Environmental risk assessment practices and applications for gold mines in EU

Assessing environmental risks at three mine sites: Kittilä mine in Finland and Roșia Montană and Zlatna mines in Romania

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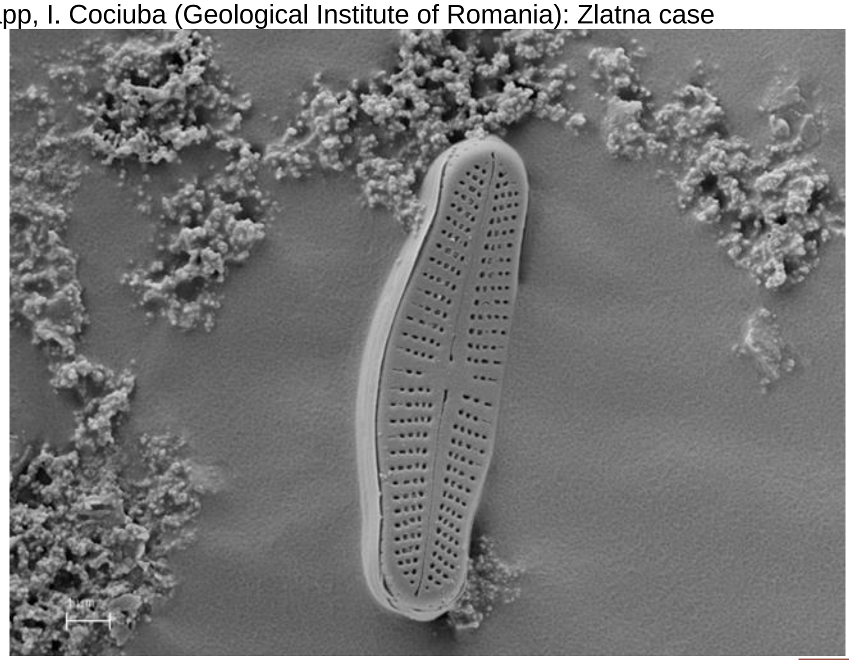

Assessing environmental risks at three mine sites: Kittilä mine in Finland and Roșia Montană and Zlatna mines in Romania

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Keywords: mining, aquatic organisms, ecological impacts, ecological risk assessment, diatom, microorganisms, invertebrates

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Abstract

Aquatic organisms represent a significant ecological indicator for assessing the physical and chemical changes and anthropogenic impact on water quality. Besides this, aquatic organisms that spend most of their life cycle in water are extremely important in maintaining the health of aquatic ecosystems and also represent a very useful instrument for rapidly assessing the contaminants risk. The aim of these risk assessment studies was to evaluate ecological and health risks related to mining, as well as to assess the usability of risk assessment tools to improve water management strategies at mine sites. This task pursues the enhancement of environmentally sustainable mining by characterizing and evaluating anthropogenic emissions relative to geogenic background, by modelling reactions and pathways of contaminants, and by assessing environmental risks. This study provides ecological perspectives from three contrasting European gold mine sites: the operating Kittilä mine in Finland, and two closed mining areas in Romania, Roșia Montană and Zlatna.

Keywords
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Synopsis

1. Purpose of research

The purpose of the environmental risk and impact research was to evaluate and test methods for environmental monitoring, modelling and risk assessment. The aim of the risk assessment studies was to evaluate ecological and health risks related to mining, as well as to assess the usability of risk assessment tools to improve water management strategies at mine sites. This task pursues the enhancement of environmentally sustainable mining by characterizing and evaluating anthropogenic emissions relative to geogenic background, by modelling reactions and pathways of contaminants, and by assessing environmental risks. This study provides ecological perspectives from three contrasting European gold mine sites: the operating Kittilä mine in Finland, and two closed mining areas in Romania, Roșia Montană and Zlatna.

The study was carried out within the SUSMIN project (Tools for sustainable gold mining in EU) in close cooperation between the Geological Survey of Romania, Babes Bolyai University and Geological Survey of Finland. This study was part of the environmental work package in which also novel monitoring tools and geochemical characteristics and isotopes for assessing migration of harmful substances were tested (Papp et al. 2018 and Lahtinen et al. 2018).

1. Material and analysis

The abandoned Roșia Montană and Zlatna sites have been mined since Roman times; they lacked environmental regulations and resulted in significant environmental consequences, still requiring monitoring and designing appropriate measures to diminish the contaminant release. In contrast, as recently opened and still operating mine, Kittilä mine has always had stricter environmental regulations and thus lesser environmental issues, but nevertheless needs monitoring. Due to different backgrounds, the case study sites had different research aims that required different ecological risk assessment methods.

In Kittilä, the aim was to assess the usability of ecological risk assessment in water management strategies, whereas in Romania biological monitoring studies on microorganisms, invertebrates, diatoms were seasonally conducted to assess the ecological status of closed mine sites and the risks of seepage water from old mining waste sites on water habitat.

2. Methods applicability

Aquatic organisms represent a significant ecological indicator for assessing the physical and chemical changes and anthropogenic impact on water quality. Besides this, aquatic organisms that spend most of their life cycle in water are extremely important in maintaining the health of aquatic ecosystems and also represent a very useful instrument for rapidly assessing the contaminants risk. The obtained data also revealed that the type of contamination (organic or inorganic) is a very important factor for the existence of certain communities. Furthermore, as the study showed that aquatic organisms indicate well both short (Kittilä site) and long-term (Romania and Zlatna sites) impacts of mining, ecological monitoring is a suitable tool to assess the environmental impact of mining on aquatic ecosystems.

3. General evaluation and awareness

As the aquatic ecosystems are controlled by various geological, physical, chemical and biological factors and identifying the exact source is not easy, a comprehensive environmental impact assessment requires further complex investigations. Moreover, as the ecological surveillance reports of Kittilä site showed, studies relying only on few organisms do not indicate the overall state of the aquatic ecosystems but may give too optimistic picture of the state. Thus the response given by the aquatic organisms indicates a certain water quality and together with practical water monitoring and the results of studies in other fields, direct to a source of contamination.

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1 Introduction

Heavy metal occurrence in soils and aquatic environments is due to either transfer from the parental rock or to anthropogenic contamination. Regions with bedrock abundant in ore and metallic minerals have naturally high amounts of harmful elements in soil, surface- and groundwater. Mining activities tend to intensify the release of geogenic metals (e.g. Cd, Cr, Cu, Pb, Ni, Zn, Ag) into the environment and concentrate them in solid wastes or drainage, which creates a serious threat to human health and an ecological risk for the environment (Sims, 2010). Unlike organic contaminants which are oxidized to carbon dioxide by microbial action, heavy metals released in the environment by the anthropogenic activities do not suffer microbial or chemical degradation and can persist for a long time after their introduction (Adriano, 2003). These inorganic contaminants may be further mobilized by physical, chemical and biological activities and especially if the ecosystem has developed in the presence of naturally low background concentrations, elevated concentrations may pose a risk to local ecosystems (González et al., 2011). Increases in total dissolved base metals generally correlate with increases in associated pyrite content (sulphide minerals), decreases in acid-neutralizing capacity, and increases in base metal content of deposits (Plumlee, 1999). Weathering and oxidation of pyrite occurs naturally when exposed to atmospheric conditions. Anthropogenic activities, such as mining, greatly accelerate the weathering of reactive sulphides because it creates conditions that tend to facilitate movement of air and water, exposes large volumes of material, increases the surface area of the reactive component, and provides the opportunity for colonization by microorganisms that catalyse the oxidation processes in the presence of acidity. Therefore, the potential environmental consequences of human activities can be significantly more noticeable than those resulting only from natural processes (INAP 2016).

Although the environmental impacts of mining tend to be rather local, water as the greatest pathway for mining related contaminants may widen the impact on ecosystems even further off the mine (e.g. Lottermoser 2002, Kauppila et al. 2013). On the other hand, the dust tends to carry emissions only around 500 m from the mine site (Kauppila et al. 2013), but as the dust derived contaminants often bond only loosely on the surface layers, they are readily leachable and thus posing a risk to ecosystems (Neitola et al. 2015). Excluding catastrophic accidents such as dam failures, the impact on the ecosystems is usually not abrupt, but often takes time to evolve. For instance, the acid mine drainage (AMD), as the most significant reason for metal-rich mine effluent, slowly generates the contamination, and often the biggest impact is seen in the post-mining period (e.g. Lottermoser 2002, Wolkersdorfer 2008, Kihlman 2012). On the other hand, bioaccumulation in organisms is also lengthy process which depends on various factors (e.g. species, chemical speciation, and bioavailability). The most characteristic feature of mine impact on ecosystems is a reduction in diversity as well as a distinction and/or flowering of certain “indicator” species. These indicator species either thrive or disappear in different conditions (e.g. pH, metals, nitrogen compounds, and salinity). This, however, applies mainly with some aquatic organisms such as benthos and diatoms. For instance fish and invertebrates are more complex and although metals have been proved to have an effect on individuals, populations and communities, the sensitivity varies not only among species, but also within species (Tipping et al. 2009).

Mining contamination has a strong influence on aquatic systems, because the contaminants are transported throughout, by surface and groundwaters. Acid mine drainage resulting from mining operations influence the viability of microorganisms and microbial-mediated metabolism rate in the contaminated waters. Due to the inhibition of microbial activity by low pH, the microorganisms are less able to metabolize cellulose and lignin, a fact that leads to greater rates of organic material deposition in acidic habitats. Thus, the rate of microbial decomposition of leaves is lower in acidified streams, which may alter the associated food webs. Diversity of algal communities and aquatic invertebrates also decreases when an aquatic system becomes acidic (Dodds, 2002). For instance, the most mayflies disappear at pH 6.5, caddis flies and stoneflies disappear at pH 5, and beetles, dragonflies and damselflies disappear at pH 4.5-5 (Jeffries and Mills, 1990).

Abovementioned data suggest that microorganisms and their metabolic activity play a significant role in the functioning of aquatic ecosystems. Therefore, the quantitative analysis of microbes as well as aquatic invertebrate monitoring in mine drainage, surface and groundwaters can be a method for assessing mining contamination risk. As the current ecological risk monitoring is often based on the exposure for a single metal and water quality criteria instead of speciation, toxicity, availability, complexation or overall exposure condition, it is unlikely to detect the short-term increases in concentrations or the bioavailability (Tipping et al. 2009). Moreover, since the ecotoxicological impacts tend to evolve in time, a single sampling period will not describe the changing conditions in long term either. Although the toxicity of dissolved metals or acidity is rather easy to attribute, increased metal concentrations will not always imply toxicity as metals are not necessarily in bioavailable from (e.g. Adams et al. 2000, ERMITE consortium 2004). Also, other types of impacts such as salinity on ecosystems are far less studied. Knowing the chemical speciation and the field conditions, such as complexation reactions, competition effects, pH, is essential for reliable prediction of the ecological impacts of mining. On the other hand, the studies are often focusing on species or individuals and ecosystem scale effects are poorly understood. (e.g. Younger 2002, Johnson & Hallberg 2005, Wolkersdorfer 2008, Tipping et al 2009). Nevertheless, as the impacts of mining on the ecosystem cannot be ruled out, reliable and better tools to detect adverse ecological impact of mining are definitely needed.

1.1 Aims of the study

This report describes three different environmental risk assessment methods applied at case study sites in Romania and Finland. The aim of the risk assessment studies was to evaluate ecological and health risks related to mining, as well as to assess the usability of risk assessment tools to improve water management strategies at mine sites. This study provides ecological perspectives from three contrasting European gold mine sites: the operating Kittilä mine in Finland, and two closed mining areas in Romania, Roșia Montană and Zlatna. All study sites share the similar environmental concerns as the majority of gold and complex sulphides mining areas. The opening of mineralized magmatic bodies by mining works and the extraction of ores may result in the formation of AMD by allowing infiltrating water to come into contact with reactive minerals resulting in release of heavy metals. The abandoned Roșia Montană and Zlatna sites have been mined since Roman times, and since they lacked environmental regulations which resulted in significant environmental consequences, they still require monitoring for designing appropriate measures to diminish the contaminant release. In contrast Kittilä, as recently opened and still operating mine, has always had stricter environmental regulations and thus lesser environmental issues, but nevertheless needs also monitoring. Therefore, due to different backgrounds the case study sites had different research aims, and different ecological risk assessment methods were applied at each site. In Kittilä, the aim was to assess the usability of ecological risk assessment in water management strategies, whereas in Romania the main target of biomonitoring and diatom studies was to assess the ecological status of closed mine sites. This task pursues the enhancement of environmentally sustainable mining by characterizing and evaluating anthropogenic emissions relative to geogenic background, by modelling reactions and pathways of contaminants, and by assessing environmental risks.

2 Case studies

This chapter provide the results of contrasting European gold mine sites: the Kittilä mine in Finland, and two mining areas in Romania, Roșia Montană and Zlatna. Whereas Romania has the largest gold resources in Europe, but no currently operating goldmine, Kittilä mine is Europe's largest operating gold mine. Characteristics from each of the three sites are summarized in Table 2.1.1.

2.1 Kittilä mine – Finland

2.1.1 Site background

More detailed site background description of the Kittilä gold mine (e.g. geology, mining operations) is presented in Papp et al. 2018, the site background is presented only shortly and mainly the issues affecting ecological status of the nearby Seurujoki River are discussed here. Moreover this case study on ecological risks at Kittilä mine is published as a Master thesis (Malinen 2016) and only the main results and overall description of the methods can be found here.

Kittilä gold mine is located in Finnish Lapland approx. 68° northern latitude (Figure 2.1.1). It is the biggest operating gold mine in Europe with more than 2 million ounces of known gold resources. At the moment the mining company is applying a permit to expand the production from 1.6 Mt/yr to 2 Mt/yr. The climate is classified as subarctic, with yearly mean temperature being below 0 °C and snow cover ranging normally from October to May. The vegetation zone is northern boreal and the mine area is surrounded by natural wetland area with 1-2 m thick peat deposits (Aluehallintovirasto 2013). The soil consist mainly of low-permeable (10^{-9}–10^{-5} m/s) glacigenic sandy and gravelly tills. (GTK 2003-2009; Pöyry 2010). Due to the local arsenopyrite bearing bedrock, the concentration of As and Sb in soil, aquatic sediments, ground and surface waters are naturally elevated in Kittilä region (Lahermo et al. 1996). The mine site is located at lower reaches of Seurujoki River catchment (ca. 307 km^2) which drains further in south to rivers of Loukinen and Ounasjoki (Aluehallintovirasto 2013).

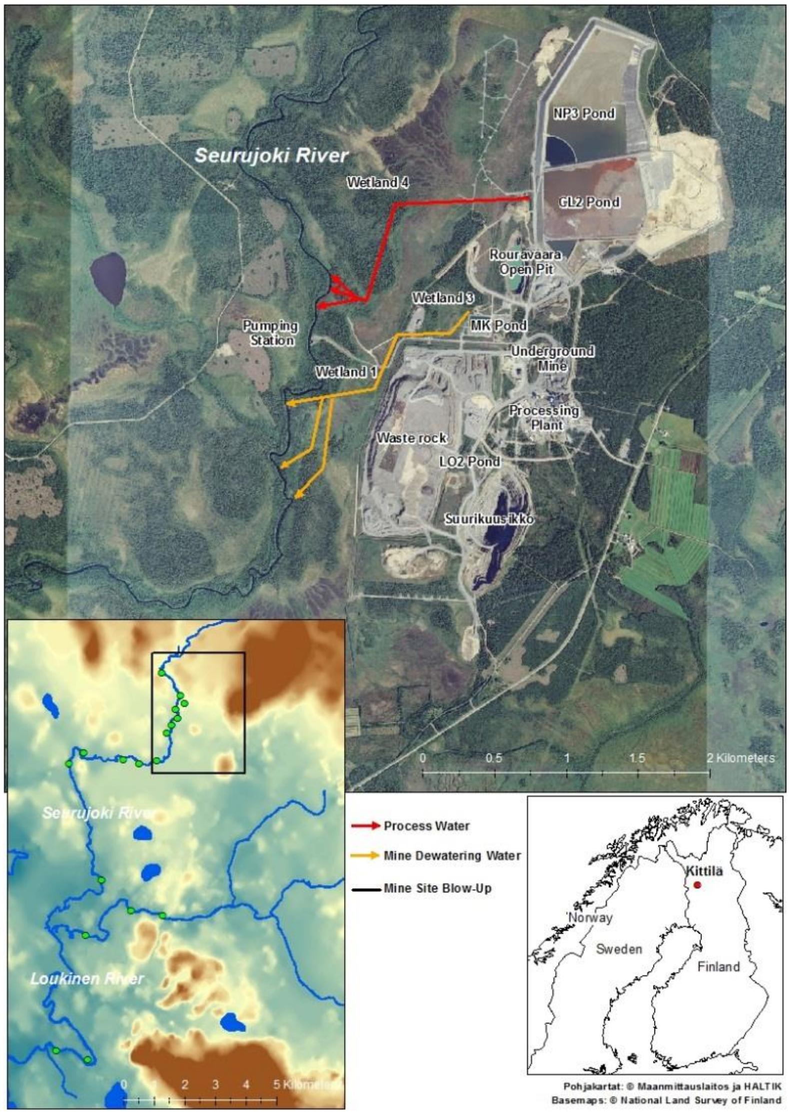


Figure 2.1.1 Location of Kittilä mine major operations, sampling sites and mine water discharge flow paths.

