Impact of dissolved metals(oids) in surface water quality of the Almadén mine district

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Abstract: - Waters from 10 sampling points in the streams of the Almadén mine district were collected in the period 2010-2013 in order to study the potential impact caused by the input of Potentially Toxic or Harmful Elements (PTHE) from the decommissioned mines of the district in the freshwater environment and then the potential risk for the aquatic organism and to the resident population. Dissolved metals(oids) in surface waters were investigated byby different analytical techniques and the results were statistical evaluated using Principal Component Analysis (PCA). The assessment of the potential risk for biota and humans using the Environmental Risk Index (ERI) and the Hazard quotient (HQ) has been made. Considering the different physico-chemical data together with their statistical multivariate treatment, the district displayed a significant metals load where the main PTHE (As, Co, Hg and Mn) may be attributed to the decommissioned mine sources. The amount of dissolved metals detected in the surface waters showed an ERI value of 11.64, corresponding to a poor environment where different metals(oids) may be transferred to the aquatic organisms. On the other hand, the assessment of the same metal load from the viewpoint of human consumption showed that the contents of Hg and As are potentially hazardous for the resident populations, where they can cause adverse health effects in sensitive populations such as children or babies.

Key-Words: - Decommissioned mining, mercury, surface waters, pollution, risk assessment, Almadén district.

1 Introduction

Heavy metals and metalloids are a great concern in the environmental sciences due to their special physico-chemical characteristics. These elements are not biodegradables; they may persist in the medium for a long time and can be accumulated in plants and food webs producing a potential risk to various ecosystem. Metals and metalloids are introduced into the environment from different sources: natural (volcanism, bedrocks weathering) and anthropogenic (traffic, industry, agriculture and particularly mining and mineral processing which have dominant influences on the biogeochemical cycles of trace metals).

One of the most important environmental compartments affected by the input of these elements is the surface waters, where pollution as consequence of anthropogenic activities is an environmental concern worldwide due to the importance of this system to the human live and activities [1-3]. The accumulation of heavy metals in aquatic ecosystems can lead to hazards on human and wildlife as demonstrated different studies linking the intake of polluted water [4-5]. Within this problematic, surface waters affected by inputs from mine activities are very important as consequence of the great amounts of potential toxic elements release from mine waste and discharged into the environment each year from mine sites. An especially concern within mine sites are the decommissioned old mines which occasionally can suppose a significant environmental risk related to pollutants that input into the environment from the weathering of the old waste materials.

The riverine system on which we focused this study, the Valdeazogues River Basin, is a large watershed, geologically and physiographically diverse with a mixture of land cover and land use types. Another significant task of the Valdeazogues River Basin, is the presence of the Almadén mine district, which is the the most important Hg mine in the world, and one of the important historic mining districts at a world scale..

The district was historically affected by the massive mining activities developed over more than two millennia, where more than 52 Hg decommissioned mines [6] are located together with a not determined number of small polymetallic

deposits (Pb, Zn and Ag) scattered in the region [7-9], which have had some extractive activities in a recent past . Together with the mercury deposits, other polymetallic deposits (Pb, Zn and Ag) have had some extractive activities in the area in a recent past. As consequence, the aquatic system in the district has been significantly affected by the inputs from decommissioned mines in the area [10-13]. Other studies from the area reported the presence of pollutants in the environment but they have not focused the objectives of the studies in the assessment of the potential risk to biota. Therefore, the goal of this study is intended as a first approach to assess the potential risk for biota and humans associated to the affection suffered by the aquatic system to serve for develop water quality conservation strategies for the area.

Fig.1-Study area and sampling points
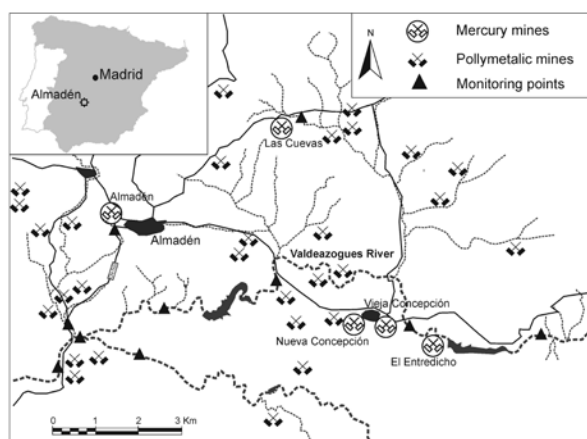
2 Materials and methods
2.1 Site description
The Valdeazogues watershed is located in Ciudad Real country (central Spain). It covers a surface of 1,213 km^2 with varied geology and land uses. The main river is the Valdeazogues that flow below different lithological materials 84.3 Km to its confluence with the Guadalmez River. In the lower part of their watershed, this is located the Almadén mine district well known as the largest mercury district in the world [14] which covers one fifth part of the total watershed surface. The hydrologic network of the district is compound by four tributary rivers (Gargantiel, Tamujar, Azogado and Alcudia) together with the Valdeazogues River which gives its name to the basin. The area is affected by a semiarid climate where the annual precipitation rate falls between 500 and 525 mm, the average

