„BABEŞ-BOLYAI" UNIVERSITY, CLUJ-NAPOCA

Faculty of Environmental Science and Engineering

# Heavy metal soil contamination and stress induced vegetation in Roşia Montană area

# ABSTRACT

PhD supervisor:

Prof. univ. dr. Laurenţiu-Călin Baciu

PhD student:

Adina-Laura Lazăr

CLUJ-NAPOCA - 2013

UNIUNEA EUROPEANĂ

GUVERNUL ROMÂNIEI
MINISTERUL MUNCII, FAMILIEI, PROTECȚIEI SOCIALE ȘI PERSOANELOR VÂRSTNICE
AMPOSDRU

Fondul Social European
POSDRU 2007-2013

Instrumente Structurale
2007-2013

MINISTERUL EDUCAȚIEI NAȚIONALE
OIPOSDRU

UNIVERSITATEA TEHNICA "GHEORGHE ASACHI" DIN IASI

Teza de doctorat a fost realizată cu sprijinul financiar al proiectului **"STUDII DOCTORALE PENTRU PERFORMANŢE EUROPENE ÎN CERCETARE ŞI INOVARE (CUANTUMDOC)"** POSDRU/107/1.5/S/79407.

Proiectul **"STUDII DOCTORALE PENTRU PERFORMANŢE EUROPENE ÎN CERCETARE ŞI INOVARE (CUANTUMDOC)"** POSDRU/107/1.5/S/79407, este un proiect strategic care are ca obiectiv general „*Aplicarea de strategii manageriale, de cercetare şi didactice destinate îmbunătăţirii formării iniţiale a viitorilor cercetători prin programul de studii universitare de doctorat, conform procesului de la Bologna, prin dezvoltarea unor competenţe specifice cercetării ştiinţifice, dar şi a unor competenţe generale: managementul cercetării, competenţe lingvistice şi de comunicare, abilităţi de documentare, redactare, publicare şi comunicare ştiinţifică, utilizarea mijloacelor moderne oferite de TIC, spiritul antreprenorial de transfer al rezultatelor cercetării. Dezvoltarea capitalului uman pentru cercetare şi inovare va contribui pe termen lung la formarea doctoranzilor la nivel european cu preocupări interdisciplinare. Sprijinul financiar oferit doctoranzilor va asigura participarea la programe doctorale în ţara şi la stagii de cercetare în centre de cercetare sau universităţi din UE. Misiunea proiectului este formarea unui tânăr cercetator adaptat economiei de piaţă şi noilor tehnologii, având cunoştinţe teoretice, practice, economice şi manageriale la nivel internaţional, ce va promova principiile dezvoltării durabile şi de protecţie a mediului înconjurător*."

Proiect finanţat în perioada 2010 - 2013

Finanţare proiect: **16.810.100,00** RON

Beneficiar: Universitatea Tehnică "Gheorghe Asachi" din Iaşi

Partener: Universitatea „Babeş Bolyai" din Cluj-Napoca

Director proiect: Prof. univ. dr. ing. Mihai BUDESCU

Responsabil proiect partener: Prof. univ. dr. ing. Alexandru OZUNU

# Table of contents

Key words: heavy metals, pollution, soil, bioaccumulation, impact on vegetation, vegetation stress, chlorophyll fluorescence, chlorophyll pigments, vegetation indices, spectral responses, wavelength,

# Introduction

Rosia Montana is one of the most significant gold deposits in Europe with a long mining history, of about 2000 years. Currently known reserves are equivalent to over 300 tons of gold and 1500 tons of silver. Due to economic problems, however, all mining activities ceased in 2006, lagging behind environmental problems with which Rosia Montana is now facing.

Environmental damage is significant: 140 km of underground galleries, two quarries and several waste rocks, two large tailings management facilities, which led to change in landscape. However, the main source of environmental pollution is acidic water resulting from exposing the sulphides to the existing atmospheric conditions. This process leads to the formation of sulfuric acid which dissolves heavy metals out of the rock. Heavy metals are easily transported in groundwater and surface water in the area, particularly affecting the aquatic fauna and sediments along the watercourses. Soil contamination with heavy metals is equally possible, but varies depending on the distance to the mining operations.

# Basis

Currently, there is a unique opportunity for performing environmental research in the area, as the environmental components have reached a relatively steady state since the mining activities have ceased in 2006. There are two alternatives for the area:

- the reopening of the mine at a much bigger scale than the previous operation, with the related effects on the environment
- the decision to prevent the restart of mining in the near future. In this case, the evolution of the environment will be influenced by the resilience natural systems, and by the remediation works that will be implemented.

In either case, a description of the environmental development in the former mining area is welcome in order to update existing knowledge.

The research is based on environmental monitoring of the Rosia Montana mining area, with an emphasis on heavy metal soil contamination and vegetation stress assessment using new ground spectrophotometric techniques compared to conventional laboratory techniques.

Although not commonly used, the plants response to the pollution stress is considered a relevant indicator of the state of the environment in the mining/industrial areas.

Our study contributes to the definition of the environmental baseline in the mining area, complements the existent environmental quality assessment study in the Rosia Montana perimeter. Soil, water, vegetation in Rosia Montana are environmental components that could be affected in the future by new mining operations, and in addition to the soil and water quality study, vegetation is a factor that deserves a more detailed approach, with the ability to record subtle changes in the environmental conditions and to provide data and new interpretations on the environmental dynamics. We believe that from this point of view, the proposed approach of this PhD thesis is innovative and represents a support for further development. Induced stress on vegetation was studied through a combination of destructive and non-destructive measurements.

# The objectives and issues addressed in the research

- Characterization of soil and associated materials. For this purpose, soil, sediment, open pit detritus material and vegetation samples were acquired, over an area large enough to include the entire mining area and adjacent areas unaffected by former mining activities; analysis of samples.
- Identification of areas with a low pH, potentially contaminated with heavy metals.
- Observation of induced stress on vegetation.

- Correlation of soil characteristics (pH, heavy metal content) with vegetation characteristics (pigment concentration, chlorophyll fluorescence, and heavy metals content) in order to identify possible contamination of vegetation.
- Usage of field spectrophotometric techniques (non destructive methods) to study vegetation stress. Comparison of efficient vegetation indices which are able to adapt to stress conditions identifications requirements.
- Correlation of the vegetation indices calculated based on vegetation spectral responses with chlorophyll concentration obtained by laboratory destructive methods.

## Questions arise

- To what extent is the vegetation in the Rosia Montana mining perimeter affected, given the increased soil acidity and the degree of heavy metal pollution? Is the distance to the deposit a factor that should be considered in terms of heavy metal contamination of the soil and vegetation?
- Could the determination of chlorophyll concentration in vegetation samples be considered a good indicator of stress on the existing vegetation in the mining area?
- To what extent can the vegetation stress be detected by using chlorophyll fluorescence technique?
- Is the concentration of chlorophyll in trees growing near the mining area affected by the existence of heavy metals in the soil?
- Is the concentrations of chlorophyll different depending on the pH of the soil?
- Is there a correlation between spectral response and chlorophyll concentration?
- Can areas affected and unaffected by former mining activities be differentiated?

# Chapter 1 – Physiological response of plants to stress

This chapter systematically presents theoretical concepts of how plants react to stress factors such as high concentrations of heavy metals in soil and increased soil acidity. The chapter also includes a brief description of the environmental behavior of the six studied metals.

Chlorophyll pigments are very important in the process of photosynthesis and carotenoids accompany chlorophyll pigments in assimilating plant cells and have a photoprotective role. Chlorophyll concentration is closely related to stress factors, fact shown by many researchers over time ( Merzlyak and Gitelson , 1995, Peñuelas and Filella , 1998). In most cases, any given stress factor of a specific intensity can lead to a decrease in chlorophyll concentration in a plant.

Photosynthesis is the physiological process of green higher plants through which they produce substances necessary in growth and development of mineral compounds. Photosynthesis is a process subject to stressors of any kind. Decrease in chlorophyll pigments can result in photosynthesis reduction. Chlorophyll fluorescence is a rapid technique to investigate the photosynthetic efficiency and can therefore be used as an indicator of the vegetation health ( Lichtenthaler et al. , 1999 , Li et al. , 2006) .