The process and dewatering waters are treated at the site. The most significant contaminants in mine waters at Kittilä site are sulphate (SO_4) and nitrogen compounds (mainly nitrate). The nitrate originates mainly from the explosives, and SO_4 from the bedrock minearalogy. Process waters contain also metals and metalloids such as Fe, Mn, As, Sb and Ni. The solids originate mainly from the underground mine workings (Palmer et al. 2015). The metals are precipitated from the processing sludge by neutralisation (liming), after which the sludge is directed to tailings pond (NP3) (figure 2.1.1). In the tailings pond the water is separated from the solids. Some of the treated process water is recycled back to the enrichment process and some is directed into treatment wetland 4 (TW 4) (Figure 2.1.2). To remove suspended solids dewatering waters are pretreated with ferrisulphate (PIX). The pretreated dewatering water is directed to treatment wetlands 1 and 3 (TW 1 and TW 3). The treatment wetlands are used to delay the flow and to improve the water quality by naturally occurring biochemical processes (sorption and precipitation). From the treatment wetlands, water is directed via ditches to Seurujoki River. The cyanide-containing water is excluded from the circle, cyanide destroyed and the water stored in separate CIL ponds. (Pöyry 2012a).

According to the previous studies (e.g. Päkkilä 2008, Palmer et al. 2015), the treatment wetlands adsorb well As and other metals and metalloids, but the retention efficiencies differ between wetlands and also temporally. It also seems that the treatment wetlands have not reached yet their maximal contaminant adsorption capacity, but the retention of Fe, Mn and SO_4 is less efficient or they were even leaching from the wetland. In places the thresholds of As, Sb and Ni for polluted soil were exceeded or likely to be exceeded within 6, 10 and 18 years respectively. At the time of the studies the treatment wetlands had been used for six years and since the targeted lifetime of the treatment wetlands is 20 years, it can be assumed that before the mine closes the wetlands will be heavily polluted (Palmer et al. 2015). As in nature the physico-chemical conditions tend to fluctuate and the quality of the mine water change, there is a risk of contaminant leaching from the wetlands to surrounding environment.

The Seurujoki River is a rather infertile river with clear water. The mining operations have increased the concentrations of nitrogen compounds, sulphate, antimony and manganese in the Seurujoki River and there is a slight increase also in levels of some other metals. It is estimated that approx. 2/3 of the N load to Seurujoki River originates from the mine (Pöyry 2012a). In addition to mining, forestry has changed the quality of the river water and the Seurujoki River is no longer in its natural state. According to the previous monitoring campaigns, the overall ecological status of the river remains good (Aluehallintovirasto 2013) and there is no indication of any significant impacts on the fish fauna or benthos communities. However, there have been changes in the diatom communities both at Seurujoki River and Loukinen River after opening the mine and the communities differ from the surrounding rivers. Thus the ecological status of the river is classified as poor. Fortunately, there has not been any indication of changes in diatom communities between years 2010-2015, meaning the ecological state of diatoms has balanced to its current state. (e.g. Hamari 2007, Kiviniemi 1999, Pöyry Finland Oy 2012-2013, Ramboll Finland Oy, 2014-2016). According to ecotoxicological studies conducted by Neitola et al. 2015, the waters directed to wetlands were ecotoxic to *Daphnia magna*, but since the water in outlet ditches was no longer ecotoxic, it seems that due to dilution and metal retention, the toxic effects are diminished at the wetland.

In future, as the mine operations expand, also the amount of mine waters and especially the nitrogen emissions and salinity are expected to increase in Seurujoki River (Pöyry 2012a, Aluehallintovirasto 2013). This may weaken the ecological status of the river. To decrease the SO_4 concentrations, the mining company plans to open a treatment plant to treat the process waters. The treatment will also reduce the concentrations of some metals and metalloids as the sulphate precipitates. However, as the mine workings extend deeper, the salinity of the dewatering waters increases, and despite of the SO_4 treatment facility, the salinity of the mine effluent remains at current level. The mine company is assessing a possibility to discharge the effluent via pipeline to lower reach of the Loukinen River. This would not increase the contaminant load in the rivers but the adverse effects would be allocated to a smaller extension of the river systems. Moreover, the water volumes and flow velocities of the Loukinen River would most likely enable faster mixing and dilution than current discharging point in Seurujoki, and the adverse effects on the river systems would be diminished.

Table 2.1.1 Summary of environmental and historical mine site conditions

	Kittilä mine, Finland	Roșia Montană mine, Romania	Zlatna mining area, Romania
Average annual temperature	-1.3 °C	7.4°C	10.5 °C
Average annual precipitation/evaporation	Precipitation: 55 cm/yr Evaporation: 25 cm/yr	Precipitation: 74.5 cm/yr (SRK 2012)	60 cm/yr 45 cm/yr
Vegetation	Northern boreal vegetation	Nemoral formations (temperate forest – deciduous and coniferous)	Nemoral formations (temperate forest – deciduous and coniferous)
Proximity to residential areas	50 km northeast of Kittilä, Finland (population 6 421)	5 km northeast from Abrud, Romania (population 5000)	4-5 km NW of Zlatna 2-3 km N of Almașul Mare 1-2 km N of Techereu
Economic mineralisation	Refractory gold (Au) within arsenopyrite and pyrite	Native gold, electrum gold (gold-silver) associated with sulphides (predominantly pyrite)	Au-Ag and Au-Ag-Pb-Zn-Cu mineralisations
Host rock	hydrothermally altered meta-oceanic sediments with 10% sulphides within tholeiitic basalt (Proterozoic Kittilä group)	Neogene maar-diatreme volcanic complex intruding Cretaceous detrital sediments	ophiolite, conglomerate, sandstones and clays, andesites
Underground workings	exist beneath open pits and are being actively expanded	140 km of underground workings	underground workings 2 mines: Haneș and Stănija
Open pits	2 open pits totalling 35 ha	2 open pits totalling 24.95 ha	no open pits
Mining operation dates	production began in 2009 and projected to continue to 2037	>2,000 years bp to 2006	>2,000 years bp to 2007
Current mining activity	Underground mining ongoing with planned expansion	No current mining, but mining permit being sought by Roșia Montană Gold Corporation	mining activities were abandoned in 2007 without planned reopening

2.1.2 Materials and methods

The aim of Kittilä case study was to assess if and how ecological risk assessment could be used to improve water management studies at mine sites. The case study focused on certain potentially harmful substances (SO_4, Cl, NO_2+NO_3, Ni, As, Sb, Fe and Al) and their impact on aquatic ecosystems in Kittilä gold mine. These elements were selected based on the monitoring data obtained from the mining company. The study assessed if the ecosystem has changed during mining and what are the ecological impacts of the mine drainage and process waters in Seurujoki and Loukinen Rivers.

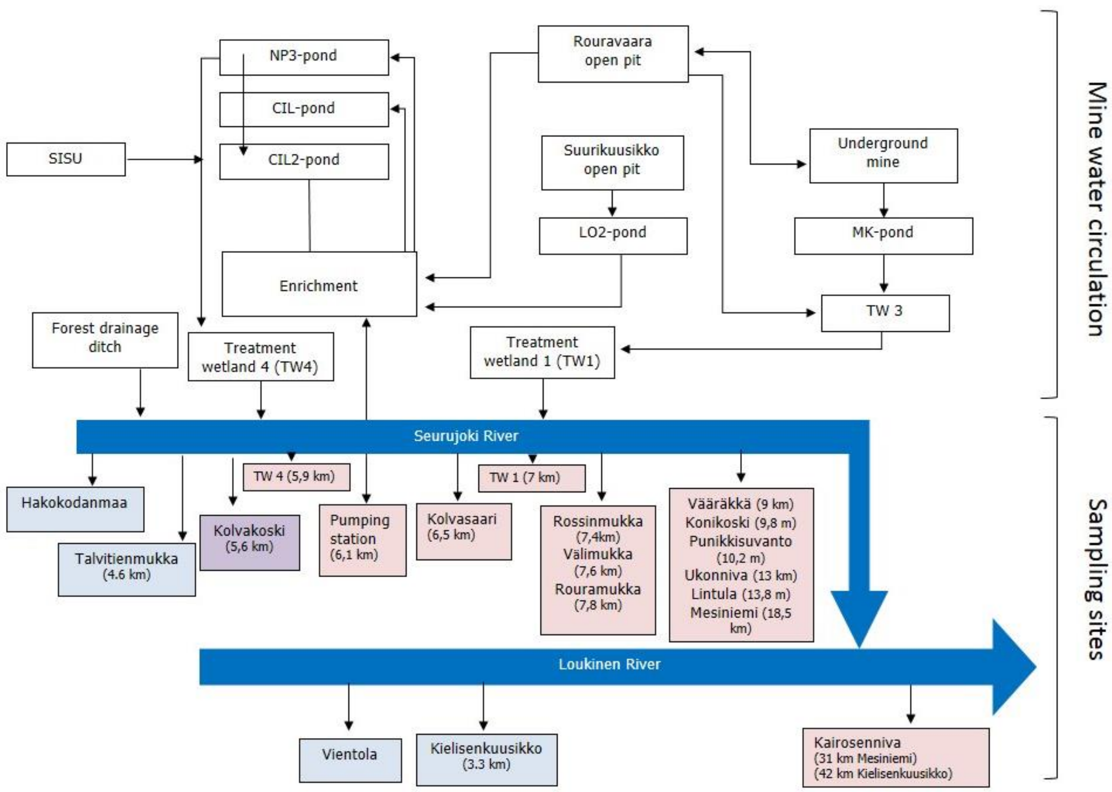


Figure 2.1.2 Chart of mine water circulation in Kittilä mine and sampling points in recipient Seurujoki and Loukinen Rivers. The distance to upstream sampling point Hakokodanmaa except for Kielisenkuusikko the distance is from Vientola is presented in brackets. (Modified from Hämäläinen 2015 and Malinen 2016).

The first step of the study was to analyse the already existing water and benthic organisms data to find trends and to create a scenario of the behaviour of the elements in treatment wetlands (retention) and recipient Seurujoki and Loukinen Rivers (load). The data comprised the annual environmental reports of water discharge and benthic fauna observation reporting done approximately every two years by various consultant companies between years 2006-2014 (LVT 2007, Pöyry 2008, 2009, 2011, 2012b and 2013, WSP Environmental 2010, and Ramboll 2014 and 2015b). In addition to already existing data, the results of the field campaign conducted at the site in July 2015 were applied for the study. This study included 15 water, 10 benthic fauna and 9 sediment samples taken from Seurujoki River, upstream and downstream from the mine site, and its recipient Loukinen River (figure 2.1.3). To be comparable with already existing data, the samples collected for the study were located as close as possible to the previous monitoring sites of the mining company. However, since the river basin is mainly stony and finer sediments exist only locally, the sediment and benthos sampling points were located where sediment was available and thus there might be slight variation in the exact sampling sites (Malinen 2016). The Figure 2.1.3 presents water and benthos sample points of the previous monitoring campaigns (AEF) and the sampling done in this study (GTK). The schematic figure of water circulation of Kittilä mine and its recipient river systems is presented in Figure 2.1.2.

The benthic fauna sampling was carried out according to a guide by Meissner et al. (2013), which applies the standard SFS 5077. The samples were collected from the riffles of Seurujoki River in June 2015 and analysed at Probenthos Oy laboratory. Since most of the benthos species mature during summer months, leaving the river basin, the benthos sampling is usually done during spring or autumn to have maximum diversity of species. Four samples were collected from each sampling point with a net. To have the total diversity for the whole width of the river present, the samples from one sampling point were taken as varying river bed type as possible (e.g. varying size of rocks, sediment type), and the results of all four samples are presented combined.

The sediment and water samples were analysed in an accredited laboratory of Labtium Oy. The bioavailable concentrations of elements in sediment samples were analysed using acetic acid extraction according to Heikkinen and Räisänen (2008). The total concentrations were analysed using nitric acid digestion in microwave oven according to Niskavaara (1995). The water samples for anion analysis were analysed from unfiltered, unpreserved water. The water samples for soluble and total trace element analyses were preserved with suprapure® HNO3 and for soluble elements also ultrafiltered (0.2 µm).To avoid contamination in field conditions, the water sample pretreatment was conducted in the laboratory facilities of Kittilä mine on the same day as sample collection. Samples were kept cold and dark and transported immediately to the laboratory for analysis. The concentrations of dissolved metal and metalloid in water samples were analysed using ICP-MS or ICP-OES – method, and anions by using IC-technique, according to standard SFS-EN-ISO 10304. Physico-chemical parameters (temperature, dissolved oxygen, pH, redox potential and electrical conductivity) were collected at field using an YSI Professional Plus multi-parameter probe (YSI Incorporated 2009).

For the ecological risk assessment the elemental concentrations in river waters (PEC) were compared to the national and international guidelines and Predicted No Effect concentrations (PNECs) which are presented in tables 2.1.2 and 2.1.3. The calculated risk assessment provides an estimate of the level of exposure, on the basis of which the risk estimate is conducted. PEC describes the exposure and PNEC the sensitivity of the ecosystem. The PEC/PNEC –ratio is rather general tool describing only the likelihood of negative impacts and not the extent of the effects. If the PEC exceeds the PNEC, the concentration may be harmful for the environment in proportion to the ratio of PEC to PNEC. The PEC being lower than PNEC, the risks are unlikely to occur (EC 2003 and ERMS 2005). PNEC is usually derived from standardized toxicity tests, in this case the Species Sensitivity Distributions (SSDs) drawn according to data from US EPA Ecotox database were used (EPA 2015b). To be able to create a representative SSDs for elements of interest in Kittilä mine environment, the chemical speciation of the elements was needed. The speciation in river water samples was modelled with PhreeqC modelling software (Parkhurst and Appelo 2015). However, in this case for most of the elements there were not enough data for a reliable SSD and thus the PNEC values should be considered as general toxicity threshold values, not site-specific guidelines.

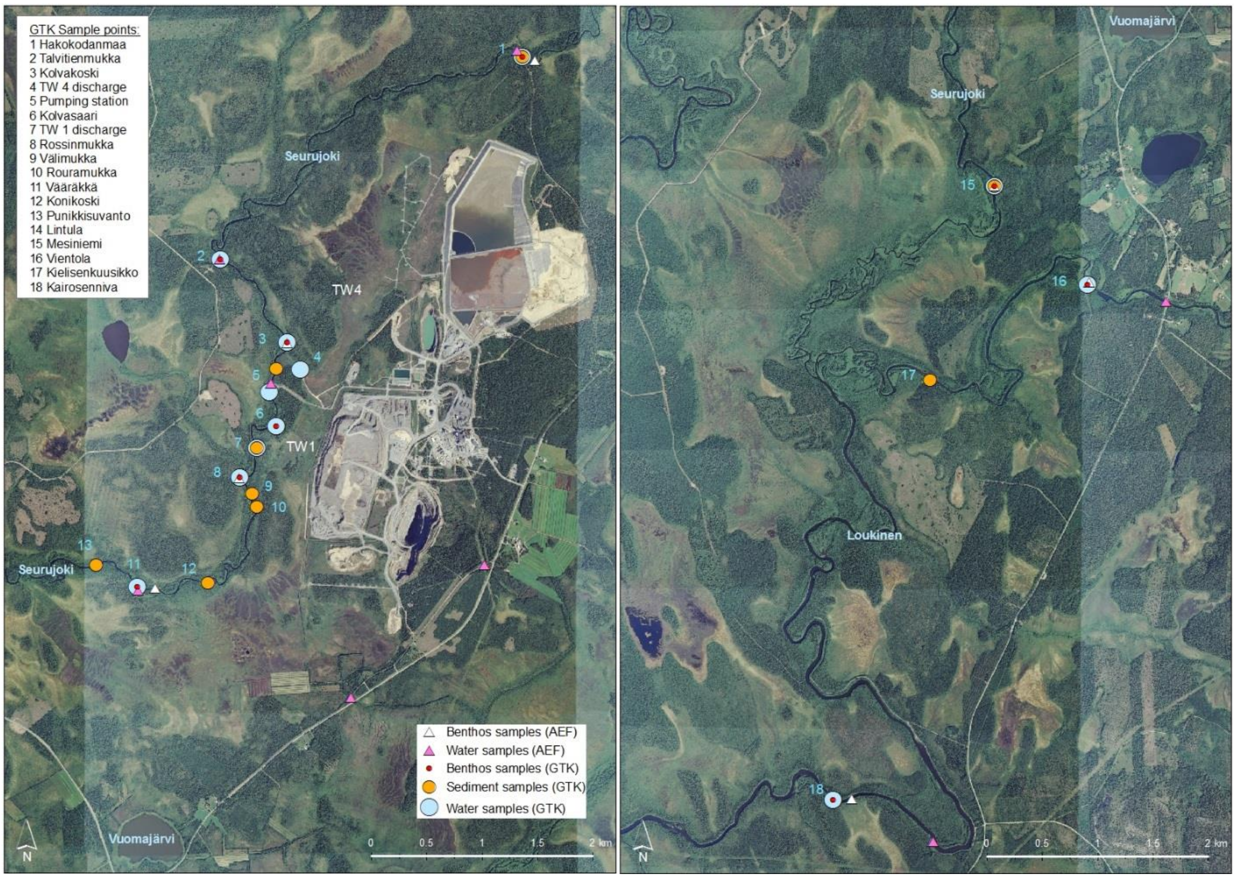


Figure 2.1.3 The benthos, water and sediment sampling locations in Seurujoki and Loukinen Rivers. GTK refers to the samples taken in this study, AEF refers to the samples taken by the mining company for surveillance of which the data was used as a part of the ecolological risk assessment in this study.