temperature is about 15–16 ºC and the average evapotranspiration amounts to 963 mm [15].

2.2 Water sampling and analytical methods
The surface water samples were collected in 10 sites distributed according to the potential contaminant sources in case of the tributary streams and following the stream flow direction of the Valdeazogues River. Nine sampling campaigns were carried out in each site in a 4-year period (2010-2013). Sampling campaigns were carried out in different months of the years including different weather periods and hydrological regimens of the streams. In each site, different types of water samples were taken to analyze dissolved metals(oids). To analysis metals(oids), samples were collected in a depth of 15 cm by means of disposables syringes previously rinsed with the site water to condition them. After that, waters were filtered at the sampling point through 0.45µm Millipore Teflon filters and storages in pre-cleaned high density polyethylene (HDPE) containers. To stabilize dissolved metals(oids) in suspension, samples were acidified to pH< 2 using suprapure nitric acid (69% Trace metals analysis HIPERPUR®), and they were immediately stored refrigerated (±4 °C) to the laboratory.

The subsamples for major elements (Ca^{2+}, Mg^{2+}, HCO^{3-}, K^+, Na^+, Cl^- and SO_4^{2-}) determinations were collected directly by the containers on the stream at the same depth as samples for metals(oids) analysis have been collected. Pre-cleaned containers were conditioned following the same methodology that container for metals(oids) analysis. After collected, the samples were stored refrigerated (±4 °C) without the addition of stabilize agents according to our laboratory recommendations. Also, together with the water samples, some physico-chemical parameters were measured in field using a CTD multiprobe YSI 556 equipped with pH, electric conductivity (EC) and redox potential (Eh) probes.

Dissolved metals(oids) in the water samples were determined using an Inductively Coupled Plasma Mass Spectrometer (ICP-MS) Agilent 7500 type quadrupole with collision cells and equipment with a meinhard concentric flow nebulizer. The gases used were Ar 99.999% purity as plasma gas and He 99.999% purity as collision gas. Analyses were performed in according to EPA method 200.8 [16]. Major elements were determined using an Ion Chromatography 883 Basic IC METROHM. All elements were analyzed in batches, which included a procedural blank and a standard reference material, each calibration curve was evaluated by analyses of quality control standards before, during and after the analyses of a set of samples.

2.3 Statistical analysis

Principal component analysis (PCA) is a powerful pattern recognition tool that attempts to explain the variance of a large dataset of intercorrelated variables. The methodology was employed to find and interpret the structure of the underlying data set through a reduced new smaller set of independent variables (principal components, PCs), arranged in decreasing order of importance. PC provides information on the most meaningful parameters, which describe the whole data set while affording data reduction without losing much original information. PCA with Varimax rotation was conducted for extracting and deriving factors respectively and those PCs with eigenvalue >1 was considered in according to of Kaiser Criterion. This method allows identify the different groups of elements and parameters that correlate and thus can be considered as having a similar or related behavior and common origin. Therefore, this statistical method was applied to the chemical results. SPSS software for MS Windows V17 and MS Excel 2012 were the software used to processing the dataset.