Heavy metals are defined as elements with metallic properties and atomic number higher than 20, which exist naturally in the earth's crust but are also found due to artificial sources . Some metals are micronutrients needed for the development of plants (eg, Zn, Cu, Mn , Ni and Co), while other metals are not involved in physiological processes of plants (eg, Cd , Pb, Hg). The most common metal contaminants are Cd, Cr, Cu, Hg , Pb , Zn .

While most plants are affected by high concentrations of heavy metals in soil, some species can tolerate high concentrations. A linear relationship between the total soil concentration and plant tissue concentrations is rare. Most plants operate as accumulators at a low background level in the soil. Often, even if the concentration of heavy metals in the soil is constant, the concentration of metals in the plants may be reduced or increased. This is strictly linked to the level of acidity of the soil, the pH influencing the mobility of the metals in the soil.

# Chapter 2 - General concepts of remote sensing and field spectrometry

This chapter presents some theoretical notions on field spectrometry technique and how this method is used to measure the spectral response of plants. This chapter also presents spectral characteristics of detecting vegetation stress and the term of vegetation indices, used in estimating some biophysical parameters.

Multispectral and hyperspectral sensors are instruments that record the electromagnetic energy emitted or reflected by different materials in several wavelength intervals called spectral bands or spectral domains ( ultraviolet , visible , near and far infrared , thermal infrared ) . Multi- and hyper- spectral sensors are also incorporated in field or laboratory spectrophotometers . Data obtained with these multi- and hyperspectral sensors are called spectral responses, representing unique information about those studied objects or phenomena, given the fact that each object has its own spectral response.

Leaves represent the main area of a tree crown where energy exchange processes take place; their optical properties are essential in understanding the transport of photons inside a leaf ( Despan and Jacquemoud , 2004).

There are three major spectral domains used for the study of vegetation: the visible domain (VIS : 400-700 nm) , red edge inflection point, the near infrared (NIR : 700-1300 nm) , the short wave IR (1300 - 2500 nm ) .

Vegetation stress is usually caused by biotic and abiotic factors . The subject of this study is abiotic factors such as soil pollution with heavy metals or soil acidity in a former mining area. Usually, the first signs of stress occur when the process of photosynthesis and metabolism decreases. Also, there are changes in the content of leaf pigment, water retention capacity of the plant and synthesis of secondary metabolites. Pollution leads to severe damage, according to the tolerance threshold of the targeted species (Panigada et al., 2010), and the effects can be visible (leaves becoming yellow, the appearance of black spots on the surface of leaves , root inhibiting , size reducing of the leaves and crown etc.) or apparent (effects take place only within the internal structure of the plant, without manifesting on the outside) .

Stressed vegetation will have lower chlorophyll content than healthy vegetation, so it will produce a spectral signature different from that of healthy vegetation. Spectral responses of healthy and stressed vegetation can be distinguished by using Red, Green and near Infrared bands but also by measuring the red edge line, located between Red and near Infrared bands (Figure 1).

Vegetation indices are derived from the combination of spectral characteristics and are generally presented in the form of dimensionless ratios (Delalieux et al., 2009). According to Huete and Jackson (1988), Qi et al. (1995) vegetation indices are mathematical transformations of spectral reflectance in order to improve vegetation signal (spectral response). They are very useful in processing and analysis of the spectral data and may be easily used in predicting the parameters of interest, without the need for traditional surveying methods

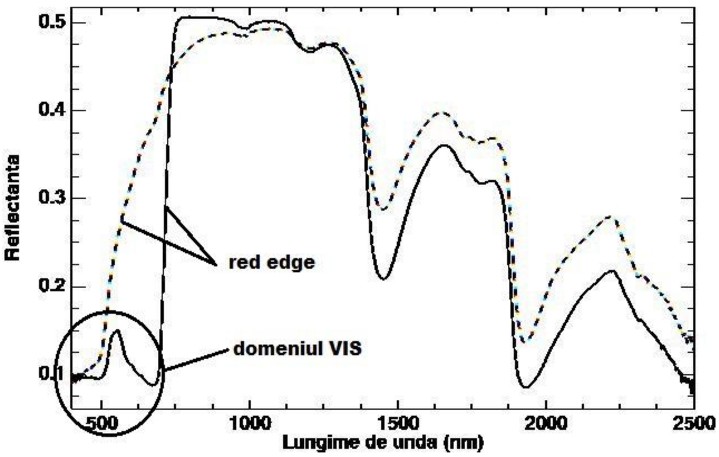



Figure 1. Healthy vegetation (solid line) and strongly affected vegetation (dotted line)

# Chapter 3 – Description of the Rosia Montana mining area

This chapter provides a brief description of the study area, including geographical location, history of mining in the study area, geology of the area, existing soil types, hydrography, climate and vegetation of the study area. The chapter also includes a brief description of the impact of former mining activities on the environment, the principle of acid water formation and a review of existing studies on environmental impact assessment in Rosia Montana.

Rosia Montana gold and silver deposit is located in the Southern Apuseni Mountains, in the central-eastern part of the Metaliferi Mountains (Alba county) and belong to the 'Golden Quadrilateral' of Transylvania (Rosia Bucium metallogenetic district) (Figure 2). It is assumed that in roman times, a significant amount of gold and silver was extracted from Rosia Montana and other neighboring fields, this territory becoming an important supplier of gold and silver of the Roman Empire. After the roman period, minig at Rosia Montana suffered some fluctuations, but it reached high values in the 19th and early 20th century.

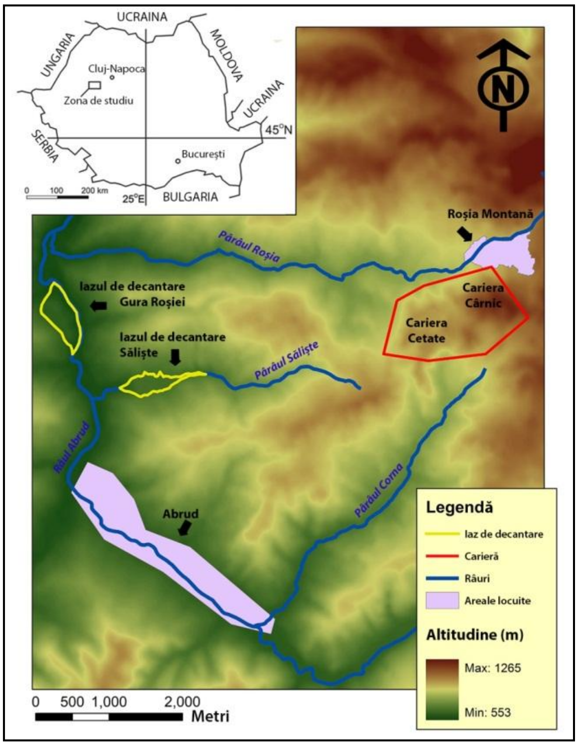


Figure 2 Rosia Montana localization

In the region, the Palaeozoic and Precambrian basement is covered by sedimentary Mesozoic deposits of marine and non-marine origin. The Cretaceous deposits, predominantly in a flysch facies, are covering most of the area. The whole pile of rocks has been intruded by Tertiary magmatites, that occur as volcanic and sub-volcanic bodies, placed along three parallel north-west trending lineaments. Three distinct magmatic episodes have been recognized during the Neogene (**Tămas, 2007**). The first stage has produced andesitic, rhyolitic, and rhyodacitic isolated bodies, lower Badenian in age (around 15 Ma). The second episode is represented by andesites and dacites of different types, that have the largest spatial extension, and were formed in the interval late Badenian to Pannonian (13.5 to 9, or even 7 Ma). The latest stage has emplaced andesites and basalts, late Pannonian to early Quaternary in age. The gold-bearing volcanites from Roșia Montană belong to the second magmatic cycle, and are part of the northernmost lineament. The Roșia Montană volcanogenic complex has been interpreted as a maar-diatreme structure, intersecting Cretaceous sediments, predominantly black shales, with sandstone and conglomerate intercalations. It includes different types of breccias and volcaniclastics, generated during successive eruptions of the volcano. Among these, phreatomagmatic breccias formed as a result of the interaction between magma and groundwater, are well represented. The breccias include fragments of metamorphic rocks from the basement, Cretaceous sedimentary rocks, and earlier dacites. Two main dacitic intrusions, with some petrographic variations, locally referred to as Cetate Dacite and Carnic Dacite, along with some smaller intrusions and dykes that are intersecting the breccias, have played an essential role in the mineralisation process. The dacite, locally affected by hydrothermal alterations, is the main host of the Au-Ag mineralisation.