For benthos the study used the EPT (Lenat 1988) and Shannon-Wiener diversity (Shannon and Weaver 1949) indices which both describe the species richness and evenness. The EPT index is based on presumption that higher water quality means higher species richness, and then also higher EPT index. In EPT index only certain fly species are taken into account since they are known to be sensitive to contamination and thus excellent indicators for water quality. Shannon-Wiener index is based on the proportional abundance of species and assesses the current conditions. The reference Shannon-Wiener's diversity index value for Northern boreal geographic region is 2.65 (Hickey and Clements 1998, WSI 2000, Swedish EPA 2000).

2.1.3 Results and discussion

The PNEC values are presented in Tables 2.1.2 and 2.1.3, excluding nitrogen and aluminium which could not be drawn and solely the existing guideline concentrations are used as PNECS. Some of the guideline and PNEC values differ a lot or there were not enough data for reliable SSD and thus the SSD derived PNEC values should be considered as general toxicity threshold values, not site-specific guidelines. The Figure 2.1.4 shows the water chemistry and the Table 2.1.3 the concentrations in sediments around Kittilä mine. According to the results, SO_4, Sb and Ni are the most significant elements elevating the potential for ecological risk in recipient aquatic systems (Malinen 2016). However, although some higher contaminant concentrations have been occasionally detected in 2006–2015 in Seurujoki River water, on average only the concentration of Sb constantly exceeds the set guideline values (Table 2.1.2) and the overall ecological state of the Seurujoki and Loukinen Rivers was good. Also, the concentrations in sediment are mostly within the common concentrations range of Finnish stream sediments (Lahermo et al. 1996) and some of the set guideline values were exceeded only locally (Table 2.1.3). The exceeding concentrations tend to focus near mine water discharge points, but are exceeded also in background sampling point. Also, the exceeding contaminants differ between these points. For instance, As concentrations are highest in background point and do not dilute as well as other concentrations towards downstream, which is most likely due to natural geogenic background derived from arsenopyrite rich bedrock and tills. In contrast, on average the NO_2+NO_3 concentrations are above 10 mg/l in treatment wetland discharge points, which might pose a risk to aquatic life (CCME 2014). However, according to Papp et al. 2018, the dilution in the Seurujoki and Loukinen Rivers seem to be quite efficient and the fast decrease of NO_2+NO_3 concentration in the river might imply that the nitrogen is taken up by the aquatic organisms.

The benthos results are presented in figures 2.1.5 (EPT index) and 2.1.6 (Shannon-Wiener index). A clear change in indices can be seen in 2009 as the mining started at the site. Whereas the peak of 2011 in EPT index might be explained by elevated precipitation (+25% of average rainfall in Kittilä region) and thus increased nutrient level or dilution of the concentrations of harmful elements. From 2011 onwards there is a clear decline which can be seen in benthos communities as well as with the diversity of benthic fauna. This might be explained by elevated SO_4 and Sb load (Malinen 2016). However, the benthos results are slightly contradictory since the high ratio of the sensitive EPT species in the benthic community would indicate a very good state of the rivers (Hickey and Clements 1998), but on the other hand locally distributed decrease in benthos diversity in water and sediment would indicate a decline in the rivers' ecological state (Swedish EPA 2000). Moreover, as with the concentrations, the decline in diversity is seen not only at the points near mine water discharge, but also in Seurujoki River upstream, which could indicate an increased load on the whole river, e.g. due to forestry. However, the lower EPT index in background point and higher in mine water discharge points (shown in figure 2.1.5 as a "humpshaped" pattern) will also indicate the natural nutrient level of the river being low, and thus the mine waters would actually increase the nutrient load to the river. Hence, the natural load of Al, Ni, Fe, and As might have altered the community by favouring the more tolerant individuals to survive. Identifying impacts of different elements on benthos is nevertheless challenging due to spatial variation of physical and chemical factors (Pallottini et al. 2015).

Table 2.1.2 Summary table of PNEC's and the guideline concentrations of contaminants used in the study.

Contaminant	Endpoint	PNEC	Guideline conc. (definition)	Notes to PNEC
Antimony	LC_{50} EC_{10}+NOEC	530 µg/l 36 µg/l	6 µg/l (1)	Acute toxicity (AF 1000) Insufficient data
Nickel	NOEC	28 µg/l	21 µg/l (2)	Insufficient data
Arsenic	EC_{10}+NOEC	141 µg/l	5 µg/l (3)	No data for insects
Sulphate	EC_{10}+NOEC	221 mg/l	309 mg/l (3)	
Chlorine	NOEC	2 µg/l	150 mg/l (3)	Chronic toxicity (AF 50)

1) EU drinking water guideline (98/83/EC), 2) Finnish fresh water guideline (VNA 1022/2006), 3) Canadian fresh water guideline (CCME 2014)

As the mine expands, also the environmental permit and the limits for discharge waters are set again. Based on the scenarios of the treatment wetland retention capacities and dilution rates calculated for recipient river systems by Malinen (2015), the safe guideline-based levels of mine discharge for Sb would be lower than the proposed permit limits. Also, based on water quality guidelines and calculated PNECs, the proposed future environmental permit limits would allow higher discharge concentrations of As, Sb and SO_4, than would be safe for the aquatic ecosystem. However, the interpretation of this study and the proposed permit limits are based on the results of a single year (2014) and implementing site-specific guideline concentrations would require more data (e.g. on toxicity) to model such a complex system (Agnico Eagle Finland 2016). If the mine will get a permit for the new pipeline to discharge the waters in the lower reach of the river system, the treatment wetlands will no longer be used for water treatment. Although this would most likely enhance the ecological status of the Seurujoki River, the retention capability of the treatment wetlands might change as the discharging via wetlands ends and the physico-chemical conditions such as pH and redox change. Especially the mobility of As and Sb is largely controlled by pH and redox conditions.

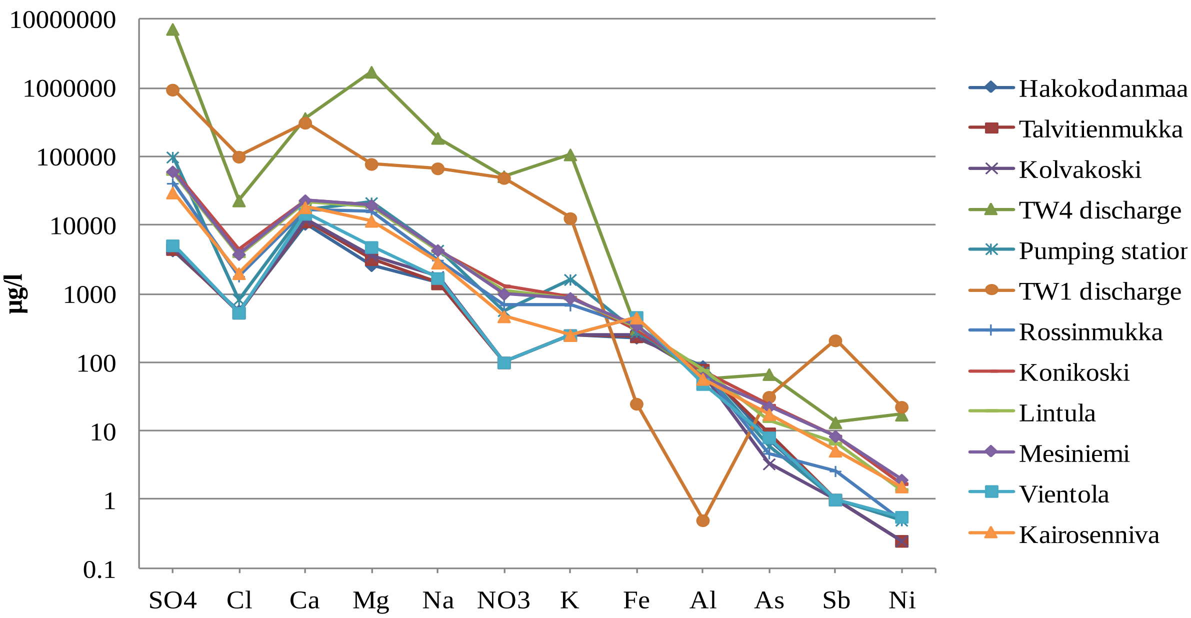


Figure 2.1.4 The water chemistry along the sampling points in Seurujoki and Loukinen Rivers

Table 2.1.3 The maximum of total and bioavailable concentrations in sample points for selected elements and the common concentrations in Finnish stream sediments and Guideline values for ecological risk assessment if available.

Contaminant	Common total concentration (90%) in Finnish stream sediments (mg/kg)	Guideline value (mg/kg), if available	Maximum concentration of sample points, total concentration (mg/kg)	Maximum concentration of sample points, bioavailable concentration (mg/kg)
Arsenic	0.8–15	10–30 (Swedish EPA, class 3)	308	15.7
Antimony	0.009–0.13	25 (WNR, probable effects)	35.6	14.8
Iron	10 000-75 000		117 000	16 900
Nickel	6–40	15–50 (Swedish EPA, class 3)	25.3	5.3
Aluminium	7 000–30 000		17 100	553
Calcium	3 000–10 000		8 910	7 450
Magnesium	1 400–9 000		13 100	11 300
Sodium	130–550		1 210	1 140
Potassium	500–7 000		1 180	804

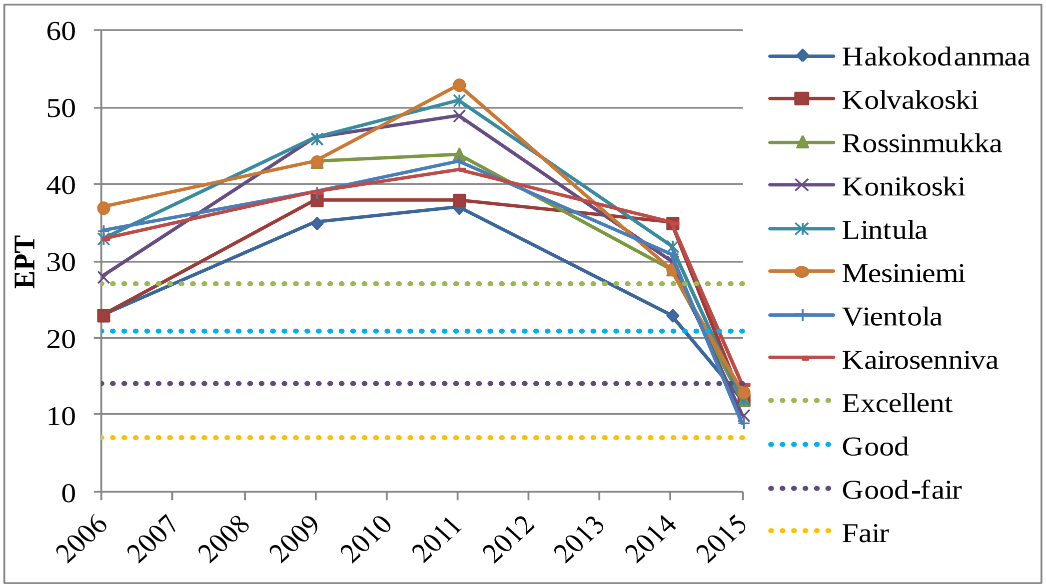


Figure 2.1.5 The EPT index value at different sample points in 2006–2015. The 2015 sampling was done during June, while others in September. The interpretation is according to WSI (2000).

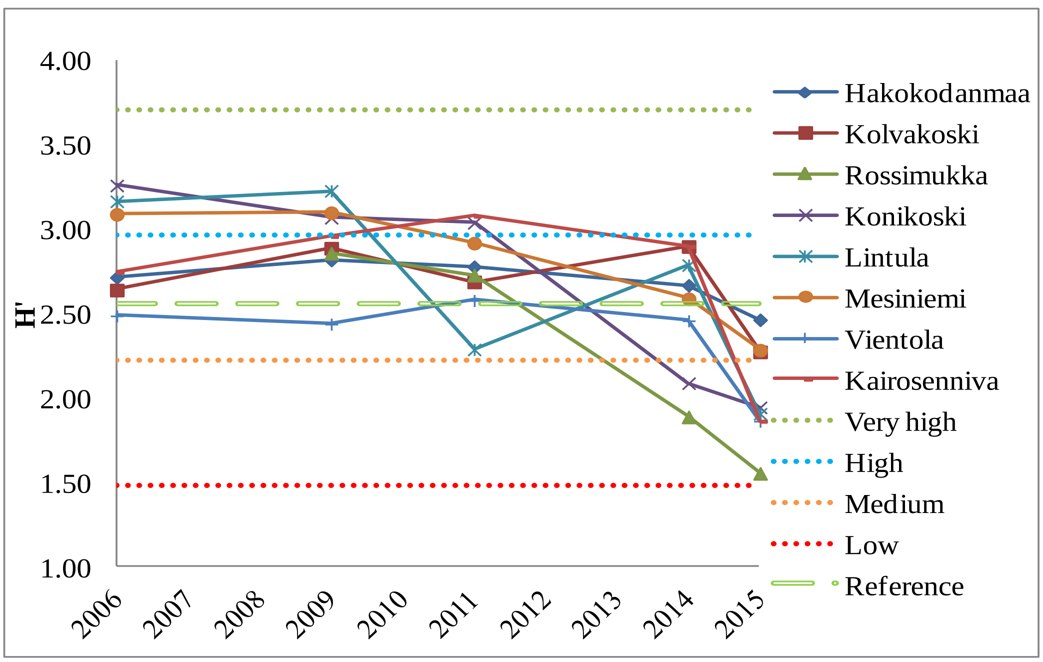


Figure 2.1.6 The Shannon-Wiener biodiversity index value at different sample points in 2006–2015. The 2015 sampling was done during June and others in September. The interpretation is according to Swedish guidelines (Swedish EPA 2000).

2.2 Roşia Montană – Romania

2.2.1 Site background

The gold and silver mineralization from Roşia Montană is hosted by volcanic rocks, mainly andesites and dacites of different types, Middle Miocene in age (Tămaş 2007). The soil cover of Roşia Montană area consists of several soil types: Districambosols, Eutricambosols, Lithosols, Regosols, Andosols and Colluvic Aluviosols (Lăcătuşu et al. 2009). The first two types are prevalent.

In Roşia Montană area, the long history of extracting ores, without proper environmental regulations and preventive actions, resulted in severe deterioration of the surrounding environment. Although the mining activity has ceased in 2006, the pollution processes are still active, mainly due to the AMD. As the human intervention in the mining area is currently very limited, it seems that the environmental processes are in a steady state. The stationary potential sources of contamination, especially affecting water, are mainly confined to the Cetate and Cârnic massifs, which host the open pits, most of the extensive network of galleries, and most of the waste rock stockpiles. Additional contamination sources are represented by the tailings piles on Sălişte Valley, and the mining works in other areas, such as Orlea and Jig. Other sources of water contamination are the uncontrolled discharges of domestic wastewater from collective dwellings and some detached houses located along watercourses. A major organic content load is also brought by the diffuse sources associated with agricultural activities and livestock (RMGC 2007b).

Several studies have been conducted in order to track the potential contaminant loads, especially of heavy metals in soil. The obtained results were compared with the allowable concentrations provided by the Romanian legislation (MO 756/1997). Previous studies were undertaken by RMGC in 2007 and 2010 on soil profiles and the upper soil horizon in order to highlight the current load or soil contamination with heavy metals prior to the mining project. According to these studies, the soils generally show low heavy metal contents, and only the concentrations of copper, lead and zinc are above the intervention threshold in few isolated cases, around the former ore processing facilities (Lăcătuşu et al. 2009). Therefore, the levels range mostly within the geochemical soil background and their abundance is not caused by the former anthropogenic activities in the area (Lăcătuşu et al. 2009).

Field campaigns have also been performed within previous projects (2011 – 2013), and more than 600 soil and sediment samples were collected from all over the study area (Lazăr *et al.* 2014). The soil samples were collected from the surface layer (0-15 cm depth). The sampling intended to cover areas with different levels of contamination, from highly impacted in the proximity of the mine, to presumably non-affected in remote zones. A selection of 262 representative samples were analysed in the laboratory for pH and heavy metal content (Cd, Cr, Cu, Ni, Pb, and Zn). It was concluded that the acidic soils predominate in the area, in relation to the geologic substrate, but the heavy metal content in topsoil is generally low in most of the cases, in the range of normal concentrations, as defined by the national environmental regulations. Lower pH and higher Pb and Cd content in soils/sediments were found in the proximity of the mining site, on the tailings and waste rock dumps, and especially along the streams carrying acid waters released by the old mining works. Nickel and chromium do not show direct relation to the rocks hosting the gold mineralization, but are more likely related to the Cretaceous flysch sediments.