2.4 Biota and human health risk assessment

Exposure of Biota that living into aquatic ecosystem could occur via three main pathways including direct ingestion, respiration through gills or other respiratory organs, and dermal absorption through skin exposures. Quality indexes can be used as direct forms to obtain quickly results that allow knowing the potential risk associated to potential harmful elements dissolved into waters. In this case, we have used the Environmental Risk Index (ERI) [17-20] that describes the possible occurrence of adverse effects from environmental exposure to one or more agents of ecological stress. The ERI allows a numerical value predicted by the probability of occurrence of a negative impact on the environment by the average concentrations of specific pollutants [21]. In this case, the mathematical index was calculated according to the equations from [17] using as reference values the levels reported by the Canadian Environmental Quality Guidelines for the protection of the aquatic life.

$$ERI = \sum_{1}^{n} Q_{Eri}$$
$$Q_{Eri} = \frac{AC_i}{RC_i} - 1$$

where, AC_i, RC_i and Q_i represent the concentration of the element in water ($\mu g\ l^{-1}$), legal risk limit to the element in waters ($\mu g\ l^{-1}$) and the environmental risk index ratio of the element. The ERI values were classified into seven categories: IRE≤0 no risk, 0<IRE≤1 very low risk, 1<IRE≤3 low risk, 3<IRE≤5 medium risk, 5<IRE≤10 high risk, 10<IRE≤15 very high risk and IRE>15 extremely risk.

In case of human exposure, only has been considered the intake by ingestion due to this is the main pathway for drinking water. The selected methods have been the Average Daily Dose by ingestion (ADD) indices and the Hazard quotient (HQ) indices, calculated according to the modified equations from the USEPA (2004):

$$ADD_{ingestion} = \frac{C_w\ x\ IR\ x\ ABS_g\ x\ EF\ x\ ED}{BW\ x\ AT}$$
$$HQI = \frac{ADD_{ingestion}}{RfD}$$

where $ADD_{ingestion}$ (Average Daily Dose by ingestion) unit is $\mu g\ kg^{-1}$/day; IR (ingestion rate) unit is $l\ day^{-1}$; ABSg, gastrointestinal absorption factor (dimensionless); EF, exposure frequency, unit in $days\ year^{-1}$; ED, exposure duration, unit in years; BW, body weight, unit in kg; and AT, averaging time (days). ABSg values originated from [22-24] and body factors, exposures and ingestions from the USEPA Exposure Factors Handbook [25]. The exposed population is assumed to be safe when HQ<1 [20].

3 Results and discussion

The results of the monthly water characteristics (mean and standard deviation) are presented in Fig. 2 and Table 1.

During the studied period, flow rates showed a seasonality variation (0 to 1.31 $m^3\ s^{-1}$) in area governed by the Mediterranean weather. Maximum flow rates were recorded in autumn and winter months and minimum in dry periods where the flow rates trend to null as consequence of the drought and the absence of important groundwater inputs. In the physicochemical parameters, pH did not show an important variation in their values, ranging from neutral to alkaline values (6.7 to 7.58). Moreover, Electric conductivity (EC) and redox potential (Eh) displayed a significant oscillation range their values between 420.5 to 984.1 $\mu S\ cm^{-1}$ and 340.7 to 631.6 mV respectively.