At Rosia Montana, there were identified 8 main soil units and 19 units that associate different types and subtypes of soils ; soils are predominantly acidic.

Permanent water courses in the area, are Rosia , Corna and Sălişte and semi-permanent streams , of which there are some that manifest only during periods of heavy rainfall or snowmelt periods . Given the geological substrate shown above, Rosia Montana is poor in groundwater, being characterised mainly by shallow groundwater horizonts . The lakes in Rosia Montana are artificial lakes, used in the past to separate gold from remaining ore. There are five significant lakes: Taul Mare, Tarina , Brazi , Anghel , Corna , being located at heights up to 1000 m ( Taul Mare).

The climate of the study area is humid continental, with annual average temperatures calculated according to seasons and the predominant vegetation is represented by deciduous forests.

Past mining activities in Rosia Montana are divided into four different mining areas : Cetate , Carnic , Orlea and Jig- Văidoaia . All four areas have been mined underground and only two of them (Cetate si Carnic) were exploited as open pits. Resulting mining waste are: waste rock obtained after processing the ore, it has no industrial value; tailing material resulting from the technological ore preparation processing (stored in Sălişte and Gura Rosiei tailing ponds) and acidic waters that represent the main impact factor. Acid drainage is one of the most important environmental problems in the mining industry (Akcil and Koldas, 2006); acid waters are formed when minerals containing sulphides are exposed to oxidising conditions (precipitation, oxygen etc). There are several types of mineral sulphides, but generally acid water forms in rocks containing pirite iron sulphides. Acid mine waters are characterized by a very strong acidity and a very high concentration of heavy metals (Cu, Fe, Cd, Zn, Pb, etc.), but they all depend on the type and the amount of oxidized sulphides. The presence of heavy metals in acid waters is due to the oxidation of sulphides and dissolution of minerals.

Taking into account the main sources of environmental pollution in the Rosia Montana area, several environmental components are affected in a direct or indirect way: surface water (Roşia stream, Corna stream, Abrud River - which collects the waters of the two streams), groundwater , soil and landscape, in general. So far, several studies on the environmental quality of the Rosia Montana area have been made, but only few attempts of rehabilitation were accomplished. Among the researchers that have conducted measurements in and around the mining area are Florea et al. (2005), Lăcătuşu et al. (2007 ), Bird et al. (2005 ) or I.NC.D.P.A.P.M. Bucharest, 2006.

# Chapter 4 – Research methodology

Multiple field campaigns have been performed in 2011, 2012 and 2013. For this project, a number of 600 soil, sediment, detritus material samples were collected along with 144 vegetation samples ( Betula pendula and Carpinus betulus). The collection area covered the whole mining area and adjacent areas. A selection of 262 representative samples have been analysed in the laboratory for pH and heavy metals contents (Cd, Cr, Cu, Ni, Pb, and Zn). On 20 the samples that were collected from the open pit area, only the pH has been measured.

All of the 144 leaf samples were first measured for reflectance and chlorophyll content directly in the field. Measurements of chlorophyll fluorescence were made before transporting the samples to the lab, immediately after leaving the area, on the same set of samples on which measurements of reflectance and chlorophyll were made. Of the 144 samples of vegetation , 84 birch and 28 hornbeam coincided with the 76 soil samples. For chemical analysis, the determined parameters were pigments concentrations and heavy metal content assimilating pigments concentration and heavy metal content were the studied parameters. Besides the 144 vegetation samples measured in the field more spectra were gathered at the end.

Hyperspectral ( reflectance ) measurements were performed using a PSR -3500 portable spectrophotometer ( Spectral Evolution, USA ) with a full spectral domain (UV -VIS -NIR ) ranged between 350-2500 nm. The process consisted in scanning the leaves and obtaining spectral responses for each leaf. Total chlorophyll was determined initially by non destructive method using an Opti Science CCM 200 chlorophylmeter, which calculates the chlorophyll content index (CCI). For the Chlorophyll fluorescence measurements we used a portable OPTI SCIENCES - OS1 -FL fluorometer, and the analyzed parameters were Fv / Fm (maximal quantum yield of PS II ) , F0 ( basal fluorescence (initial ) ) , Fm ( maximum fluorescence ) Fv ( variable fluorescence ) . Determination of assimilating concentrations pigments ( clf a, b and carotenoids CLF ) was performed using Metertek SP -850 spectrophotometer , and the heavy metal content of the leaves was measured using the inductively coupled plasma mass spectrometry technique ( Perkin Elmer Sciex Elan DRC II ( Canada ) .

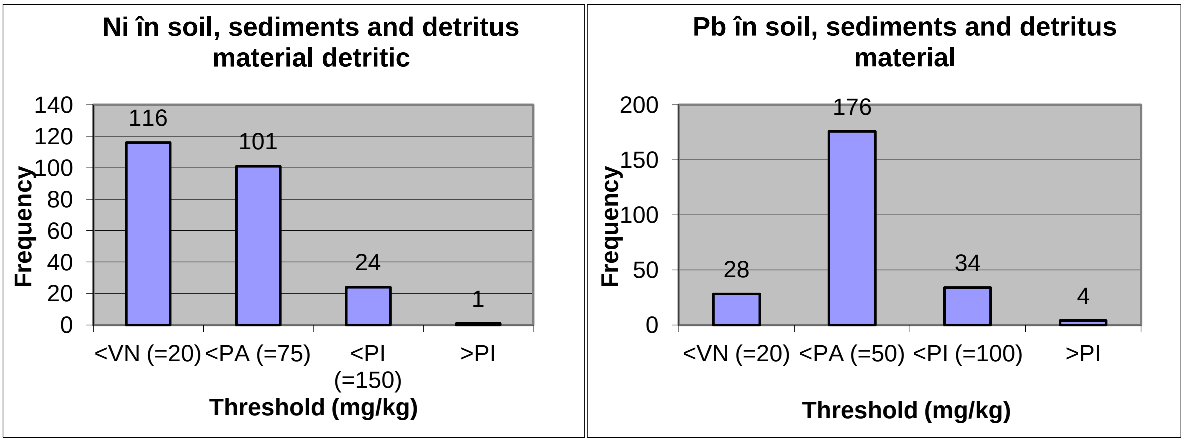
The content of heavy metals in soil, sediment, and detritus material was determined using a ZEEnit 700 atomic absorption spectrometer , and pH measurements have been achieved using a portable multimeter WTW Multi 350i (Germany).

For interpreting the results, all the existing database was subjected to analysis of statistical terms. The data were tested for normality by using the Shapiro-Wilk test, and described using mean and standard deviations. The level of significance was set at 0.05. Kruskal-Wallis test for $n \geq 3$, and Mann-Whitney nonparametric test for $n = 2$, were used to describe the not normally distributed data. To summarize the strength of the relationship between two variables, Pearson test was also applied.Results were described using the arithmetic mean, median, minimum and maximum values , followed by the standard deviation and upper and lower quartiles.

# Chapter 5 – Results and discussions

**Soil, sediment, detritus pit material results**

Further on, the heavy metals contents in soils and stream sediments have been compared with the standard reference values of RMO 756/1997. This regulation is setting up the values considered as normal contents of these chemicals in soils, and values representing alert, and response thresholds for sensitive soil use. Accordingly, four classes of values for each metal were considered: below the normal value, between the normal value and the alert threshold, between the alert threshold and the intervention threshold, and above the intervention threshold. (Figure 3).