Potential water-bearing rocks in the area of interest are sedimentary Jurassic and Cretaceous rocks, volcanic formations, and the shallow alluvial and colluvial deposits. Due to their structure, the sedimentary rocks, although detrital layers are included, do not provide significant amounts of water. Most Cretaceous sediments, including thick shale sequences show very low permeability. Volcanic dacites and black breccias also have a low primary permeability. The occasional permeability of the sedimentary and volcanic sequences is due to secondary structural features such as fractures and faults. The unconsolidated shallow deposits and the weathered rocks situated closer to the surface can have significant capacity for storing water in some parts, but they are too thin to be exploited as large water supplies and are more suitable as small supplies used for domestic needs. Most often, the flow of groundwater is restricted to the narrow horizon of weathered rock under colluvial soils, reflecting topography, and to alluvial sediments. The piezometric level of the shallow groundwater layer closely reflects the terrain, showing groundwater flows from the highlands to the valley bottoms and local streams. This flow model indicates that the stream flow is fed by groundwater along the entire valley length (RMGC 2007b). Also the artificial lakes are assumed to be fed by the springs and by runoff water and it is presumed that the contribution of water from the springs feeding is balanced through evaporation and seepage.

The acid water originates from the Cetate (19.75 ha) and Cârnic (5.2 ha) open pits, from the tailings facilities (Sălişte and Gura Roşiei), from the several open tunnels and numerous waste dumps which are placed very close to former extraction sites. Some open tunnels release notable amounts of acid water with pH often below 3 (e.g. Adit RO88, Cetate Adit, 714 Adit). These acid waters are collected by the local streams (Roşia, Sălişte, Corna, etc.), and finally conveyed to the main watercourse in the region, the Aries River. High concentrations of metals such as Cu, Zn, Pb, Cd, Ni are present also in stream sediments, disturbing the aquatic environment (Florea et al. 2005; Bird et al. 2005).

2.2.2 Materials and methods

In 2014, samples from 10 soil profiles, 100 cm deep, were collected within Roşia Montană area. The soil samples were analysed for heavy metals and physico-chemical features (pH and redox potential). In addition, a sampling network for monitoring the water quality has been setup. Initially, this network included 28 sampling points, and after a few months it was restricted to 24 points, based on the samples representativity. The sampling network covered a representative area in the mining-influenced zone and in more remote background areas, taking samples from surface water, groundwater, and mine water (Table 2.2.1). The water samples were analysed for anions, cations, heavy metals and stable isotopes. The physico-chemical parameters were measured in the field concurrently with the sampling. Beside the chemical feaures, the state of surface waters was assessed by studying the benthic diatom communities in rivers. The sampling sites for water and soil profiles are presented in Figure 2.2.1 and for diatoms in Figure 2.2.2.

In lotic habitats (surface running freshwater), diatom communities' structure and dynamics are influenced by a number of external factors such as light, salinity, temperature, concentration of inorganic nutrients, dissolved oxygen and nature of the substrate. Some diatoms can tolerate a small range of environmental conditions and are used as indicators of physico-chemical characteristics of water. Thus, in unpolluted rivers, diatoms are represented by many different species, each having a low number of individuals. Algal communities signal changes in physico-chemical and biological properties of aquatic ecosystems by the presence or absence of species and increasing or decreasing populations. In practice, a number of benthic algae are often used as bio-indicators in assessing the quality of natural waters (Potapova and Charles 2002; McCormick and Cairns 1997) particularly in assessing the degree of saprobity (the amount of organic contaminants) and salinity (Kiss, 1998). If the water quality is impaired due to human activity and the change exceeds the tolerance limits of species, their population will decrease or disappear. Identifying the sources that caused this decline is nevertheless easy and requires further complex investigations, but the response given by the algae community indicates a certain water quality and direct to a source of contamination.

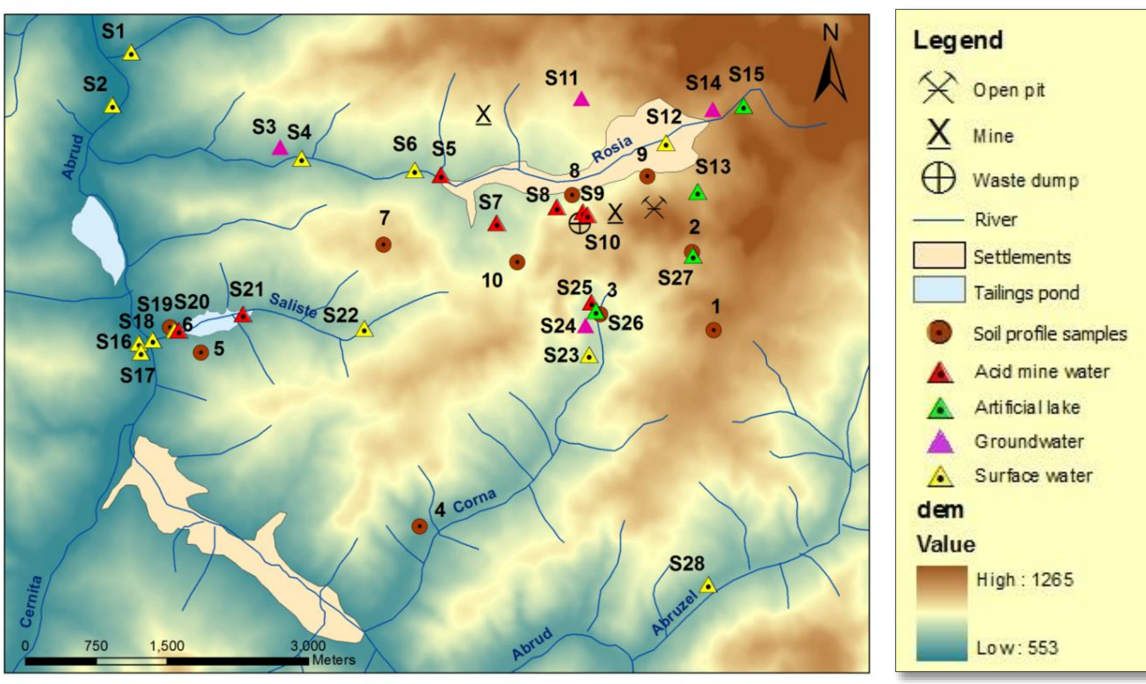


Figure 2.2.1 Roşia Montană study site and the location of water and soil sampling sites.

Table 2.2.1 The water and diatom sampling site codes, names and locations around Roşia Montană mine site

Sample ID	Name	Water type	Long E	Lat N
S1	Vârtop	Surface water	23 03 37	46 19 01
S2	Downstream confluence Roșia river with Abrud river	Surface water	23 03 27	46 18 43
S3	RO11 spring	Groundwater	23 04 50	46 18 27
S4	Downstream Roșia river	Surface water	23 05 00	46 18 23
S5	Adit 714	Acid mine water	23 06 09	46 18 16
S6	Downstream adit 714	Surface water	23 05 56	46 18 18
S7	Cetate Dump 2	Acid mine water	23 06 36	46 17 59
S8	Adit RO88	Acid mine water	23 07 06	46 18 04
S9	Cetate Adit	Acid mine water	23 07 19	46 18 02
S10	Cetate Dump 1	Acid mine water	23 07 21	46 18 01
S11	RO78 dug well	Groundwater	23 07 20	46 18 41
S12	Upstream Roșia river	Surface water	23 08 01	46 18 25
S13	Tău Brazi Lake	Artificial lake	23 08 16	46 18 08
S14	RO43 spring	Groundwater	23 08 25	46 18 36
S15	Tău Mare Lake	Artificial lake	23 08 40	46 18 37
S16	Downstream confluence Sălişte river with Abrud river	Surface water	23 03 37	46 17 21
S17	Upstream confluence Săliște river with Abrud river	Surface water	23 03 38	46 17 18
S18	Downstream Săliște TMF	Surface water	23 03 44	46 17 22
S19	Săliște Gallery	Surface water	23 03 55	46 17 25
S20	AMD exfiltration (Săliște tailing)	Acid mine water	23 03 57	46 17 25
S21	Săliște pond	Acid mine water	23 04 29	46 17 30
S22	Upstream Săliște tailing	Surface water	23 05 29	46 17 24
S23	Corna Valley	Surface water	23 07 20	46 17 13
S24	C120 dug well	Groundwater	23 07 19	46 17 23
S25	C122 Adit	Acid mine water	23 07 22	46 17 31
S26	Tău Cartuş Lake	Artificial lake	23 07 24	46 17 28
S27	Tău Corna Lake	Artificial lake	23 08 13	46 17 46
S28	Abruzel Valley	Surface water	23 08 16	46 15 53

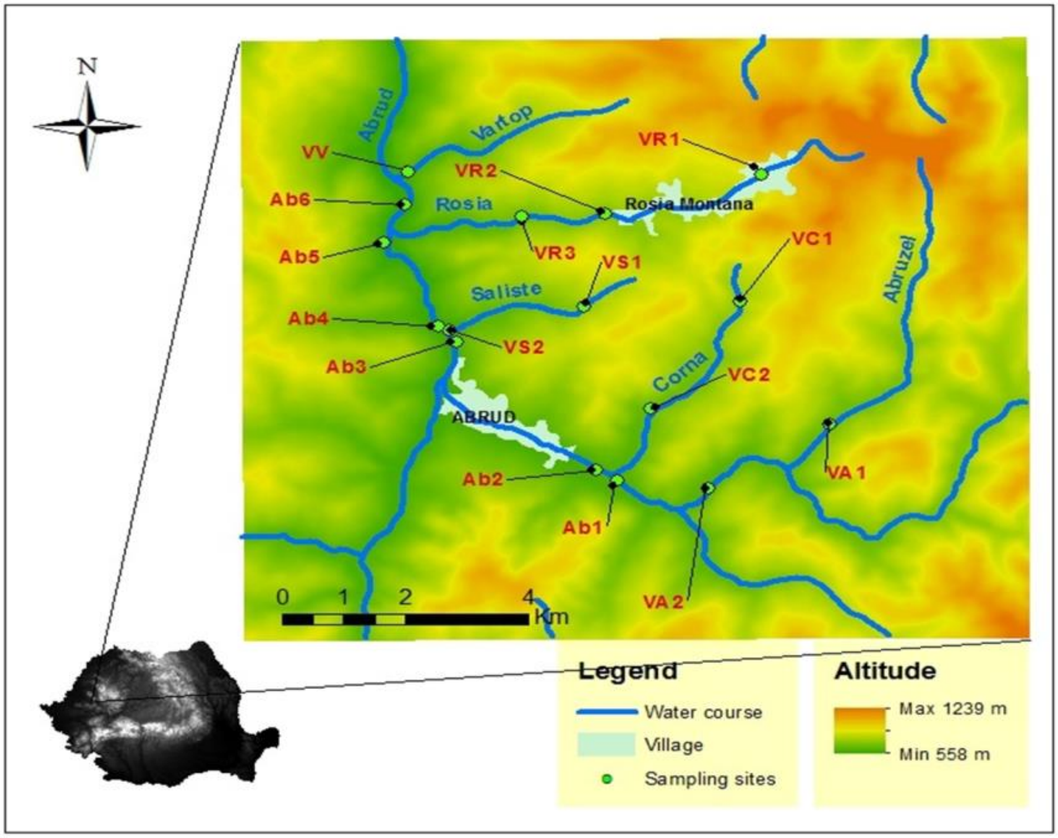


Figure 2.2.2 The map of the study area with the diatom sampling points

Studying aquatic ecology together with practical water monitoring and other fields allows an integrated multidisciplinary approach, which includes not only diatoms, but also other groups of aquatic organisms (Jonge De et al. 2008). However, a realistic and complex image on the integrity of an aquatic ecosystem as an assemblage is a mosaic image composed of the results of all studies targeting all the components (geological, physical, chemical and biological). In this context, we tried to correlate the identified diatom communities with the physicochemical features, and highlight the major factors controlling the algal assemblages. The standard maximal concentrations imposed by the Romanian legislation were applied on the mean values of the specified parameters for all water sources under study.

2.2.3 Results and discussion

2.2.3.1 *Impact on soil*

Most of the soil samples are classified as weakly acidic soils (according to RMO 278/2011), which are typical for mountain areas. It was demonstrated that most of the results fall in the range of the normal values, or between normal threshold and alert threshold, which indicates that in most cases, heavy metal concentrations correspond to the geological setting of the area (Figure 2.2.3).

Values between the alert threshold and intervention threshold have been recorded for the following metals: zinc, lead, copper, and nickel. The highest concentrations for zinc, lead, and copper were recorded close to the former mining area, near Tăul Cartuş, Cetate dump and Cetate Adit (Figure 2.2.3). High concentrations of nickel, exceeding the alert threshold, but still below the intervention threshold, were recorded in two soil profiles (profile 5 and 6) close to the Sălişte TMF. On both profiles, Ni concentration increases with depth, thus suggesting a link with the geological background. These results were consistent with the previous studies taken in 2013 on the soil superficial layer. The zinc and lead concentrations tend to decrease on most of the soil profiles, while for the other elements, differences are quite minor from one level to another. Near the mining areas, a slight increase with depth of zinc, cadmium, and copper concentrations was noticed, which indicates the influence of the mineralized rock bodies on the soil quality.

The factor analysis between trace elements suggests correlation between Cd, Zn and Cu, and between Cr, Ni and Pb (Table 2.2.2). The principal component analysis confirmed these two associations, which were related to the potential origin of the elements. In accordance with the geochemical characteristics of these elements, the association between Cd, Zn, and Cu as well as between Cr and Ni is normal, whereas the connection of Pb with the latter group was less expected. High concentrations of lead were observed at the base of Sălişte TMF, where higher values were recorded at a depth of 30 cm. This area is heavily influenced by the presence of the tailings that severely modify the concentrations of metals in soils.

Table 2.2.2 The factor analysis between trace elements (Eigenvectors F1 and F2 after Varimax rotation)

Elements	***Factor - 1***	***Factor - 2***
Cd	0.936424	-0.198692
Zn	0.961572	0.132132
Pb	0.262632	0.834835
Cu	0.986610	-0.006424
Ni	-0.130750	0.946925
Cr	-0.082583	0.962575
Expl.Var	2.867803	2.577146
Prp.Totl	0.477967	0.429524

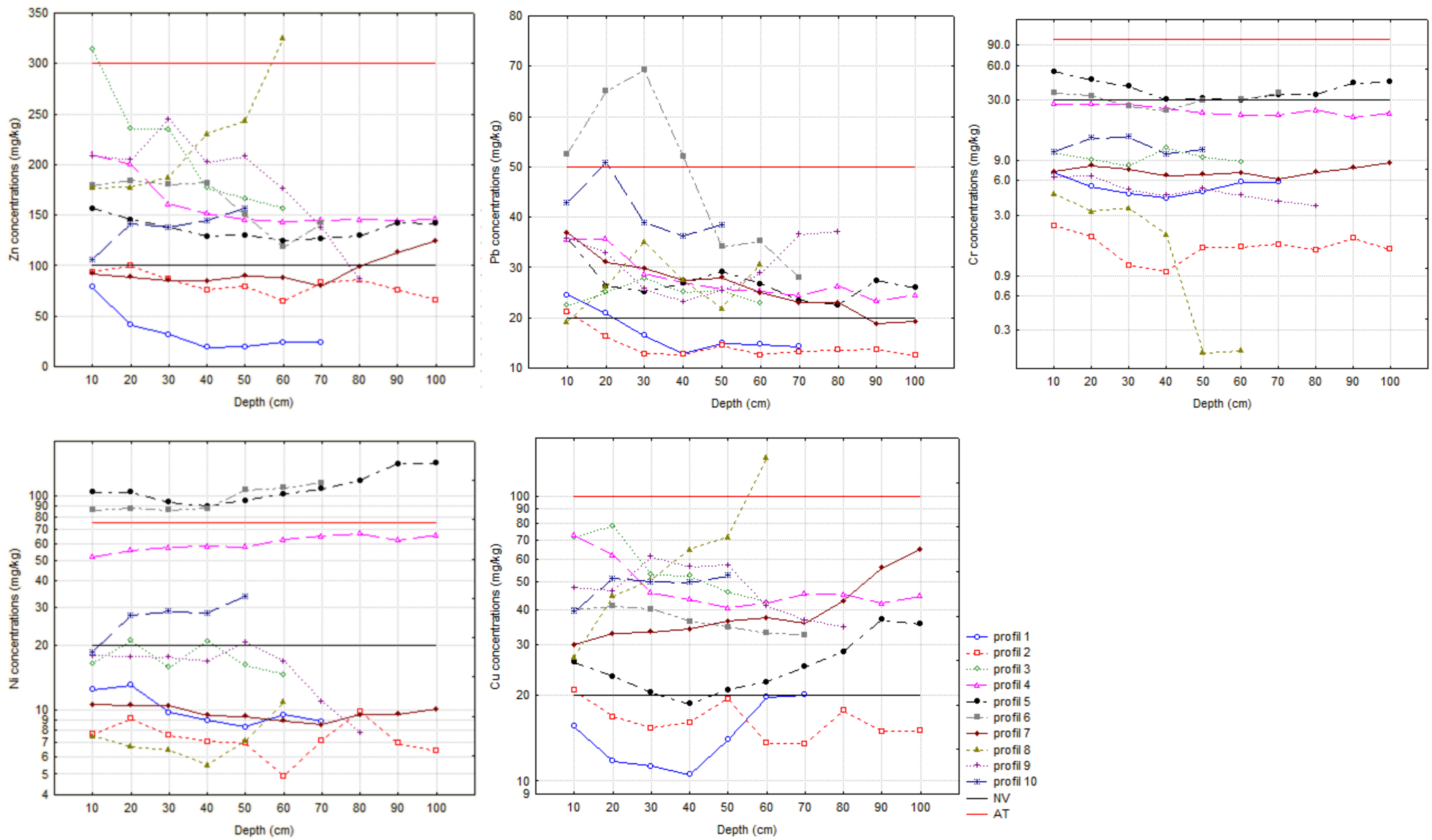


Figure 2.2.3 Heavy metal (Zn, Pb, Cr, Ni, Cu) concentrations in soil profiles depending on Depth (NV – normal values; AT – alert threshold, according to RMO 756/1997)

2.2.3.2 *Water quality*

Waters can be roughly classified into two different groups based on their acidity and metal contents (Figure 2.2.4). The waters in the vicinity of the former mining area and downstream of it show low pH and high content of heavy metals and dissolved solids, due to the acid mine drainage process. Upstream, and also on Vârtop River which is considered as a background point, the surface water quality is good. The water in artificial lakes, used in historical periods as reservoirs for process water, is also of good quality. Springs and dug wells have a relatively good water quality with some exceptions.