Figure 2: Boxplots of dissolved metals(oids) concentration in comparison with flow rates along the sampling period. Data in µg l⁻¹
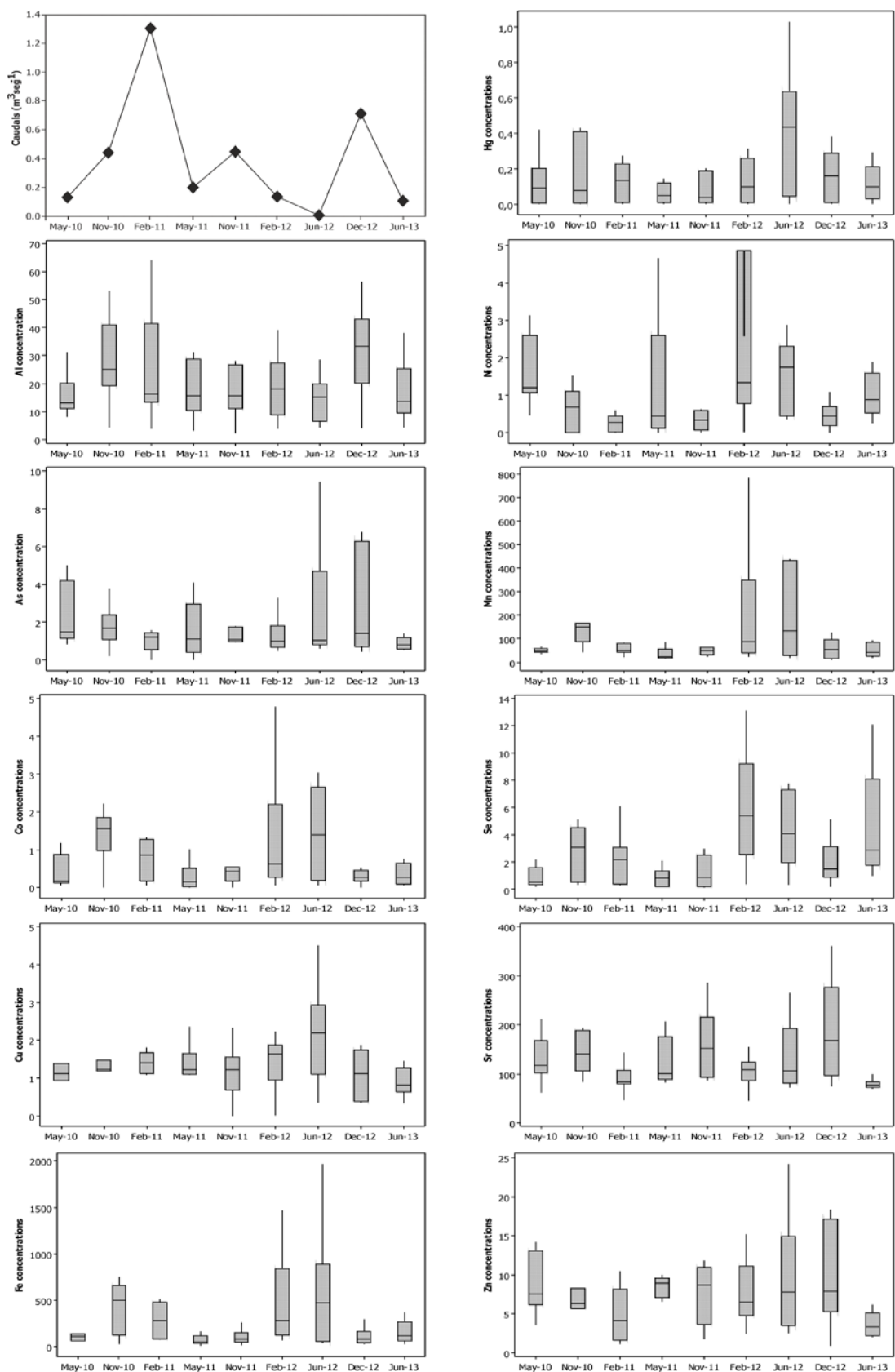
Table 1: Dissolved anions, cations and physicochemical parameters in the Valdeazogues watershed (units: cations and anions in mg l^{-1}, Electric conductivity in µS cm^{-1}, and Eh in mV).

	may-10		nov-10		feb-11		may-11		nov-11		feb-12		jun-12		dic-12		jun-13	
	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
HCO_3^-	123.9	56.9	140.5	55.9	96.2	42.4	101.0	50.8	105.2	48.2	169.9	72.1	197.6	100.1	98.4	54.9	111.3	45.8
Ca^{2+}	31.0	14.3	45.4	25.2	25.70	11.5	24.5	12.5	30.9	18.3	72.1	71.0	61.4	68.1	27.5	18.8	43.5	38.8
Cl^-	18.1	4.7	23.9	16.2	16.18	11.6	18.5	12.8	23.8	17.8	44.9	8.8	31.0	14.8	21.4	11.3	16.8	6.4
Mg^{2+}	21.8	18.2	25.2	13.0	13.79	8.5	18.1	8.1	19.6	8.0	77.3	126.3	50.2	60.5	19.0	11.7	31.2	34.7
K^+	3.1	2.9	4.3	3.2	2.20	1.8	3.2	2.9	3.6	3.5	5.4	4.1	6.0	5.8	2.4	1.7	2.5	2.4
Na^+	24.6	18.0	20.4	9.6	14.98	11.2	14.5	10.7	21.2	16.0	37.8	16.9	32.6	21.5	17.7	10.7	22.6	18.5
SO_4^{2-}	78.48	92.2	103.2	62.4	47.88	29.1	46.8	23.7	73.8	49.9	407.3	750.6	256.7	427.2	61.1	56.2	151.1	218.1
pH	7.5	0.3	7.5	0.2	7.58	0.3	7.5	0.5	7.3	0.3	6.7	0.5	7.2	0.4	7.3	0.2	7.4	0.3
EC	568.6	364.5	542.5	369.5	471.70	346.3	526.9	296.1	465.2	194.5	726.3	729.8	984.9	792.9	420.4	202.3	592.2	410.4
Eh	422.6	30.3	381.8	24.3	340.69	21.7	381.5	41.9	381.7	20.89	412.9	40.3	631.0	51.9	357.	27.6	420.8	29.9