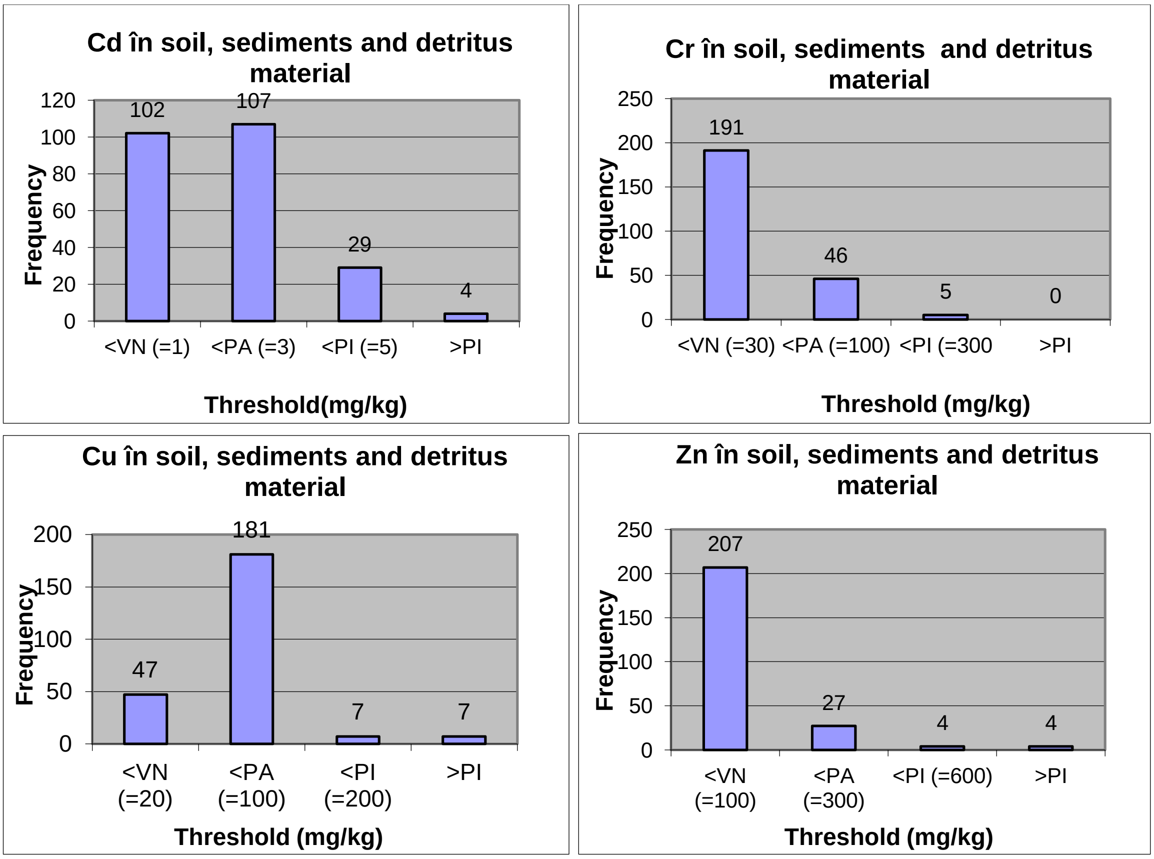



Figure 3. Classification of soil, sediment, and detritus samples according to standard reference values required by RMO 756/1997. VN - normal, PA - alert threshold, PI = intervention threshold

After the statistical analysis and comparison with the reference values, it is found that all the metals showed a very large deviation from the normal distribution profile, most of the returned analytical results are in the range of the normal values, with some variations. Copper, Pb, Cd, and Zn concentrations are in the range of normal values, slightly increased in the case of Cu, Pb, and Cd, but still below the alert threshold. Increased values can be observed in the proximity of the mining areas and along the streams with low pH waters. From the results, it appears that Ni and Cr don't have a direct link to mineralization, but rather with Cretaceous sediments appearing in the west and south of the deposit. Near the open pit Ni and Cr concentrations are quite low, which can be observed in Sălişte tailings pond where technological processed material comes from the open pit. Ni and Cr concentrations increase with distance from the deposit (Lazar et al., 2013).

According to RMO 278/2011 of the Romanian legislation, most of the soil, sediment, and detritus pit material samples were classified as acidic soils, as expected, according to the literature. The results were compared with reference values established in RMO 278/2011, which divided the obtained values in five classes of pH : strongly acidic (pH <5.00), moderately acidic (pH = 5.01 - 5.80), weakly acidic (pH = 5.81 - 6.80), neutral (pH = 6.81 - 7.20) and alkaline (pH = 7.21 - 7.67). pH values classification and their distribution in space are shown in Figure 4. Very few samples were recorded with neutral pH values (n = 4) or slightly alkaline (n = 5). The lowest pH values corresponding to strongly acidic soils can be observed in Cetate and Cârnic open pits, Sălişte TMF and also along Rosia valley, heavily polluted by water leaking mines upstream. Strongly acidic soils can be encountered on Orlea and Jig-Văidoaia massifs, small mineralized areas that have been exploited in the past.

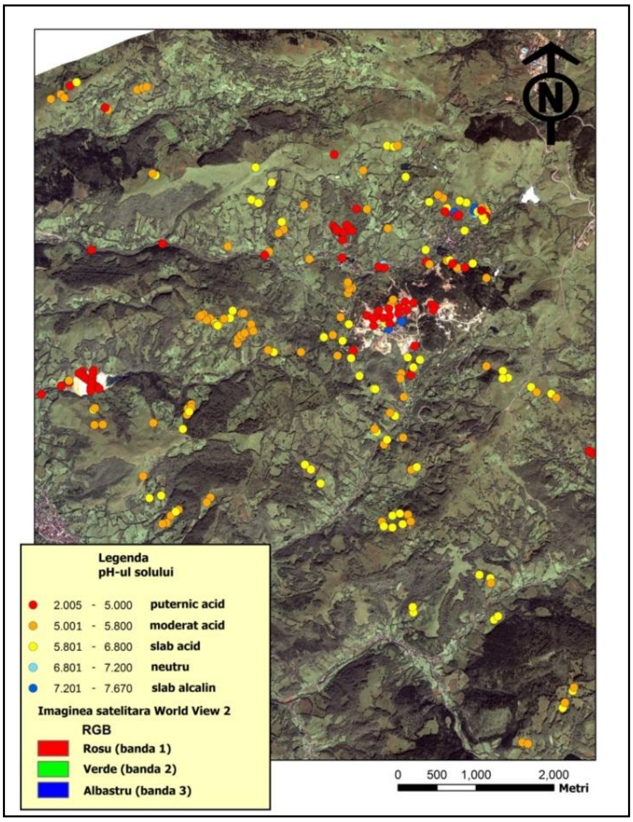



Figure 4 Spatial distribution of soil sampling points and acidity in the study area

Simple correlation coefficients between pH and heavy metal content as well as metal-metal interactions, have been determined on the entire set of 242 samples to highlight the intensity of the dependency relationship between the studied variables or the inexistent dependency relationship between the studied variables. The significance of the simple correlation coefficients values was compared with the calculated value of 0.1800. From the results, the best correlation are in the case of zinc, with geochemical chalcophile affinity. The best correlations are formed with Pb (r = 0.458), Cd (r = 0.376) and with Cu (r = 0.518), while the concentrations of Ni, Cd and Cr in soil and sediment have only one correlation with the concentration of other metals (Lazar et al., 2013). These metals coexist near and inside the mining deposit. Also copper and lead are strongly correlated (r = 0.370), elements with geochemical chalcophile affinity, which generally have a good association.

## Vegetation results

Several studies have investigated the impact of heavy metals on *Betula pendula*, but less on *Carpinus betulus*. From several authors studies, including Ehlin (1982), Wislocka et al. (2006), Dmuchowski et al. (2011) some values for metal concentrations accumulated in the trunk, bark or leaves were selected. These values were recorded in the areas affected by heavy metal pollution (industrial areas, mining areas, etc.) and also in control areas (if applicable). In addition to the references presented above, normal (sufficient), excessive and toxic values given by *Kabata-Pendias & Pendias (1992)* and *Pais & Jones (1997)* have been used for comparing the results of the present study.