Roşia and Corna Streams exhibit the highest concentration of contaminants that exceed the national quality standards for surface water. Beyond Roşia Montană area, the surface waters near the Bucium area, located in the north-eastern part of Abruzel sub-basin show high contaminant concentrations (Tables 2.2.3 and 2.2.4). High acidity and concentrations of SO_4, Ni, Cu, Zn Cd, Pb, and NO_3 were found in the monitoring points S1, S4, S6, and S28. The concentrations exceed the standard limits for the 3rd class of water quality, therefore representing the 4th and 5th class, with very poor quality, according to RMO 161/2006. The high metal and dissolved solid concentrations in Roşia Stream (the most affected river) is a consequence of numerous mine water discharge points from Cetate and Cârnic open pits, from the numerous waste rock piles of various sizes, and also from the underground workings. The quality of Roşia Stream, downstream of the mining site, is very poor, being classified in the 5th quality class of surface waters.

Corna Stream is the second most polluted stream in Roşia Montană area, due to the mine waters seeping from the waste dumps located upstream of the valley. On Sălişte valley the mining influenced water coming from the tailings is mixed with the relatively clean water conveyed along Sălişte gallery (S19), this way diminishing the contaminant load. High concentrations of NO_3, SO_4, Ni, Zn, Cd, and Pb, but neutral pH were recorded downstream of Săliște TMF (S18) and on Corna Valley (S23). Although Abrud River collects all the contaminated mine water from Roşia Montană area, it has the lowest SO_4 content, less total dissolved solids, as well as lower heavy metal concentrations due to the highe flow rate. The less affected surface waters in the area are upstream of the mining workings on Roşia valley (S12), and on Vârtop Stream (S1). In lakes of Roşia and Corna Valleys, only Cd and Pb concentrations may exceed the standard maximum value.

All water discharging from old mine workings are acid and have large amounts of contaminants. The concentrations of F, NO_3, SO_4, Ni, Cu, Zn and Cd exceed the standard values for wastewater. According to the standard maximal values, the mine water from Cetate Adit (S9), C122 Adit (S25) and 714 Adit (S5) are the most contaminated, while Cetate Dump 2 (S7) shows moderate concentrations of pollutants, with the exception of SO_4. Its pH is also higher than in the case of other mine water sources. The main water supply for the Roşia Montană municipality relies on springs and dug wells. Most of the wells were dug in the shallow alluvial or colluvial deposits, whereas the springs are believed to occur due to the difference in permeability of shallow deposits. All springs and wells exhibit low concentrations of NO_3, below the standards for drinking water. In S24 the concentration of SO_4 is above the standard value for drinking water, while the other sources are below this concentration.

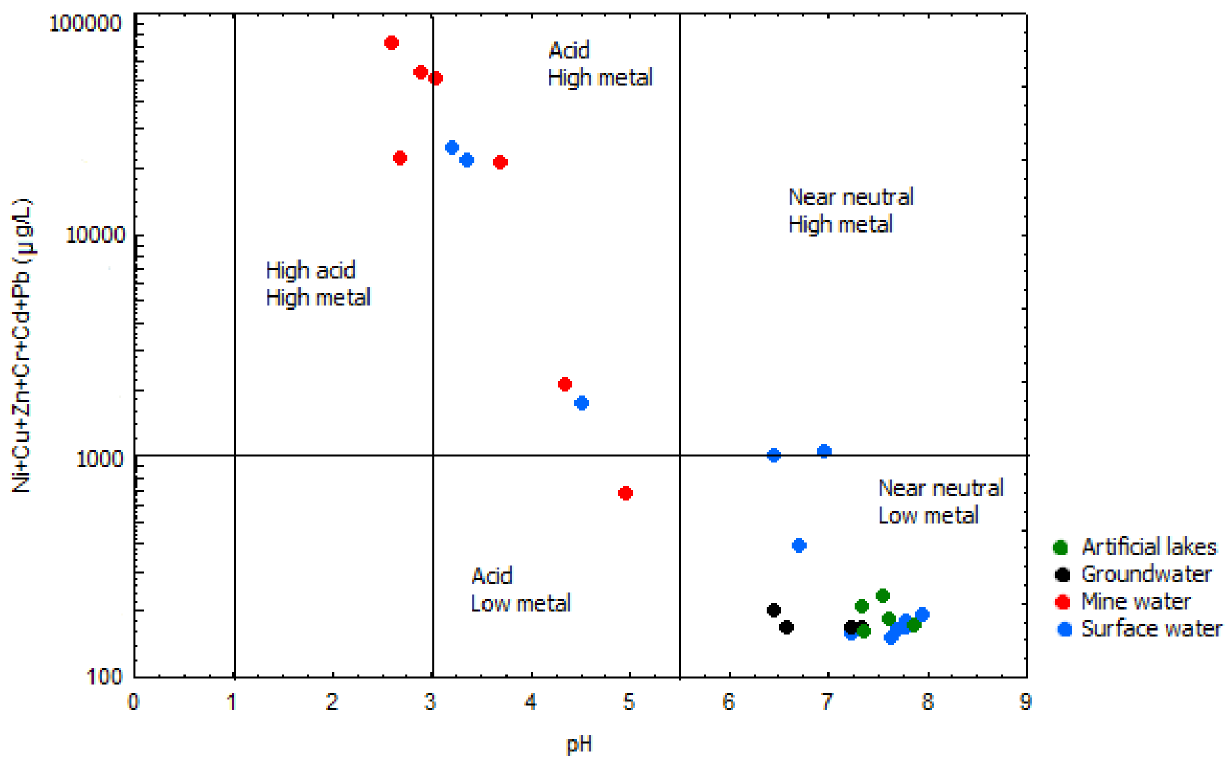


Figure 2.2.4 Ficklin diagram of water samples showing the sum of base metal concentrations vs. pH (averaged data from January 2014 till August 2015 sampling period)

2.2.3.3 *Impact on aquatic life*

Most waters in the study area are at least slightly acidic, which affected the diatom communities in Corna Stream, Săliște Stream, Roșia Stream, and Abrud River, downstream from the mine water discharges. In some sampling points the pH was neutral. The elevated electrical conductivity of water is closely linked to sulphate, alkali and earth alkali metal contents in most of the sampling sites (Savolainen *et al.* 2016). It was observed that the high concentrations of Cd, Cu and Zn are one of the reasons for the presence of the teratological (deformed or abnormal) individuals in Abrud River and even for the disappearance of the diatom communities in some sampling points in Abrud River, Corna, Săliște and Roșia streams. Moreover, the high concentrations of Ni, Fe and SO_4 led to the disappearance of the algal assemblages in the sampling sites that were mentioned before. This fact could be also attributed to the large amount of suspended matter recorded in the river and in its tributaries, displaying an orange-brownish colour. On the contrary, the relative contribution of other parameters such as O_2 and Pb concentrations, or water temperature was negligible. It could also be seen that due to the lower temperature, waters contained more dissolved oxygen in spring and autumn. The O_2 concentration was determined by the yield of the water courses and by the amount of mine water mixing with the clean water in the stream. Regarding oxygen concentrations, the various portions of the streams are classified within different classes of surface water quality, from very good to poor. The concentrations of heavy metals increased considerably during dry seasons, especially autumn, due to the low flow. Qualitatively, the number of diatom species exhibited significant variation among sampling sites, as the algal communities were strongly affected by the presence of the AMD in the study area. Some taxa suggesting critical saprobic levels on the Corna Stream were observed. This indicates a large quantity of organic matter that together with the high concentrations of NO_3, draw attention on the mediocre quality of water.

Mine drainage exerts chemical stress (low pH, dissolved metals) as well as physical stress (deposition of metal oxides) on stream biota (Niyogi et al., 2002). In Abrud River, the presence of abnormal individuals of many species was observed as a consequence of the AMD (figure 2.2.5). Furthermore, there was clear seasonal variation in benthic diatom communities, not only regarding the number of species or their relative abundance, but also regarding the type of teratology. In some seasons only individuals that presented deformed valve outlines were found, but in other seasons also individuals that presented other types of deformed valves were identified: modification of the raphe canal system (displaced fibulae) or abnormal central area location and irregular striation. The presence of teratological individuals indicates that the diatom communities were strongly affected by acid mine waters. The diatom taxa that presented a high abundance of deformed individuals were *Achnanthidium minutissimum* and *Achnanthidium macrocephalum*. The maximal abundance of abnormal individuals of both species of *Achnanthidium* was identified during summer, when the concentrations of heavy metals and ions are lower in comparison with other sampling points. Another taxon that was very affected by the presence of AMD in the study area was *Fragilaria rumpens*. More than 90% of the identified valves were deformed.

According to Ivorra *et. al.* (1999), diatom communities responded more completely to the metal-polluted conditions than to the reference water quality, but metal tolerance is not the only selective factor, as other ecological variables may influence the composition of algal communities. Moreover, the dominance of *A. minutissimum* in polluted assemblages indicated that those stations were subjected to continuous chemical and/or physical stress (Ivorra et al., 1999). The most evident response of natural diatom populations to metal contamination is the development of frustule deformations (Tonolli, 1961 and Yang and Duthie, 1993), and diatom teratology is considered the more significant indicator of metal contamination (Falasco et al., 2009). Falasco et al. (2009) reported in their review that copper contamination appears to mainly affect the valve outline, leaving the ultrastructure (striae, raphe, etc.) unaffected.

Table 2.2.3 Average concentrations of metals and major ions in different water sources in Roșia Montană mining area

ID	Type	pH	Metals (μg/L)						Anions (mg/L)					Cations (mg/L)			
			Ni	*Cu*	*Zn*	*Cr*	*Cd*	*Pb*	*F^-*	*Cl^-*	*$NO3^-$*	*$SO4^-$*	*HCO_3^-*	*Na^+*	*K^+*	*Mg^{2+}*	*Ca^{2+}*
S1	sw	7.71	27.82	7.27	22.95	6.44	17.49	21.43	0.09	10.94	3.12	22.77	343.9	7.32	18.2	10.39	49.97
S2	sw	6.23	73.36	30.32	1052.30	6.68	23.97	25.58	2.19	25.25	4.13	269.52	294.44	11.17	34.51	12.15	75.15
S4	sw	3.15	296.97	444.44	25041.95	25.74	80.24	74.50	1.76	12.07	14.99	2227.31	BDL	20.95	31.95	55.49	235.06
S6	sw	3.25	262.12	559.95	24515.36	24.53	69.81	83.46	1.92	19.17	81.62	2097.97	BDL	15.39	32.02	48.94	229.62
S12	sw	7.60	25.84	7.16	22.23	7.30	17.98	17.11	0.06	3.67	3.4	116.32	313.27	6.09	5.16	4.58	30.52
S16	sw	7.13	23.62	10.49	60.55	8.58	3.87	22.30	0.18	12.67	3.88	130.14	323.12	10.23	12.89	8.93	63.18
S17	sw	7.62	22.20	11.65	56.78	4.80	4.77	28.91	0.3	15.88	22.83	105.77	338.08	10.37	8.81	7.33	57.47
S18	sw	6.64	44.91	12.80	143.13	10.79	19.57	36.80	BDL	5.56	6.09	898.34	412.86	7.78	23.6	26.46	109.95
S19	sw	7.68	27.01	5.89	24.96	9.66	19.40	19.90	0.61	4.24	2.26	56.8	340.83	5.18	7.86	8.7	50.27
S22	sw	7.87	28.83	12.25	21.04	7.10	14.02	25.13	0.13	4.3	1.49	15.31	330.94	3.87	7.12	6.11	36.37
S23	sw	6.88	151.78	33.04	704.87	18.77	16.57	72.13	0.14	17.62	16.74	2250.5	358.63	20.69	38.05	126.55	403.22
S28	sw	4.48	86.13	906.55	1059.16	12.09	28.39	43.99	1.71	7.81	7.76	1182.45	250.08	8.61	19.62	40.88	110.15
S3	sp	7.25	17.05	11.39	23.47	5.21	BDL	23.38	0.15	3.49	1.86	27.53	431.75	9.35	7.89	7.28	54.11
S11	dw	6.61	20.62	10.30	22.41	7.44	BDL	19.48	0.07	4.85	1.75	15.47	357.18	9.72	13.26	3.82	29.58
S14	sp	7.36	21.19	9.13	25.43	7.26	BDL	18.33	0.05	1.74	1.12	10.21	293.82	5.09	4.45	2.97	22.35
S24	dw	6.54	30.08	10.40	21.45	10.06	BDL	28.84	BDL	4.4	7.19	405.20	288.78	11.26	13.04	25.78	118.54
S13	la	7.44	25.33	11.53	30.58	30.02	BDL	13.95	0.06	2.55	1.19	18.25	312.3	5.19	5.03	2.17	13.81
S15	la	7.82	23.32	8.67	22.70	7.78	BDL	25.24	0.06	4.3	1.84	28.8	302.82	4.87	4	3.12	23.68
S27	la	7.61	40.47	9.99	29.75	6.11	BDL	23.58	0.09	4.02	2.21	66.91	288.05	6.31	8.06	4.9	37.22
S26	la	7.37	26.19	9.53	31.08	8.08	BDL	16.36	0.1	7.4	1.36	26.66	319.02	1.58	11.85	1.61	19.14
S21	la	7.21	26.02	6.73	83.54	6.92	BDL	31.96	0.12	2.3	1.19	41.88	309.9	2.78	5.95	4.59	28.62
S5	mw	2.82	868.07	1535.20	52227.91	62.98	217.42	178.08	9.59	17.33	62.27	11085	BDL	38.67	59.25	222.11	659.72
S7	mw	4.71	83.36	64.01	414.79	13.32	16.64	50.17	0.36	5.6	14.08	934.21	271.23	5.1	17.95	20.78	229.97
S8	mw	3.05	359.43	806.18	46665.16	44.76	236.67	89.13	2.02	15.06	33.43	4024.77	BDL	16.14	34.97	80.84	351.51
S9	mw	2.54	1730.30	2185.59	75516.71	103.00	649.06	197.19	6.91	42.47	98.23	31051.9	BDL	32.47	68.23	359.99	841.04
S10	mw	2.65	565.93	2409.75	18621.73	62.22	42.63	98.48	2.81	18.26	24.83	5960.22	BDL	22.44	48.06	175.12	452.8
S20	mw	4.35	172.81	30.63	1284.08	38.98	15.42	156.52	BDL	11.6	243.62	9773.13	349.27	24.34	90.94	311.54	874.41
S25	mw	3.51	581.57	148.17	19052.21	34.98	42.39	163.38	4.74	141.72	46.4	5535.73	157.88	19.55	49.97	164.35	685.24

RMO 188/2002 industrial/waste water
RMO 458/2002 drinking water
RMO 161/2006 surface water, 3rd class

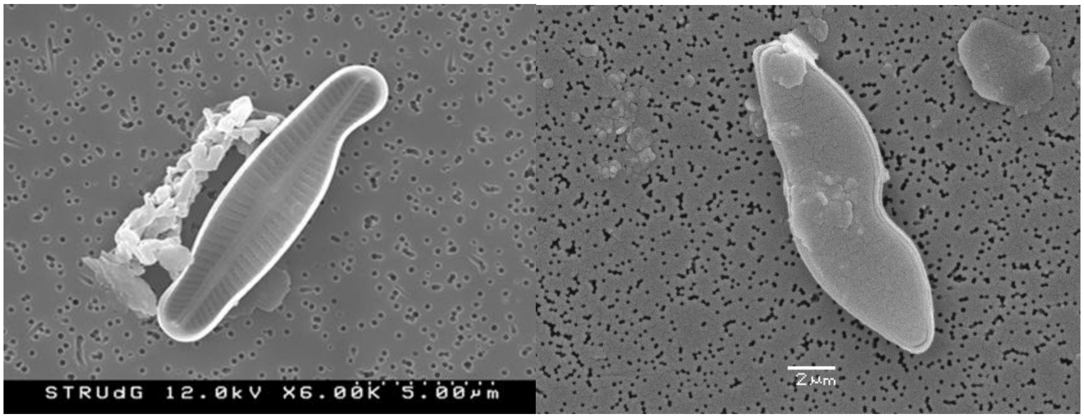
sw – surface water; sp – spring; dw – domestic well; la – lake; mw – mine water

In some sampling sites situated along the tributaries and the Abrud River, where EC was very high, algal assemblages were absent, as diatom communities were strongly affected by the presence of acid mine waters. This confirmed that the presence of elevated SO_4 and heavy metal concentrations affected the diatom communities, having as consequence a very low relative abundance, the presence of the teratological individuals, and even the disappearance of the algal communities. Additionally, in three sampling points in Vârtop, Roșia, and Săliște Streams, upstream of the mine water discharges, a high diversity of the diatom communities was observed, as the physicochemical properties were normal, this fact confirming once again that the presence of the AMD affects very strongly the algal assemblages.

As mentioned before, the number of diatom species exhibited significant variation among sampling sites and it was observed that in the study area the Zn and Cu concentrations and the high values of conductivity control the characteristics of the algal assemblages, but in our opinion it is possible that also other unmeasured factors had affected the diatom communities in the study area. In areas severely affected by the acid mine drainage, the diatom communities are almost totally disappearing. A larger number of species was found in Vârtop Valley, Roşia Valley (upstream of the waste water treatment plant) and Sălişte Valley (upstream of the TMF), while on the downstream points (Roşia Valley, Sălişte Valley and Abrud River), the number of taxa is very low or none. It was found that besides drastic reduction of diatoms and their disappearance in some sampling points, another consequence of the high concentration of heavy metals in the study area is the presence of deformed individuals of diatoms (Figure 2.2.8). In terms of flora affinity relationships, a high level of similarity was found between algal communities from the points affected by discharges of acidic waters. Also a low level of similarity was observed between algal communities from the affected points and the communities collected from the points on less polluted water courses.

2.2.3.4 *Issues of concern*

Previous studies on water quality were conducted by RMGC between 2000 and 2010 (e.g. RMGC 2007a). Comparing those results with the values obtained in 2014 and 2015, and by analysing the temporal variations of contaminant loading, it becomes clear that although mining in Roşia Montană was closed down in 2006, the water quality in the region has not improved compared to the period prior to closure. High concentrations of major ions (especially SO_4, NO_3), acidity and metal content in mine waters represent the main issues of concern in Roșia Montană mining area. These flow directly into the surface waters and affect the water quality of Roșia, Corna and Săliște streams. The most affected one is Roșia stream, with the downstream point having very low pH and greater Zn and Cu concentrations during the entire sampling period. In lakes, the concentrations of heavy metals are within normal limits, excepting Cd and Pb for some months in 2015. The Cd concentration exceeds the limits for drinking waters in most of the cases. Another concern is the high concentration of nitrogen compounds. This could be a consequence of using nitrogen fertilizers in agriculture, and/or could be related to the former mining activities, where nitrate-based explosives have been used.



Achnanthidium minutissimum *Fragilaria rumpens*

Figure 2.2.5 Deformed diatoms (*Achnanthidium minutissimum* and *Fragilaria rumpens*).

The poor water quality is the major factor affecting the benthic diatom species in Roşia Montană. In areas severely affected by the acid mine drainage, the diatom communities have almost totally disappeared. As mentioned before, the number of diatom species exhibited significant variation among sampling sites and it was observed that in the study area the Zn and Cu concentrations and the high values of conductivity control the characteristics of the algal assemblages. However, it is possible that other unmeasured factors had also affected the diatom communities.

As the release of metals due to AMD is a long-term process, the monitoring of soil, surface water, groundwater, and mine water quality is still necessary for designing appropriate measures to diminish the contaminant release that could harm the human health or the environment.

2.3 Zlatna site – Romania

2.3.1 Site background

Zlatna gold mining area (approx. 40 km^2) is located in the Metaliferi Mountains of the South Apuseni mountain range (Romania) approximately 24 km south of Roșia Montană (Figure 2.3.1). It also belongs to the "Golden Quadrilateral" mining district. The Zlatna mining group includes three mines: Haneş, Almaș and Stănija. The Almaș mine was closed during the World War II and at present the amount of drainage water from the Almaș mine is insignificant. The mining operations at the Haneș and Stănija mines ceased in 2007. Currently, the mine openings are secured, but significant flow of acid mine water continues to discharge into the river system.

Figure 2.3.1 Location of the Zlatna study area

The basement of the area consists of Jurassic ophiolites (Figure 2.3.2). The sedimentary cover, with a thickness of few hundred meters, consists of Late Jurassic – Hauterivian reef limestones, followed by a suite of detrital formations, Cenomanian – Early Turonian in age, and Late Cretaceous molassic sediments. The Neogene volcanic activity had two peaks at 14.6 - 10.8 Ma and 9.3 - 7.4 Ma, ending 1.6 Ma ago (Roșu et al. 2004). Quartz andesite with amphibole and pyroxene, and quartz andesite with amphibole and biotite are the main petrographic types. Three types of polymetallic mineralization occur: (1) porphyry type (with or without veins at the upper part); (2) vein type, and (3) breccia pipe type with transitional forms generated by remobilization. All mineralizations contain Au ± Ag, Cu, Pb, and Zn in various proportions. The Zlatna mining site has a long history of mining with significant environmental consequences related to the former ore extraction and processing. In most cases, the mine water infiltrates into the waste rock pile located in front of the mine adits, before it discharges into the river system. Waste rock piles may contain large amounts of trace metals and metalloids (e.g. Pb, Zn, Ni, Al, Cu, Mn, Fe and As), that are released through acid drainage, thus increasing the initial load of mine water. Additionally, significant amounts of SO_4 are formed as a consequence of the oxidation of sulphides. The AMD and the related mobilization of heavy metals still represent the main contaminant factor affecting the environment at the site.

2.3.2 Materials and methods

Different water quality parameters are set by several international and national regulations. In order to assess the quality of water at the Zlatna mining area we applied the following standards:

- the European 98/83/EC standard for drinking water → for springs and wells that are currently used for drinking water;
- the national NTPA-013/2002 standard for water that is suitable for treatment to produce drinking water → for running water;
- the national standard for waste water HG 188/2002 → for mine water.

The table 2.3.1 presents a summary of some of the most common water quality parameters which are regulated under these 3 standards. These standards were applied on the mean values of the specified parameters for all water sources under study.

Table 2.3.1 Water quality parameters regulated under different standards

Parameter	98/83/EC standard (drinking water)	NTPA-013/2002 standard (surface water)	HG 188/2002 standard (waste water)
pH	>6.5	>6.5	>6.5
F (mg/l)	1.5	1.7	5
Cl (mg/l)	250		500
NO_3 (mg/l)	50	50	25
SO_4 (mg/l)	250	250	600
Cd (mg/l)	0.005	0.005	0.2
Cr (mg/l)	0.05	0.05	1
Pb (mg/l)	0.01	0.05	0.2
Cu (mg/l)	2.0	5	0.1
Ni (mg/l)	0.02	0.1	0.5
Zn (mg/l)	3	5	0.5
Fe (mg/l)	0.2	1	5

The presence of microbes and their quantitative evaluation represent an indirect way to assess the ecological risks of contamination in mine water, groundwater and surface water around mining areas. Besides these, aquatic invertebrates that spend most of their life cycle in water are extremely important in maintaining the health of aquatic ecosystems and also represent a very useful instrument for rapidly assessing the contaminant' risk. Biomonitoring based on water sampling, identification and quantitative determination of living organisms is a method of assessing the quality of water sources, because the contamination and changes in habitats influence greatly the specific structure and abundance of these organisms.

Altogether 25 water samples for biological monitoring were seasonally collected from mine drainage, groundwater (wells and springs), and running surface water (Figure 2.3.2). Two types of sanitation tests (RIDA®COUNT, R-BIOPHARM AG, Germany) were used for monitoring the microbial concentration in mine, groundwater and surface waters: RIDA®COUNT TOTAL for quantitative determination of total aerobic bacteria, and RIDA®COUNT YEAST&MOLD for quantitative determination of yeast and mold. RIDA®COUNT ready-to-use compact dry plates represent a simple and safe method for determination and quantification of different microorganisms from the environment. The test principle is based on specific chromogenic substrates which are converted into coloured products by the metabolism of microorganisms.

Rida®Count Total (Total Count, TC) indicates the aerobic mesophilic count expressed by the number of colony forming units (CFU) formed on a plate count medium during a specified incubation time at mesophilic temperature (approx. 30-37°C). Compact dry TC contains a standard nutrient medium for detection of total plate count. Due to a redox indicator which is integrated into the medium, the grown colonies will be red coloured and therefore easy to identify from possible residues on the plate. The time for incubation is 48±3 h, at 35±2°C. The following taxa may grow on the dry compact medium for TC: *Escherichia coli, Klebsiella pneumonie, Pseudomonas aeruginosa, Bacilus subtilis*, and *Staphylococcus aureus*.

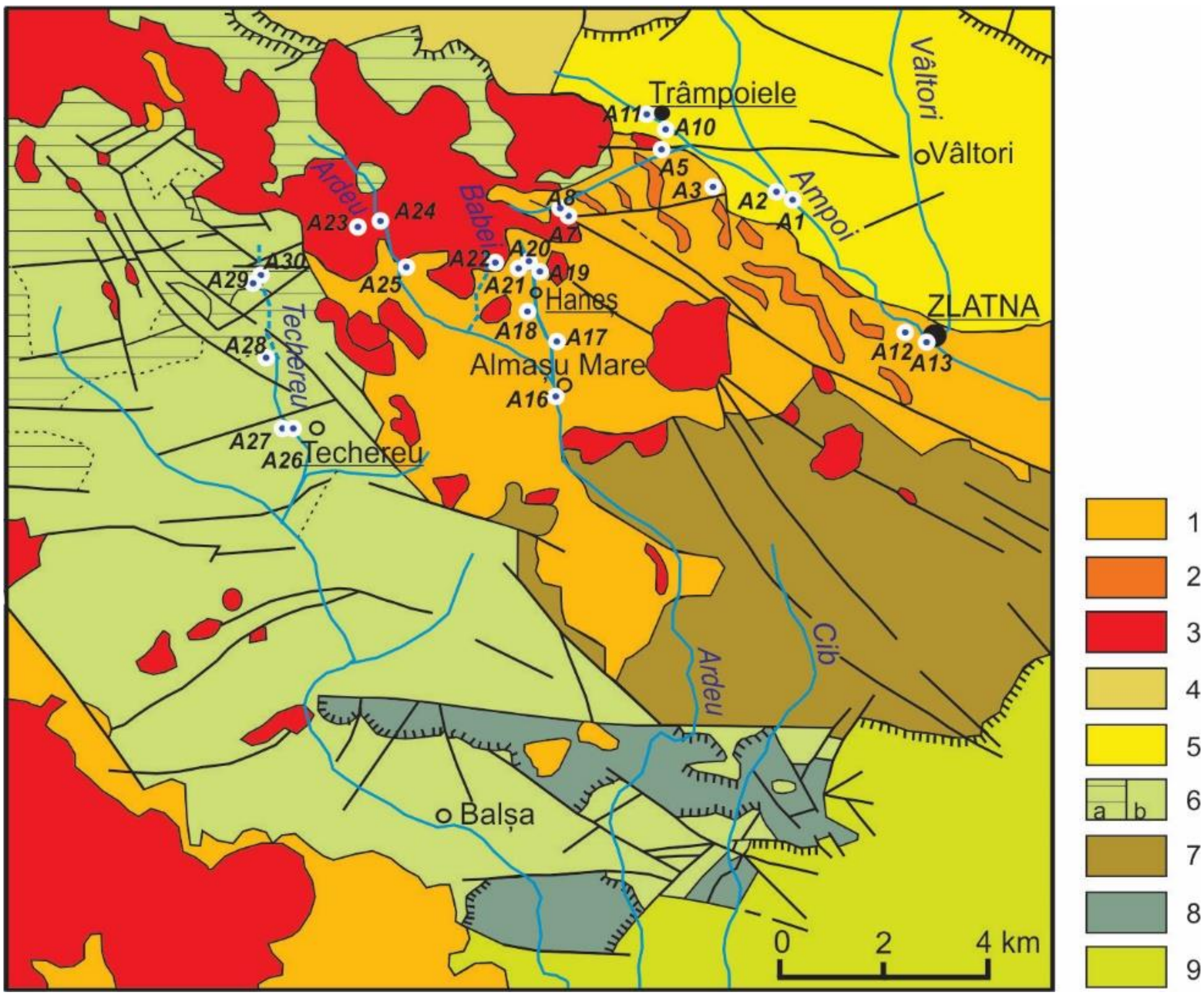


Figure 2.3.2 Geological sketch map of the Zlatna mining area showing the location of the 25 sampling sites (redrawn after the Geological map of Romania scale 1:50000, sheet 74C-Zlatna)

Legend: 1 – Neogene molasse; 2 – Neogene magmatites stage II (rhyolites and quartz andesites with Miocene sedimentary intercalations); 3 – Neogene magmatites stage I (biotite, amphibole andesites +/-pyroxene); 4 – Early Cretaceous – Middle Jurassic flysch, Bucium Unit (sandstones and conglomerates); 5 - Early Cretaceous – Late Cretaceous flysch, Feneș Unit (sandstones, conglomerates and limestones; 6a – Early Cretaceous – Turonian flysch, Techereu Unit; 6b – Late Jurassic ophiolites (basalts, pyroxene andesites and rhyolites); 7 – Late Cretaceous wild flysch, Galda Unit; 8 – Jurassic – Early Cretaceous reef limestones, Ardeu Unit; 9 – Senonian – Maastrichtian flysch, Bozeș Unit (marls, sandstones)).

Rida®Count Yeast&Mold (Y&M) are chromogenic compact dry plates with a specific nutrient medium suitable for the growth of yeasts and molds after its activation by water samples. Yeasts are facultative anaerobic, monocellular fungi (*Ascosporidae*), fermenting sugar substrate to CO2 and H2O under aerobic conditions. Under anaerobic conditions yeasts ferment sugar to alcohol and CO2. *Candida* is one of the most common yeasts playing a major role in food spoilage and also in hygiene monitoring. The term *mold* is commonly used for the visible part of the fungi present on the contaminated surfaces. It grows in the form of hyphae filaments that contain multiple identical nuclei. In culture fungi grow in the form of mycelium with different shapes and colours. After the nutrient medium activation by the water sample, during the incubation, yeast and mold colonies show different colour reactions. Almost all yeast colonies show a blue coloration and therefore they are easily differentiated. Molds will form their typical three-dimensional structures in the air space between nutrient pad and plate cover. The coloration of the air structures might be different according to the special type of mold. Growth of bacterial species on Compact Dry YM is inhibited by antibiotics which are added to the medium. The incubation time of the activated medium is 3-7 days at 25-30 °C. *Candida albicans* and *Aspergillus niger* grow on the Compact Dry YM.

In microbial monitoring, all water samples were taken seasonally, in May, August, October and December of 2015. A volume of 1 mL of water was directly applied on the plates, right after sampling, transforming the dry medium into a nutrient gel. After inoculation the TC plates were placed in a portable incubator at 36 °C for 48 hours, and the YM plates were stored at 25-30 °C for 120 hours in dark conditions. The red colonies of TC, the blue colonies of yeast and the specific coloured colonies of molds were counted in order to determine the viable microbial concentration (CFU/mL) in mine drainage, groundwater and surface water. Water invertebrates were sampled by qualitative filtration of the water sources in August, October and December of 2015. Fauna was collected from a 100 μm mesh-sized hand net. Animals were fixed in 70% ethanol, counted and sorted to different taxonomic levels, using a stereomicroscope Optika SZR-10. The biological determinations were performed at the Institute of Speleology "Emil Racoviță" Cluj-Napoca (Romania) by Dr. Daniela Borda.

2.3.3 Results and discussion

2.3.3.1 *Water quality*

The water quality parameters were chosen based on the national and European quality standards for drinking water, waste water and surface water. The standards are presented in table 2.3.1. These standards were applied on the mean values of the specified parameters for all water sources under study. The marked values in the Table 2.3.2 are above the standard limits.