The results showed that in the case of the two studied species (*Betula pendula* and *Carpinus betulus*), most of the obtained concentrations were within normal limits reported in plants. The exception is for zinc and to a lesser extent for nickel.

In about 30 samples of leaves, Zn concentrations in *Betula pendula* exceed the normal values reported by Kabata Pendias and Pendias (1992), this phenomenon being reported in only one case of hornbeam. These values fall within the excessive concentrations of plant tissue. In general, Zn concentrations in *Betula pendula* leaves exceed Zn concentrations in corresponding soil samples. Table 1 presents the average metals concentrations in leaves of Betula sp. and Carpinus sp. Although *Carpinus betulus* is in the same family, *Betulaceae*, it is not as susceptible to Zn accumulation as birch, but rather the concentration of Zn in hornbeam leaves is usually lower than in the soil.

The hyper-accumulation of Zn in different species of Betula has been described in previous papers. *Betula papyrifera, B. pendula, and B. pubescens* are known to be tolerant, and have the ability to accumulate metals, such as Zn and Pb (Gussarsson, 1996; Prasad, 1999; Margui et al., 2007; Gallagher, 2008).

Table 1. Mean concentrations of heavy metals in leaves of *B. pendula* and *C. betulus* in Roșia Montană area depending on mean concentrations of heavy metals in soil. ( standard deviation ).

| Species | Ni (mg/kg) | Pb (mg/kg) | Cd (mg/kg) | Cr (mg/kg) | Cu (mg/kg) | Zn (mg/kg) |
|---|---|---|---|---|---|---|
| *B. pendula* | 3.97 (2.70) | 0.34 (0.21) | 0.34 (0.20) | 1.24 (0.60) | 5.49 (1.60) | 110.31 (84.72) |
| *Soil* | 14.45 (17.05) | 39.44 (75.37) | 1.62 (1.15) | 9.36 (9.18) | 57.01 (91.70) | 70.43 (93.37) |
| *C. betulus* | 5.14 (5.15) | 0.42 (0.22) | 0.08 (0.08) | 1.26 (0.60) | 6.43 (2.39) | 25.81 (26.66) |
| *Soil* | 27.69 (31.49) | 50.67 (112.14) | 1.99 (1.29) | 14.57 (14.06) | 77.40 (126.15) | 103.67 (138.30) |

In order to verify to what extent the heavy metals concentration in vegetation samples depend on the soil characteristics - pH and heavy metals - multiple correlation coefficients are calculated, starting with the simple correlation coefficients. The following simple correlation coefficients were considered: $r_{f \cdot s} = r_{s \cdot f}$ - simple correlation between heavy metal content in leaves and heavy metal content in soil; $r_{f \cdot pH} = r_{pH \cdot f}$ - simple correlation coefficient between the heavy metal content in leaves and soil pH; $r_{s \cdot pH} = r_{pH \cdot s}$ - simple correlation between heavy metal content in soil and soil pH. Simple correlation coefficients significance test was performed using $\boldsymbol{r'_{\alpha v}}$ criterial value (calculated according to the sample size). The significance level ($\alpha$) chosen has a value of 0.05, and the number of degrees of freedom ($v$) is calculated as follows $v = 47\text{-}2 = 45$, or $v = 26\text{-}2 = 24$. For birch $\boldsymbol{r'_{\alpha v}}$ value was set at 0.2818, and for hornbeam $\boldsymbol{r'_{\alpha v}}$ value is 0.3746. To test the significance of multiple correlation coefficient value for metals Fischer criterion (Fc) was applied. The size obtained is compared to the table value of Fischer criterion (Ft). For birch, $F_T = 3.214$ and for hornbeam, $F_T = 3.42$. Table 2 summarizes all the values of simple and multiple correlation coefficients, together with criteria values $r'_{\alpha v}$ or $F_T$. The calculations were made for the two species separately and for each metal individually.

Table 2. Simple and multiple correlation coefficients compared to Fisher tabular values. Values in bold are above the criterial $r'_{\alpha v}$ or $F_T$ limits, meaning the linear correlation is good and significant. (**Lazăr et al., 2013**)

| **No.** | **Species** | **Heavy metals** | **r f·s** | **r f·pH** | **r s·pH** | $r'_{\alpha v}$ | **r f·s, pH** | **$F_C$** | **$F_T$** | **Conclusions** |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | *B. pendula* | Ni | **0.642** | -0.084 | 0.119 | 0.2818 | **0.662** | **17.163** | 3.214 | ***Correlation*** |
| 2 | *B. pendula* | Pb | **0.372** | 0.044 | -0.113 | | **0.382** | **3.759** | | ***Correlation*** |
| 3 | *B. pendula* | Cd | 0.260 | 0.233 | 0.065 | | 0.338 | 2.838 | | *Non-correlation* |
| 4 | *B. pendula* | Cr | -0.204 | -0.120 | 0.082 | | 0.229 | 1.218 | | *Non-correlation* |
| 5 | *B. pendula* | Cu | 0.024 | -0.097 | 0.016 | | 0.100 | 0.222 | | *Non-correlation* |
| 6 | *B. pendula* | Zn | **0.462** | -0.274 | -0.070 | | **0.522** | **8.24** | | ***Correlation*** |
| 7 | *C. betulus* | Ni | **0.598** | -0.070 | 0.088 | 0.3746 | **0.611** | **6.851** | 3.42 | ***Correlation*** |
| 8 | *C. betulus* | Pb | **0.705** | -0.296 | -0.252 | | **0.716** | **12.097** | | ***Correlation*** |
| 9 | *C. betulus* | Cd | 0.042 | 0.006 | 0.098 | | 0.043 | 0.021 | | *Non-correlation* |
| 10 | *C. betulus* | Cr | 0.061 | -0.193 | -0.069 | | 0.199 | 0.474 | | *Non-correlation* |
| 11 | *C. betulus* | Cu | **0.530** | -0.239 | 0.152 | | **0.621** | **7.219** | | ***Correlation*** |
| 12 | *C. betulus* | Zn | 0.291 | -0.002 | -0.194 | | 0.296 | 1.104 | | *Non-correlation* |

Analyzing multiple correlation coefficient values, it can be concluded that for birch leaves, nickel, lead and zinc concentrations depend mostly on soil characteristics and for hornbeam, Ni, Pb and Cu concentrations depend on soil heavy metal concentrations and soil acidity.

An interesting thing to note and to consider is the fact that lead located in soil (toxic element to plant metabolism) is closely related to Pb concentrations found in birch and hornbeam leaves. A significant correlation for birch, between Pb concentrations in soil and Pb leaf concentrations is also reported by Gallagher (2008) demonstrating the ability of birch to carry Pb in the leaves.

In this study we wanted to see whether a very acid soil pH can influence the chlorophyll concentration in vegetation samples of both species separately. Also we wanted to see whether chlorophyll concentration can be influenced by increasing the distance from the deposit and increasing heavy metal content in soil. It was found that for birch, chlorophyll a, chlorophyll b and carotenoids show a slight concentration decrease, in terms of increased acidity, and average values of total chlorophyll concentration recorded by soil pH are very close to one another (Figure 5). The difference in concentrations between the three classes of pH is statistically insignificant. An exception is hornbeam because the total chlorophyll concentration is greater for trees that grow in moderately acidic soils compared to those that grow in slightly acidic soils. However, chlorophyll a (primary photosynthetic pigment) is more abundant in leaves collected from *Carpinus betulus* trees that grow in slightly acidic soils. In this case, it is found that the optimal chlorophyll a and chlorophyll b ratio (CHL / CHL b) is situated in the pH range 5-5.80 for both species.