In the Larga creek (A8), the Ardeu stream (A16) and the Techereu stream downstream (A26), the NTPA-013/2002 standard values for pH, NO_3, SO_4, Fe and Zn are exceeded. High concentrations of NO_3 were found in the Ampoi stream (A1 and A13) and the Trâmpoiele stream (A2 and A10). The NO_3 and SO_4 concentrations in the Techereu stream upstream (A30) exceed the standard values. To a lesser extent the concentration of Pb is exceeded in the Trâmpoiele stream (A2), the Ardeu stream downstream (A16), the Larga creek (A8) and the Techereu stream downstream (A26). In all surface waters the standard value for Cd concentration is exceeded by up to 6 times. The least affected running water in the area is the Ardeu stream upstream (A24). In this source only the Cd concentration standard is exceeded.

Table 2.3.2 Values of water quality parameters in different water sources at Zlatna mining area

Source and type		***pH***	***Anions mg/L***				***Heavy metals mg/L***						
			F^-	***Cl^-***	***NO_3^-***	***SO_4^-***	***Cd***	***Cr***	***Pb***	***Cu***	***Ni***	***Zn***	***Fe***
A1	rw	**8.04**	**0**	**7.52**	**1082**	**16.51**	**0.069**	**0.02**	**0.044**	**0.001**	**0.019**	**0.05**	**0.088**
A2	rw	**7.58**	**0.09**	**5.54**	**146**	**150**	**0.027**	**0.023**	**0.054**	**0.004**	**0.028**	**0.357**	**0.081**
A3	mw	**7.36**	**0.51**	**5.69**	**2759**	**1323**	**0.03**	**0.03**	**0.07**	**0**	**0.05**	**1.124**	**0.07**
A5	sp	**7.22**	**0.11**	**4.2**	**269**	**183**	**0.027**	**0.028**	**0.057**	**0**	**0.023**	**0.009**	**0.028**
A7	mw	**4.04**	**2.82**	**4.42**	**3962**	**3615**	**0.043**	**0.084**	**0.19**	**0.233**	**0.216**	**19.36**	**1918**
A8	rw	**3.88**	**0.85**	**1.77**	**1076**	**1314**	**0.04**	**0.055**	**0.053**	**0.338**	**0.13**	**11.05**	**1175**
A10	rw	**7.91**	**0.06**	**4.27**	**116**	**51.28**	**0.027**	**0.032**	**0.024**	**0.001**	**0.019**	**0.011**	**0.072**
A11	we	**7.02**	**0.04**	**10.55**	**158**	**59.44**	**0.022**	**0.029**	**0.038**	**0.001**	**0.017**	**0.008**	**0.039**
A12	sp	**7.75**	**0.15**	**2.69**	**56**	**64.69**	**0.027**	**0.027**	**0.034**	**0.001**	**0.021**	**0.009**	**0.062**
A13	rw	**8.11**	**0.14**	**9.44**	**65**	**57.64**	**0.027**	**0.023**	**0.036**	**0.009**	**0.022**	**0.033**	**0.049**
A16	rw	**5.61**	**0**	**4.07**	**547**	**534**	**0.045**	**0.033**	**0.074**	**0.06**	**0.083**	**38.59**	**481**
A17	sp	**7.45**	**0.05**	**2.13**	**21**	**14.26**	**0.028**	**0.029**	**0.034**	**0.001**	**0.019**	**0.017**	**0.03**
A18	mw	**7.3**	**1.74**	**2.84**	**189**	**1289**	**0.028**	**0.028**	**0.057**	**0.001**	**0.028**	**0.039**	**0.282**
A19	mw	**3.83**	**0**	**41.11**	**2298**	**27679**	**0.091**	**0.064**	**0.148**	**0.248**	**0.327**	**63.59**	**2610**
A20	rw	**7.06**	**0.09**	**1.4**	**57**	**40.61**	**0.029**	**0.047**	**0.027**	**0.001**	**0.02**	**0.099**	**0.727**
A21	mw	**4.16**	**14.78**	**56.4**	**2519**	**21063**	**0.133**	**0.083**	**0.266**	**0.028**	**0.481**	**108**	**3098**
A22	mw	**4.55**	**24.35**	**82.1**	**2308**	**67541**	**0.143**	**0.109**	**0.26**	**0.024**	**0.461**	**108**	**3041**
A23	mw	**7.67**	**0.04**	**1.72**	**68**	**286**	**0.03**	**0.05**	**0.063**	**0**	**0.019**	**0.203**	**2.641**
A24	rw	**7.77**	**0.07**	**1.48**	**14**	**17.47**	**0.029**	**0.038**	**0.045**	**0**	**0.014**	**0.023**	**0.404**
A25	we	**7.55**	**0.06**	**3.01**	**221**	**47.58**	**0.023**	**0.034**	**0.054**	**0**	**0.017**	**0.035**	**0.123**
A26	rw	**7.39**	**0.17**	**1.91**	**217**	**547**	**0.029**	**0.023**	**0.063**	**0.004**	**0.084**	**6.15**	**533**
A27	we	**7.37**	**0.1**	**23.39**	**165**	**820**	**0.032**	**0.03**	**0.059**	**0**	**0.035**	**0.18**	**0.056**
A28	sp	**7.64**	**0.32**	**5.13**	**62**	**26.47**	**0.029**	**0.028**	**0.051**	**0.001**	**0.024**	**0.016**	**0.096**
A29	mw	**5.91**	**3.13**	**8.47**	**1127**	**4220**	**0.029**	**0.043**	**0.111**	**0.004**	**0.336**	**45.00**	**2523**
A30	rw	**7.59**	**0.19**	**1.69**	**390**	**284**	**0.031**	**0.042**	**0.043**	**0.001**	**0.032**	**0.214**	**0.405**

rw – running water; mw – mine water; sp – spring; we – domestic well; marked values exceed the quality standards: green – 98/83/CE for drinking water; orange – NTPA-013/2002 for surface water; purple – HG 882/2013 for waste water

In groundwater, Zn and Fe concentrations do not exceed the standard values for drinking water. The Pb and Ni concentrations exceed these standards by less than 1 % and can be considered borderline values. On the other hand, Cd concentration is higher than the standard value for drinking water and could be a consequence of groundwater contamination by mine water. However, if we take in consideration the fact that Cd concentrations and their seasonal variation are very similar in all sources of groundwater and in unaffected surface water, then the elevated Cd concentration could be due to geochemical background which, due to the presence of sulphide ores in the area, is high. All springs and wells show much higher values of NO_3 concentration than the standard for drinking water (98/83/EC) allows. The high values of NO_3 were recorded in March and April. In the Techereu well (A27) the concentration of SO_4 is also above the standard value for drinking water. The Larga spring (A5), the Zlatna spring (A17) as well as the Techereu well (A27) and the Techereu spring (A28) have Ni and Pb concentrations slightly higher than the standard values. All springs and wells show more elevated Cd concentrations than the standard permits.

The mine waters from the Haneș 2 and the Valea Babei (A21 and A22) adits discharge the largest amount of contaminants. They are acid waters and in their composition F, SO_4, NO_3, Pb, Zn and Fe exceed the standard concentrations for waste water (HG 188/2002). The mine water from the 23 August adit (A23), with the exception of NO_3 concentrations, is within the standard limits for waste water.

2.3.3.2 *Impact on aquatic life*

Total aerobic count (TC) is an indicator for the microbial status of the environment. The analysis of water samples from Zlatna mining area shows a lower bacterial load in the mine drainage than in groundwaters and surface waters, more than half of drainage waters being incompatible with the survival and multiplication of the aerobic mesophilic bacteria. The drainage waters from the Haneş Mine (A19) and from the Haneş 2 adit were sterile in all seasons. The Valea Babei adit (A22) and the Podul Ionului adit (A29) showed very few viable microorganisms (1-2 CFU/ mL). The IPEG (A3), Larga (A7) and Toţi Sfinţii (A18) adits showed a low concentration of aerobic bacteria (0-99 CFU/mL), but the 23 August adit (A23) indicated medium microbial growth (100-199 CFU/mL), similar with the surface waters.
In most of the surface waters, the total aerobic count had medium values, showing concentrations of more than 100 CFU/mL, with most values being around 200 CFU/mL. But in the Ampoi stream upstream (A1), the Ardeu stream upstream (A24) and the Techereu stream A30 and A26) the total count of aerobic bacteria was high, more than 300 CFU/mL. The highest concentration was recorded in the Haneş stream (A20) (522 CFU/mL). The lowest concentrations of aerobic bacteria, less than 100 CFU/mL, were observed in the running waters (Larga Creek (A8) and Ardeu stream (A16)) when samples were taken downstream from the mine drainage.

The groundwater (draw-wells and springs) in the mining areas indicated reduced concentrations of aerobic bacteria with less than 100 CFU/mL in the Larga spring (A5), medium values (100-200 CFU/mL) in the Trâmpoiele draw-well (A11), as well as high concentrations (>300 CFU/mL) in the draw-wells from the Techereu (A27) and Ardeu streams (A25) (Table 2.3.2. and Figure 2.3.3).

Fungi show a similar pattern in the three water types from Zlatna mining area, but with lower concentrations of viable cells than bacteria. Thus, the mine drainage comprised the smallest number of fungi, with a maximum (2 – 30 CFU/mL) observed in Toţi Sfinţii and 23 August adits. The acid waters of four mines (Podul Ionului adit, Valea Babei adit, Haneş Mine, and Haneş 2 adit) did not contain any viable fungi in any season, whereas the IPEG and Larga adits showed only one colony growth on the nutrient medium, when the waters were sampled in the warm seasons. In winter, the mine drainage did not contain any viable yeast or mold. The running waters show the highest number of fungi (>300 CFU/mL), in contrast with the groundwaters, which had a medium content in yeast and mold (1-176 CFU/mL). The seasonal influence also determined fluctuation in fungi content in the surface water, groundwater, and mine drainage, with a maximum concentration of viable fungi in spring and summer and a minimum in winter (Table 2.3.3 and Figure 2.3.4).

The diversity and types of aquatic invertebrates are very useful for evaluating water quality. The diversity of aquatic organisms is highest at intermediate values of pH and O2 (pH about neutral, and O2 about 8 mg/L). Mine waters heavily polluted by acid drainage and metal contamination are distinguished by the absence of any signs of life, both microorganisms and invertebrates. This category of deadly waters includes those observed at the Haneş Mine and the Haneş 2 adit. The absence of aquatic invertebrates was also observed in the surface waters of the Ardeu stream downstream of the Haneş Mine, as well as in the Techereu stream downstream of the Podul Ionului adit, although these running waters had an elevated microbial content. In the IPEG, Valea Babei and Podul Ionului mine drainage very few invertebrates (less than five) were recorded, and only in the warm seasons. The last category of mine waters, represented by the Larga, 23 August and Toţi Sfinţii adits were distinguished by a large number of aquatic invertebrates, like the surface waters. The running surface waters generally had less abundant invertebrate fauna, similar to the mine waters' mean invertebrate counts. Meanwhile, groundwaters showed the most abundant aquatic fauna, with a total of 150 invertebrates recorded in the summer (Table 2.3 3 and Figure 2.3.5).

In terms of taxonomic diversity, 13 groups of aquatic invertebrates were found in the warm seasons, compared to 9 groups recorded in winter. In surface waters the most abundant invertebrates were insects, represented by *Diptera* larvae, however in groundwaters copepods (*Copepoda* and *Harpacticoida*) were the most abundant (Figure 2.3.6). The aquatic invertebrate community provides a significant indication of the health of water ecosystems. A basic invertebrate community indicator of clean streams is the total number of taxa in the groups *Ephemeroptera, Plecoptera*, and *Trichoptera* (EPT), with more species commonly found in cleaner waters (Dodds, 2002). *Ephemeroptera*, *Plecoptera*, and *Trichoptera* were found in all seasons in all types of waters: in running surface waters (Ardeu stream downstream, Haneş stream, Ampoi river downstream and upstream, Larga Creek, Trâmpoaiele stream downstream and upstream, and Techereu stream upstream), in mine waters (Toţi Sfinţii, 23 August, and Larga adits), as well as in groundwaters (Larga spring and Ardeu well).

Comparing the three types of analysed waters, the mine drainage, groundwaters and running surface waters, it is evident that surface waters had the highest concentration of microorganisms, followed by groundwaters. The mine drainage was either sterile or showed low concentration of bacteria and fungi (Figure 2.3.7). In all types of analysed waters the aerobic bacteria were higher than fungi. The running surface waters exhibited a reduced concentration of bacteria and fungi downstream, compared to those situated by the main drainage upstream. A constant exception was the Techereu Valley, where the distance from the two sampling points was greater (about 6 km), including a village, which contributed to the increase of microorganism content.

Table 2.3.3 Total count of the microorganisms and invertebrates monitored in mining drainage, groundwaters and running surface waters at the Zlatna mining area

Source	**Type**	**Total count of aerobic bacteria**				**Yeast&Molds**				**Aquatic invertebrates**		
		(CFU/48 ore/mL)				(CFU/96 ore/mL)				(no of taxa)		
		May	Aug.	Oct.	Dec.	May	Aug.	Oct.	Dec.	Aug.	Oct.	Dec.
A1 Ampoi upstream	rw	155	>300	110	246	4	34	2	1	1	3	1
A2 Trâmpoiele downstream	rw	138	157	114	2	44	13	1	1	3	0	2
A3 IPEG adit	mw	98	4	22	3	0	1	0	0	4	0	0
A5 Larga spring	gw	75	13	76	66	91	5	3	1	0	13	1
A7 Larga adit	mw	0	4	17	0	1	1	0	0	16	29	0
A8 Larga creek	rw	3	22	5	49	0	2	3	0	4	29	4
A10 Trâmpoiele stream upstream	rw	59	189	121	138	12	>300	9	11	10	1	0
A11 Trâmpoiele well	gw	28	Sec	152	36	6	Sec	1	3	sec	sec	1
A13 Ampoi downstream	rw	115	174	81	133	39	26	7	7	4	0	2
A16 Ardeu downstream	rw	99	55	35	90	3	22	0	4	2	2	0
A18 Toți Sfinții adit	mw	7	46	20	63	2	20	8	0	4	4	2
A19 Haneș mine	mw	0		0	0	0	0	0	0	0	0	0
A20 Haneș stream	rw	522	>300	123	67	46	37	3	30	11	4	8
A21 Haneș 2 adit	mw	0	0		0	0	0		0	0	-	0
A22 Valea Babei adit	mw	0	1		0	0	0		0	4	-	0
A23 23 August adit	mw	167	63	28	9	30	13	0	0	7	13	3
A24 Ardeu upstream	rw	108	>300	100	73	30	>300	3	1	10	2	0
A25 Ardeu well	well	68	>300		53	33	17		2	88	-	2

A26 Techereu downstream	rw	-	>300	75	279	67	>300	3	0	0	0	0
A27 Techereu well	well	352	93	89	99	176	1	9	5	62	4	9
A29 Podul Ionului adit	mw	0	0	2	1	0	0	0	0	0	1	0
A30 Techereu upstream	rw	65	227	20	0	11	1	0	0	1	6	0
Total no. of individuals		**2059**	**2553**	**1190**	**1407**	**595**	**1096**	**52**	**66**	**231**	**111**	**36**

rw – running water; mw – mine water; gw – groundwater (springs and wells)

The seasonal variation of aquatic organisms reveals the fundamental role of temperature for the microorganism viability and growth, but also for invertebrate development. The most favourable seasons were spring and summer, as demonstrated by a greater number of organisms sampled from the mine drainage and surface waters. In groundwaters, the seasonal influence on organism populations was not so evident for aerobic bacteria, as their concentration was rather uniform. However, the fungi concentration and the aquatic invertebrates followed the seasonal pattern of surface water, being more abundant and diverse in the warm seasons. Thus, the summer samples indicated the highest number of aquatic invertebrates (228 individuals), followed by autumn (109 individuals), while during the winter season, the fewest animals were recorded (36 individuals).

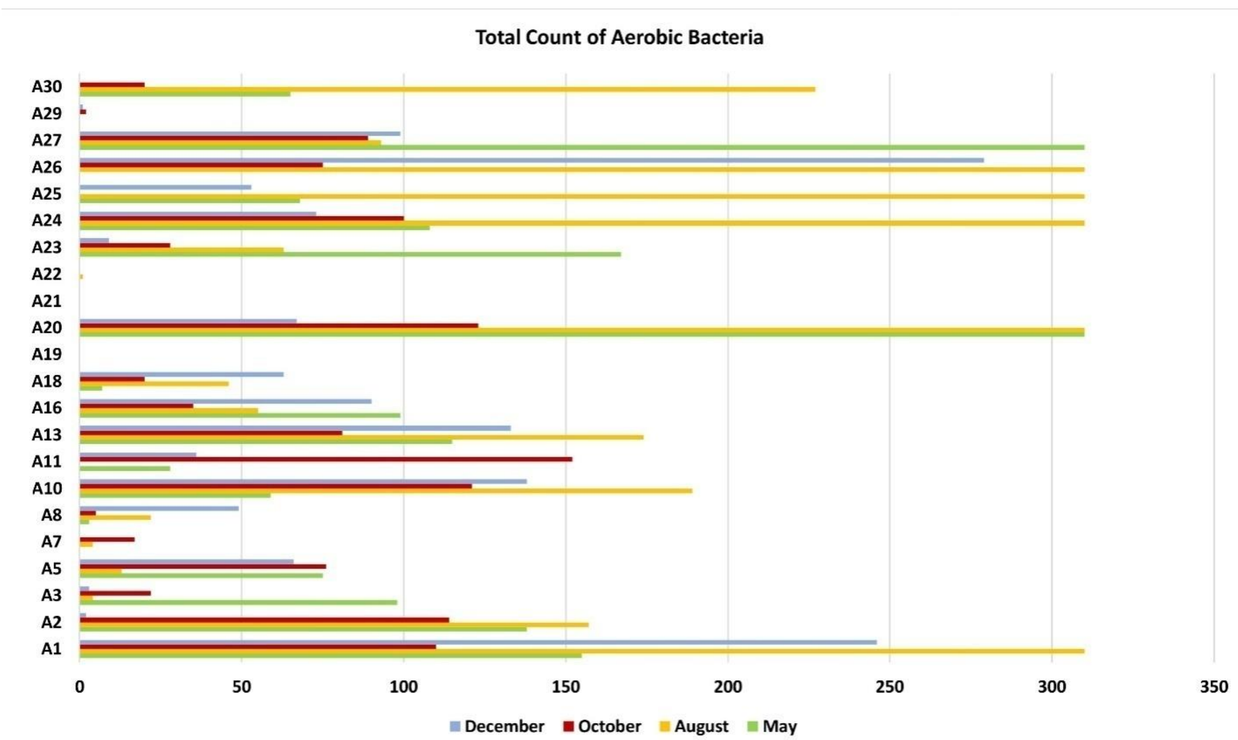


Figure 2.3.3 Total count of aerobic bacteria (CFU/mL) in mine water, groundwater and running water at Zlatna mining area

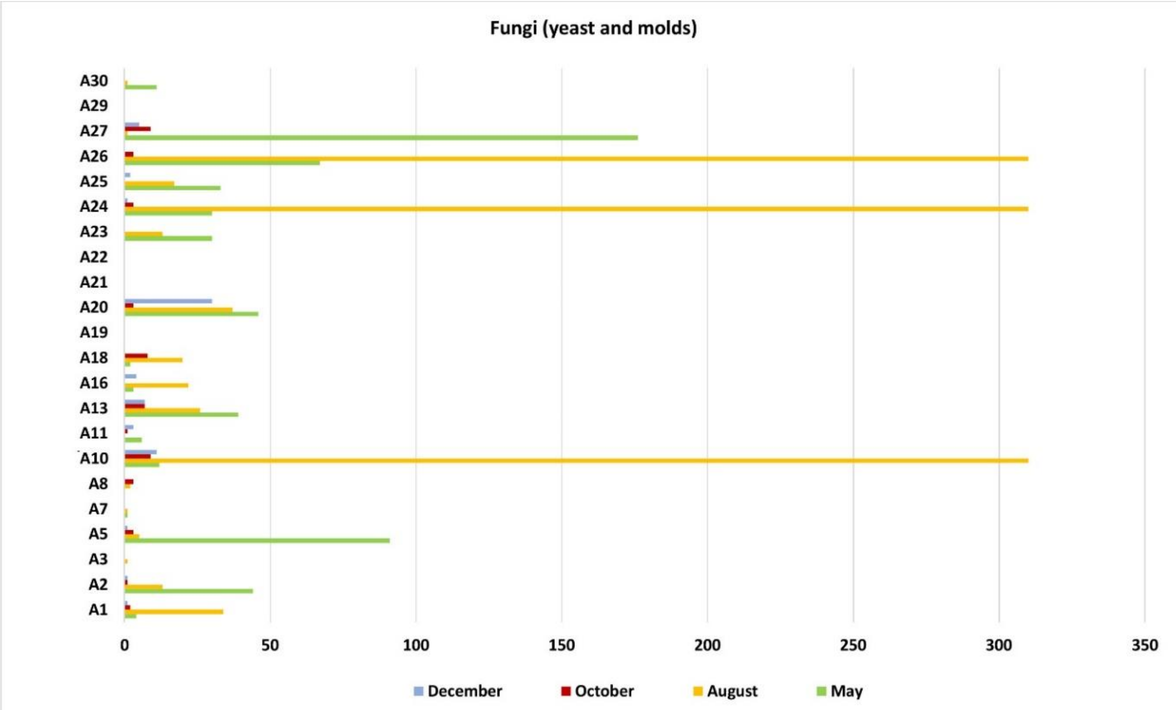


Figure 2.3.4 Fungi (yeast & molds) in mine water, groundwater and running water at Zlatna mining area (CFU/mL)

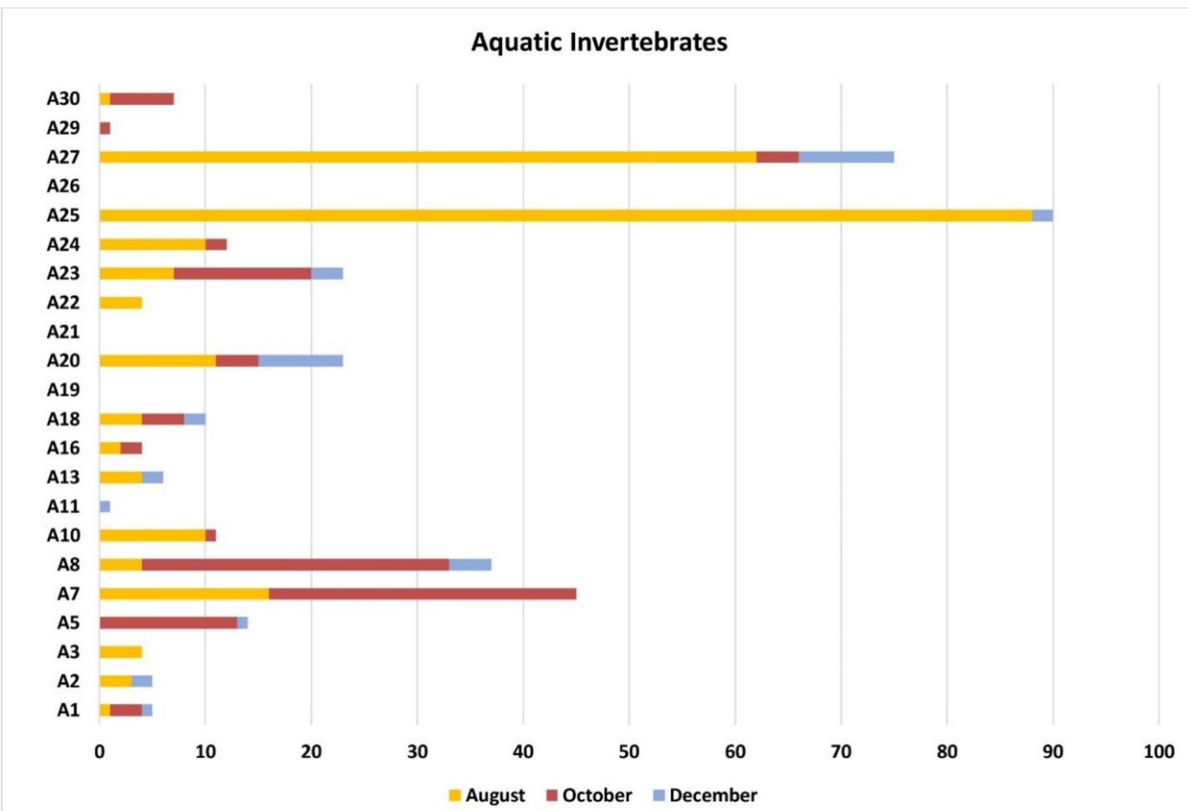


Figure 2.3.5 Aquatic invertebrates (number of individuals) in mine water, groundwater and running water at Zlatna mining area

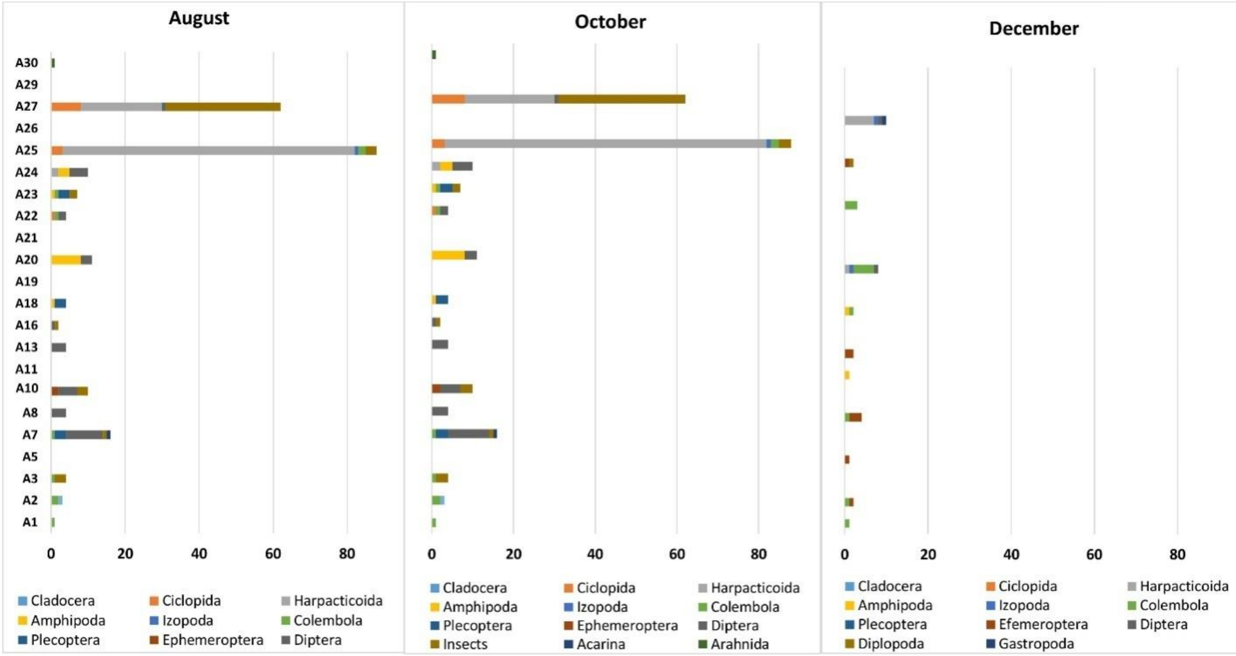


Figure 2.3.6 Taxonomic groups of aquatic invertebrates in mine water, groundwater and running water at Zlatna mining area

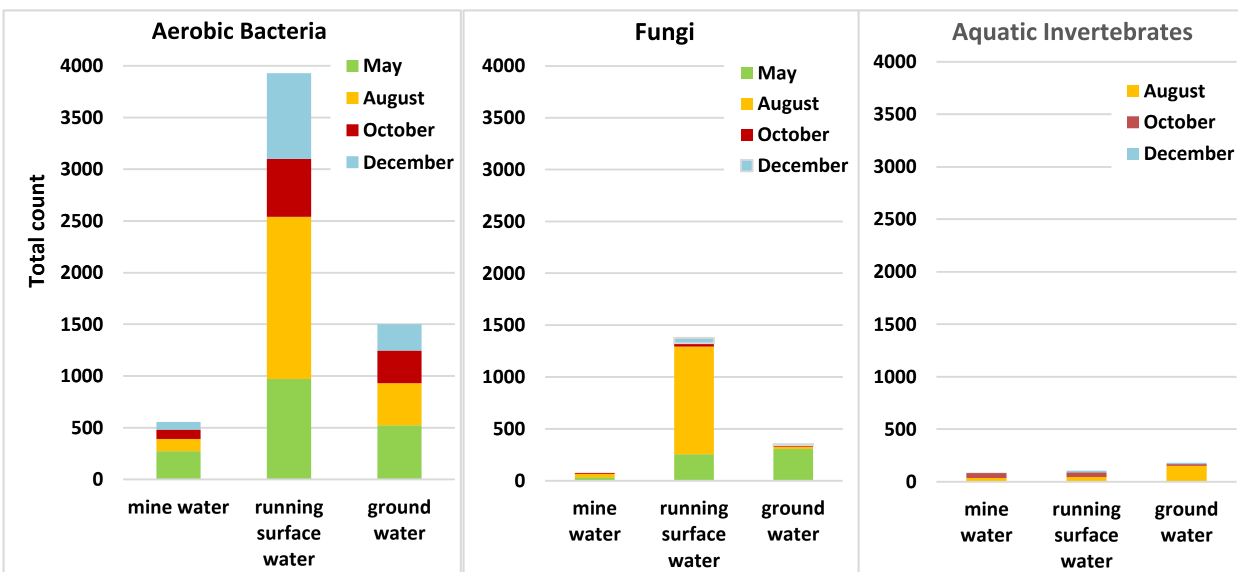


Figure 2.3.7 The aquatic organisms seasonally monitored in mine water, groundwater and running water at Zlatna mining area

2.3.3.3 *Issues of concern*

The elevated concentration of SO_4, Zn and Fe in mine waters are the main issues of concern at Zlatna site. The most affected watercourses are the Ardeu stream and the Techereu stream, downstream of mine water discharge. Another concern is related to the high concentrations of NO_3 that was recorded in all water sources in March and April. However, this could be due to organic matter or could be a consequence of using fertilizers in agriculture and is not necessarily directly related to mining contamination. The analytical results for the entire year would allow the source of NO_3 to be better constrained.

In groundwater, the concentrations of Zn, Fe, Pb and Ni are within the standard values for drinking water. In contrast, the Cd concentrations are exceeding the standard and could be a consequence of groundwater contamination by mine water. However, when taking into account the seasonal variation of the concentrations in all sources of groundwater and in unaffected surface water, the elevated Cd concentration might also be originating naturally from the sulphide ores in the area. Supplementary isotopic analysis of Cd and Pb is needed for accurate determination of the source of these elements in groundwater, as well as in surface water.

In conclusion, the results of the biomonitoring of mine drainage, groundwater and surface water from the Zlatna mining area showed:
(1) The mine drainage was inadequate for supporting life, or exhibited a low concentration of mesophilic aerobic bacteria, yeast and molds, and aquatic invertebrates;
(2) The running surface water had the highest concentration of microorganisms, the upstream areas being richer in aquatic organisms than those located downstream the mines;
(3) The groundwater showed an intermediate pattern of microorganism content, but domestic wells were the richest in aquatic invertebrates.
Furthermore, in all types of analysed waters the aerobic bacteria were higher than fungi.

As former mining areas still represent the main contaminant factor that affects the environment in the vicinity of Zlatna area, monitoring of water quality and aquatic ecosystems is still necessary. Moreover, designing appropriate measures to diminish the contaminant release that could harm human health or the environment should be applied.

3 Methods applicability. General evaluation and awareness

Aquatic organisms represent a significant ecological indicator for assessing the physical and chemical changes and anthropogenic impact on water quality. Beside this, aquatic organisms that spend most of their life cycle in water are extremely important in maintaining the health of aquatic ecosystems and also represent a very useful instrument for rapidly assessing the contaminants risk. This affects also the recreational use of the water systems, as the state of the lakes and rivers are often important not only to locals but also to tourism. Clear differences were observed between communities of all aquatic organisms living in water with varying levels of contamination at all three case study sites, in Finland and Romania. The obtained data also revealed that the type of contamination (organic or inorganic) is a significant factor for the existence of certain communities. Furthermore, as the study showed that aquatic organisms indicate well both short (Kittilä site) and long-term (Romania and Zlatna sites) impacts of mining, ecological monitoring is a suitable tool to assess the environmental impact of mining on aquatic ecosystems.

However, as the aquatic ecosystems are controlled by various geological, physical, chemical and biological factors and identifying the exact source is not easy, a comprehensive environmental impact assessment requires further complex investigations. Moreover, as the ecological surveillance reports of Kittilä site showed, studies relying only on few organisms do not indicated the overall state of the aquatic ecosystems but may give too optimistic picture of the state. Thus the response given by the aquatic organisms indicates a certain water quality and together with practical water monitoring and the results of studies in other fields direct to a source of contamination.

One of the aims of this study was to evaluate the usefulness of ecological risk assessment tools in supporting water management strategies. Ecological risk assessment tools appeared to be useful in estimating the safe discharge levels of different elements in a certain environment. However, ecological risk assessment alone would be too simple to model such a complex system as mine environment, and more defined modelling of the water flows and reactions could give more robust results. Moreover, since there was not enough toxicity data available to allocate PNEC to a specific environment, PNECs was not seen to be a very practical tool for setting environmental permit limits without conducting toxicological tests.

This type of a modelling method could be best when applying an Environmental Impact Assessment (EIA), which is the foundation of setting the environmental permit. Furthermore, ecological risk assessment would help monitoring the efficiency of the water treatment and management methods during mine operations. However, when implementing site-specific guideline concentrations, for example by the use of SSD, would require more precise information on *e.g*. the river characteristics, toxicity data and geochemical modelling of the behaviour of different mine water constituents in the environment after discharge.

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