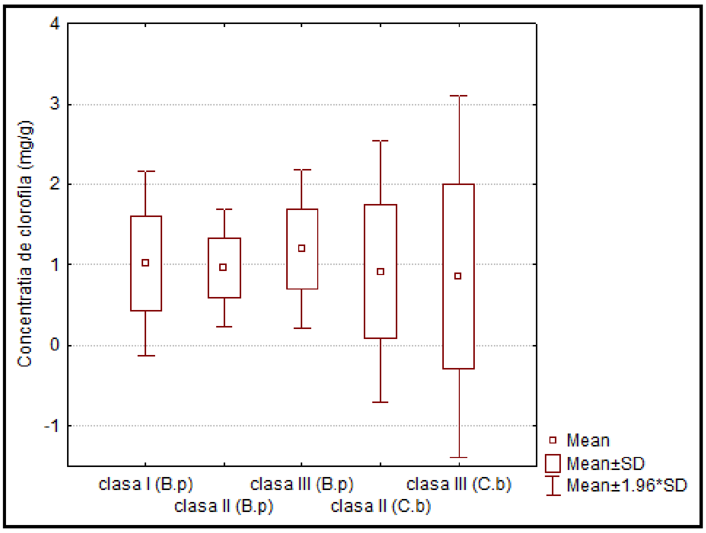



**Fig. 5** Box plots for mean total chlorophyll concentration in *B. pendula* and *C. betulus*, in relation with the pH classes (I – strongly acidic; II – moderately acidic; III – weakly acidic).

Regarding the distance from the deposit, an increase of chlorophyll concentration is visible far from the mining deposit, however, carotenoids concentration remains constant in both locations. Near the deposit, chlorophyll a, chlorophyll b and total chlorophyll decreases, even though the content of heavy metals in soil does not exceed normal limits.

There are many other disturbing factors when speaking of vegetation stress. The object of this study, however, is the potential impact of former mining activities on vegetation and soil, and this study shows that mining perturbing factors, as increased soil acidity, high heavy metals content in soil, are not a major negative impact.

The chlorophyll concentrations and chlorophyll fluorescence parameters were analyzed according to soil pH and depending on the species. For the two species found in different pH classes, no significant changes in chlorophyll fluorescence parameters measured on intact and fully developed leaves were detected. Fv / Fm and (F0) recorded normal values for all pH classes, in both species, with the exception of basal fluorescence for hornbeam with higher significant values, for the pH range of 5.80-6.80. Some lower values of Fv / Fm ratio were observed for birch and hornbeam, but in areas with moderately and weakly acidic pH. These values, however, are extremely rare (ten birch trees and five hornbeam trees with potential quantum yield values below 0.70).

## Spectra processing

The spectral responses of both species are shown in Figure 6.

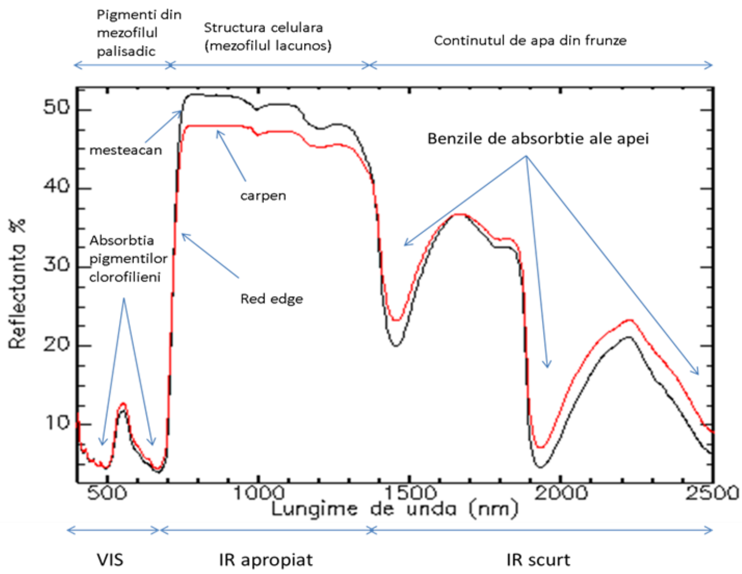



Figure 6. Spectral responses for *Betula pendula* and *Carpinus betulus*

Both spectra have similar shape and same adsorption bands. The reason for this similar shape is the fact that both species belong to the same family. The highest spectral response is occurring in the NIR domain (700-1300 nm) because of multiple scattering at the air cell interfaces in the leaf internal tissue. Vegetation has low reflectance in the visible spectral domain (400-700 nm) since the majority of solar energy is absorbed by photo-active pigments.

Applying nonparametric t test (Mann Whitney test) for the spectra of the two species, it was found that although there are some visual differences between the two spectra, the differences are not statistically significant, but the p value is very close to 0.05 ($p = 0.0655$).

For the spectra classification according to soil pH, spectral libraries were created and reflectance values were saved in ASCII format to calculate vegetation indices.

All the leaf samples were averaged in order to obtain only one spectral curve for each tree. The obtained means were grouped according to soil pH and according to species. For each pH class, a spectral library was created with spectral responses of all the trees that are entering one specific class.

The difference in spectral response depending on soil pH classes was calculated by dividing the trees spectral responses in six spectral domains: blue (400-495 nm), green (500-600 nm), red + red edge (620-750 nm), NIR (760 - 1300 nm), SWIR - 1 (1300 -1800 nm) and SWIR - 2 (1800 - 2500 nm). This division into separate areas proved to be useful in observing subtle differences in spectral responses between pH classes and between the two species.

Nonparametric test results applied on the spectral values of the 6 different areas, showed generally insignificant changes in spectral responses according to the soil pH classes. Significant differences occur only in case of birch trees, in the near infrared, where there is a slight increase in reflectance with decreasing soil acidity.

In order to compare the spectral responses according to the distance to the deposit, averaged spectral responses were calculated for trees near the deposit and for trees selected at a distance of about 2 kilometers from the mining deposit. The spectra obtained are plotted in Figure 7. Nonparametric tests have determined that between the two spectra there are small differences, mainly in the visible and near IR ($p = 0.0302$).

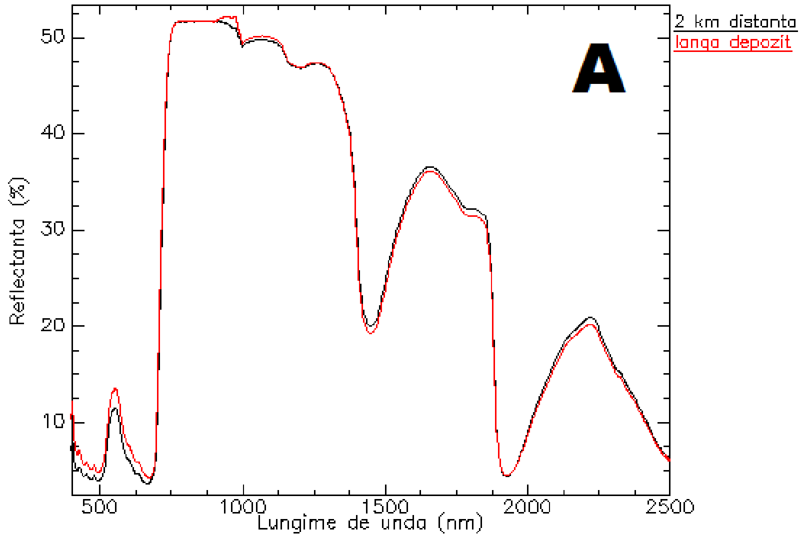



Figura 7. Graphic representation of the tree spectral means for the two locations

In order to verify which wavelength is more sensitive or is not sensitive at all to the chlorophyll concentration, coefficients of correlation between reflectance values for each wavelength and assimilating pigments, were calculated. The wavelengths of maximum sensitivity pigment content are indicated by negative strong values, as the reflectance decreases with increasing concentration of assimilating pigments at these wavelengths. In this study, the strongest correlation coefficients obtained varies between -0.400 and -0.470, for birch and between -0.250 and -0.283, for hornbeam. The values obtained for hornbeam are weaker, unable themselves to establish a better connection with field measurements.

A number of vegetation indices, very widely used in literature, are also calculated in this study, to test their sensitivity to concentrations of assimilating pigments. Most of the indices are using the NIR wavelength, in relation to green and red bands, which shows the highest sensitivity in the presence of assimilating pigments.

Links between pigments and vegetation indices are investigated using correlation and regression analysis. For the calculation of vegetation indices, each obtained spectral response curve (reflectance) was converted into a series of reflectance values. The values obtained were transferred to an Excel table where vegetation indices were calculated based on mathematical formulas for each indicator separately. The correlation coefficients between the concentration of photosynthetic pigments (Chl a, Chl b, Chl a + b and carotenoids) and vegetation indices are presented in Table 3 for *Betula sp* and in Table 4 for *Carpinus sp*.

Table 3. Coefficients of correlation between vegetation indices and photosynthetic pigments (*Betula pendula* (N = 84)

| Vegetation indices | Chl a (mg $g^{-1}$) | Chl b (mg $g^{-1}$) | Carotenoids (mg $g^{-1}$) | Chl a + Chl b (mg $g^{-1}$) |
|---|---|---|---|---|
| R430/R680 | 0.232 | 0.160 | 0.239 | 0.229 |
| R440/R690 | 0.150 | 0.102 | 0.148 | 0.147 |
| R740/R720 | 0.353 | 0.387 | 0.364 | 0.406 |
| R750/R550 | 0.382 | 0.380 | 0.403 | 0.424 |
| R750/R710 | 0.373 | 0.392 | 0.386 | 0.422 |
| R800/R550 | 0.381 | 0.375 | 0.402 | 0.421 |
| R800/R635 | 0.466 | 0.418 | 0.483 | 0.498 |
| R800/R680 | 0.447 | 0.228 | 0.440 | 0.408 |
| R760/R695 | -0.452 | -0.400 | -0.463 | -0.482 |
| R750/R705 | 0.389 | 0.397 | 0.403 | 0.435 |
| NPCI | -0.226 | -0.165 | -0.230 | -0.227 |
| (R800-R635)/(R800+R635) | 0.453 | 0.439 | 0.475 | 0.497 |
| (R800-R680)/(R800+R680) | 0.455 | 0251 | 0.449 | 0.423 |
| REIP | 0.302 | 0.327 | 0.315 | 0.345 |
| NDLI | 0.172 | 0.029 | 0.114 | 0.133 |
| (R750-R705)/(R750+R705) | 0.377 | 0.397 | 0.392 | 0.427 |
| R800,7/R513,7 | 0.502 | 0.345 | 0.495 | 0.494 |
| (R800,7-R513,7)/( R800,7+R513,7) | 0.498 | 0.365 | 0.497 | 0.499 |

Table 4 Coefficients of correlation between vegetation indices and photosynthetic pigments *Carpinus betulus* (N = 28)

| Vegetation indices | Chl a (mg $g^{-1}$) | Chl b (mg $g^{-1}$) | Carotenoids (mg $g^{-1}$) | Chl a + Chl b (mg $g^{-1}$) |
|---|---|---|---|---|
| R430/R680 | -0.167 | -0.024 | -0.162 | -0.135 |
| R440/R690 | -0.039 | 0.069 | -0.013 | -0.008 |
| R740/R720 | 0.264 | 0.320 | 0.327 | 0.299 |
| R750/R550 | 0.216 | 0.280 | 0.271 | 0.250 |

| | | | | |
|---|---|---|---|---|
| R750/R710 | 0.240 | 0.314 | 0.299 | 0.278 |
| R800/R550 | 0.200 | 0.264 | 0.254 | 0.232 |
| R800/R635 | 0.172 | 0.272 | 0.221 | 0.213 |
| R800/R680 | -0.134 | -0.013 | -0.125 | -0.106 |
| R760/R695 | 0.106 | 0.209 | 0.149 | 0.144 |
| R750/R705 | 0.226 | 0.307 | 0.283 | 0.266 |
| NPCI | 0.179 | 0.031 | 0.175 | 0.146 |
| (R800-R635)/(R800+R635) | 0.206 | 0.284 | 0.252 | 0.243 |
| (R800-R680)/(R800+R680) | -0.163 | -0.056 | -0.161 | -0.141 |
| REIP | 0.226 | 0.289 | 0.292 | 0.260 |
| NDLI | -0.432 | -0.295 | -0.427 | -0.419 |
| (R750-R705)/(R750+R705) | 0.240 | 0.313 | 0.300 | 0.278 |

In general, correlations between vegetation indices and chlorophyll pigments are weak and very weak for *Betula pendula* and very weak in case of *Carpinus betulus.* Mathematical relationship between the ICC and chlorophyll pigments extracted varies depending on species, growing conditions, etc.

In this case, the chlorophyllmeter the measurements were performed on two species, and not on the entire set of samples, because the device was not available during the entire field period. Therefore, in the case of birch, from 84 samples were measured only 66 samples and in the case of hornbeam from 28 samples were measured only 24. The values obtained for birch are in the range 5.20 - 28.20 and hornbeam values for the range 8.30 - 25.73.

After calculating the relationship between photosynthetic pigments and chlorophyll index of both species, the results indicate that there is a weak correlation between pigment chlorophyll index determined in the laboratory. We assume that the reason for this is damaging the pigment content during handling of samples under very high temperature (over 30 ° C) and transport to the laboratory.

Correlations between spectral responses and chlorophyll index values obtained in the field gave good results. It is assumed that since the measurements of the devices were made consecutive field correlations between the two variables are stronger. The strongest correlation coefficients obtained for birch varies between -0.60 and -0.65 and the strongest correlation coefficients obtained for hornbeam varies between -0.75 and -0.82. Compared with the values of correlation coefficients between each wavelength reflectance and chlorophyll pigments determined in the laboratory, the correlations obtained in this case are significantly stronger.

In general, correlations between vegetation indices and ICC are visibly improved in case of both species, with very high correlation coefficients for most of the vegetation indices.

# Chapter 6 – Conclusions

General conclusions drawn from the study are

- Results indicate an increased acidity of soils in Rosia Montana, according to the existing acidity classes in RMO 756/1997 . This increased acidity is typical for mountain soils. A very low pH was recorded in the open pits, in the existing sediment along rivers that collect acid mine waters, but also within the tailings.
- Most of the results are within the normal range, regarding the heavy metals soil concentrations, the obtained values are compared with reference values provided by the Romanian legislation. Normal, below alert threshold values were recorded throughout the study area, even near the mining deposit.
- The highest metals concentrations generally coincide with highly acidic pH soils: in the mining deposit, lead concentration exceed the intervention threshold, zinc concentrations are high, with some values above alert threshold; along Rosia and Corna streams, sediment samples have values above the intervention threshold for cadmium, lead and copper the TMF soil samples indicate high values of lead, mostly above the alert threshold. These high concentrations from the former mining areas are correlated with the existing geological substratum.
- Ni and Cr concentrations are higher in areas distant from the mining deposit, between the two metals there is a strong dependence. They are dominant in flysch areas (Cretaceous sedimentary).
- Zinc concentration (chalcophile element) correlates best with other geochemical chalcophile affinity metal concentrations (three significant correlations with Pb, Cd and Cu).
- In general, we can confirm that soil pollution is minimum near the mining area, very few locations registering values above the alert threshold.
- In general, heavy metals concentrations in the leaves are not exceeding normal values. Exceptions occur for Ni and Zn, in both species; however, excessive Ni concentrations are connected to other geological formations than the ones associated with the ore deposit.
- *Betula pendula* can be considerate a Zn hyper-accumulative species, even when the Zn concentrations in soil are low.
- *Carpinus betulus*, although a part of the same family *Betulaceae*, does not have the same tendency to build-up zinc on the leaves, the Zn concentration in hornbeam leaves being generally lower than the ones in the soil.

- There are no strong correlations between the metal concentrations in soil and in leaves (in both species), this phenomenon is due mainly to the relatively low metal concentrations in soil, for the original measurement locations. Ni, Pb, Zn for birch and Ni, Pb, Cu in the case of hornbeam are metals with leaves concentrations depending on the soil metal concentrations and soil acidity.
- For birch, there is a slight increase in assimilating pigments concentration once the soil acidity decreases, but the difference between the three pH classes is statistically insignificant;
- Hornbeam shows a slight increase in chlorophyll concentration in moderately acidic soils (statistically insignificant), but chlorophyll a (primary photosynthetic pigment) is has higher values in leaves collected from trees growing in slightly acid soils.
- *Betula pendula* has a significant increase of the chlorophyll concentration in relation to the distance from the mining deposit. Near the deposit, assimilating pigments concentration decreases, even if the content of heavy metals in soil has values that don't exceed normal limits.
- The optimum chlorophyll a chlorophyll b ratio occurs in the pH range 5-5.80, for both species.
- In general, all correlation coefficients between the content of heavy metals in soil and chlorophyll concentration are negative and close to 0, as long as the heavy metal soil concentrations remain within the normal range, they are not a stress factor stress for the sampling areas.
- In order to identify possible failures in the leaf photosynthetic apparatus, chlorophyll fluorescence measurements suggest that leading indicators (Fv / Fm, F0 and Fm) are not significantly affected by soil acidity values in the study area.
- Using the field spectrophotometry method, spectral domains or wavelengths that showed high sensitivity to the chlorophyll pigments content in birch and hornbeam leaves were identified. The strongest correlation coefficients ranged between -0.400 and -0.470 for birch and between -0.250 and -0.283 for hornbeam, which showed a weak correlation between laboratory determined chlorophyll pigments and the spectra acquired in the field.
- Of all the indices tested for correlation between spectra and chlorophyll pigments concentration values, the fallowing indices: R800 / R635, (R800 - 635) / (R800 + R635) and indices R695/R760 R800, 7/R513, 7, (0.7 R800-R513, 7) / (R800, R513 7 +, 7) correlated best with the chlorophyll pigments concentration determined in the laboratory.

- Based on spectrometric determination, a decrease in the chlorophyll concentration inside the mining deposit, where detritus open pit material recorded very low pH (2.5), was shown.
- ICC recorded strong correlations with spectra acquired in the field. The strongest correlation coefficients obtained for birch trees vary between -0.60 and -0.65 and between -0.75 and -0.82, for hornbeam trees. In comparison with the obtained correlation coefficients values between each wavelength reflectance and laboratory determined chlorophyll pigments, the obtained correlations in this case are significantly stronger; an expected result, due to the fact that the measurements with the two devices were taken successively.
- Most tested vegetation indices showed strong correlations with ICC values.

# Selected references

Akcil, A., Koldas, S., 2006. Mine Drainage (AMD): causes, treatment and case studies, Journal of Cleaner Production 14, pp. 1139-1145

Bird, G., Brewer, P., Macklin, M., Șerban, M., Bălteanu, D., Driga, B., 2005. Heavy metal contamination in the Arieş river catchment, western Romania: Implications for development of the Roşia Montană gold deposit Journal of Geochemical Exploration, 86(1), pp. 26-48

Delalieux, S., Somers B., Verstraeten, W. W., van Aardt, J. A. N., Keulemans, W., Coppin, P., 2009. Hyperspectral indices to diagnose leaf biotic stress of apple plants,considering leaf phenology, International Journal of Remote Sensing Vol 30, nr.8, pp. 1887-1912

Despan, D., Jacquemoud, S., 2004. Optical properties of soil and leaf: Necessity and problems of modeling. In: Schönermark, M. von, Geiger, B., Röser, H. P. (Eds.): *Reflection properties of vegetation and soil.* Berlin. Wissenschaft und Technik Verlag, pp. 39-70

Dmushowski, W., Gozdowski, D., Baczewska, A.H., Bragoszewska, P., 2011. Che comparison bioindication methods in the assessment of environmental pollution with heavy metals, Ochrona Środowiska i Zasobów Naturalnych Nr. 50, 2011 r.

Ehlin, P.O., 1982. Seasonal variations in metal contents of birch, Geologiska Föreningen i Stockholm Förhandlingar, 104:1, pp. 63-67, http://dx.doi.org/10.1080/11035898209454538

Florea, R., Stoica, A., Baiulescu, G., Capota, P., 2005. Water pollution in gold mining industry: a case study in Roșia Montană district, Romania. Environmental Geology, 48, pp. 1132-1136

Gallagher, F.J., 2008. The role of soil metal contamination in the vegetative assemblage development of an urban brownfield. New Brunswick, New Jersey

Gussarsson, M., Asp, H., Adalsteinsson, S., Jensen, P., 1996. Enhancement of cadmium effect on growth and nutrient composition of birch (Betula pendula) by buthionine sulphoximine (BSO), Journal of Experimental Botany, vol. 47, No. 295, pp. 211-215

Huete, A.R., Jackson, R.D., 1988. Soil and atmosphere influences on the spectra of partial canopies. Remote Sensing of Environment, 25(1), pp. 89-105

I.NC.D.P.A.P.M. Bucureşti, 2006. *Capitolul 4.4. Solul* din *Raportul la studiul de evaluare a impactului asupra mediului* depus de RMGC

Kabata Pendias, A., Pendias, H., 1992. Trace elements in soils and plants, CRC Press: Boca Raton, Florida. 315 p

Lazăr, A.L., Baciu, C., Roba, C., Dicu, T., Pop, C., Rogozan, C., Dobrotă, C., 2013. Impact of the past mining activity in Roșia Montană (Romania) on soil and vegetation (**în curs de revizuire**)

Lăcătuşu, R., Câtu, G., Lungu, M., Rizea, N., Lazăr, R., 2007. Possible impact of mining and processing the Roşia Montană gold-bearing ore upon soil, PRESENT ENVIRONMENT AND SUSTAINABLE DEVELOPMENT, NR. 1, 2007, pp. 107-114

Li, R., Guo, P., Baum, M., Grande, S., Ceccarelli, S., 2006. Evaluation of chlorophyll content and fluorescence parameters as indicators of drought tolerance in barley. *Agricul. Sci. China*, 5, pp. 751-757

Lichtenthaler, H., Wenzel, O., Buschmann, C., Gitelson, A., 1999. Plant Stress Detection by Reflectance and Fluorescence. Annals New York Academy of Sciences, pp. 271–285

Margui, E., Queralt, I., Carvalho, M. L., Hidalgo, M., 2007. Assessment of metal availability to vegetation (Betula pendula) in Pb-Zn ore concentrate residues with different features. Environmental Pollution 145, pp. 179-84

Merzlyak, M.N., Gitelson, A.A., 1995. Why and what for the leaves are yellow in autumn? On the interpretation of optical spectra of senescing leaves (*Acer platanoides* L.). J. Plant Physiol. 145, pp. 315-320

Pais, I., Jones, J.B.Jr., 1997. The handbook of trace elements. St. Lucie Press, Boca Raton, Florida

Panigada, C., Colombo, R., Meroni, M., Rossini, M., Cogliati, S., Busetto, I., Fava, F., Picchi, V., Migliavacca, M., Marchesi, A., 2010. Remote sensing of vegetation status using hyperspectral data, Proc. 'Hyperspectral 2010 Workshop', Frascati, Italy, 17–19 March 2010 (ESA SP-683, May 2010)

Penuelas, J., Filella, I., 1998. Visible and near-infrared reflectance techniques for diagnosing plant physiological status. Trends Plant Sci. 3, pp. 151-156

Prasad, M. N. V., 1999. Metallothioneins and metal binding complexes in plants. In: Prasad M.N.V., Hagemeyer J., (Eds), Heavy Metal Stress in Plants: From Molecules to Ecosystems. Springer, Berlin, pp. 51-72

Qi, J., Moran, M.S., Cabot, F., Dedieu, G., 1995. Normalization of sun/view angle effects using spectral albedo-based vegetation indices. Remote Sensing of Environment, 52(3), pp. 207-217

Tămaş, C.G., Bailly, L., Ghergari, L., O'Connor, G., Minut, A., 2006. New occurrence of tellurides and argyrodite at Roșia Montană, Apuseni Mountains, Romania and their metallogenetic significance. Canadian Mineralogist 44 (2), pp. 689-705

Wislocka, M., Klink, A., Krawczyk, J., Morison, L., 2006. Bioaccumulation of Heavy Metals by Selected Plant Species from Uranium Mining Dumps in the Sudety Mts., Poland, Polish J. of Environ. Stud. Vol. 15, No. 5 (2006), pp. 811-818