Magnesic-carbonated facies is predominant in water as consequence of the great amounts of carbonates presents in soil and lithological material in form of secondary carbonates [9] and in case of Mg^{2+}, their enrichment in the area were as consequence of the presence of magnesium lithology materials related basic volcanic processes suffered in the area. Within the group on major elements, SO_4^{2-}, despite not being the most abundant anion, presented the high variability range its values between 46.7 to 407.3 mg l^{-1}. In Almadén, the decommissioned mine activities were related to sulfide ore deposits and the mining liabilities nowadays remains with great amounts of these minerals. The weathering of these areas produces as consequence of biogeochemical processes significant amounts of sulfate salts derived from oxidation processes of the sulfides. As consequence of the easy leachable of these sulfate salts by rain, they flow to the river from the liabilities, altering the normally mineralization load and consequently other system parameter such as the electric conductivity or the potential redox by the physico-chemical reaction produced to hydrolyze theses salt compounds.

In case of metals(oids), results boxplots are showing in the Fig. 2. The average concentrations displayed similar to the major elements a great monthly variability. For example, between the different sampling months mean values of Fe and Mn ranges between 78.87-677.71 µg l^{-1} and 37.75-319.81 µg l^{-1} respectively, reaching vary up to seven times comparing between the low and the high concentrations measured. The rest of metals(oids) varied less than the previous three but showing significate differences between sampling. Analyzing the flow rates with the metals(oids) concentrations, Al, Ni and Sr showed their high concentrations with high flow rates suggesting an input from rain runoffs and in consequence, they may be attributed from soil and lithological materials more than from mine inputs. On the other hand, Fe, Mn, and Hg together with SO_4^{2+} showed high amounts with low or null flow rates. This fact together with the chemical-physical variations that occurred in this months, may be indicative to anthropogenic activities as enrichment source of these key elements from pollution processes, which in our case, attributed to the decommissioned mines since in the area there are no other significant potential contaminant sources.

3.2 Multivariate statistical analysis

The PCA analysis performed on data from surface waters of Almadén mine district showed 5 PCs with eigenvalue higher than 1 for the dataset and justified more than 80% of the total variance (see Table 2). The factors or components which were related to the sources of the elements were extracted after varimax rotation and factor loadings were classified in according to the methodology of [26] as strong (>0.75), moderate (0.75-0.50) and weak (0.50-0.30) respectively corresponding to their absolute loading values.

The first factor (PC1) explaining 37.41% of the total variance, comprised the elements and parameters with high loadings to this factor: Co, Fe, Hg, Mn, Ca^{2+}, Mg^{2+}, SO_4^{2-} and EC. In the second factor (PC2), the elements with high loadings were: Sr, HCO_3^-, K^+ and Na^+ and the factor explained 21.47% of the dataset total variance. The third factor (PC3) explained 12.89% of the total variance, being As and Zn the elements which presented high loadings. The fourth factor includes pH and Ni, and the fifth factor includes Al, but each of these two components explains less than 10% of variance, and may be considered insignificant.

In relation to the first factor, the group of elements seemed to be associated to mine sources. Metals and SO_4^{2-} are key element of mine water inputs into the river. Moreover, this fact is supported by the presence of Ca^{2+} and Mg^{2+} in this group, attributed to processes of acidity buffering of the system related to altering in the water

hydrochemistry produced by physico-chemical reaction that occur in mine waters. The second factor is associated to ions from commonly Earth's crust materials, related in this case to the soils and the geological formation of the basin, therefore from a natural origin. As consequence of the elements with high loading in the third factor, it may be considered another mine component. This factor groups the elements As and Zn related with the other important Pb-Zn-Ag polymetallic ore deposits scattered in the district. The fourth and the fifth factor were considered natural due to their elements and parameters were much related from a natural origin than from mine activities.

Table 2- Total variance explained and component matrix for dataset (principal component analysis with Varimax rotation; the significance of KMO and Bartlett's sphericity test is <0.001).

	PC1	PC2	PC3	PC4	PC5
Al	-0.062	-0.172	0.236	0.057	**0.771**
As	-0.027	0.250	**0.875**	-0.083	0.073
Co	**0.798**	0.099	0.077	0.115	0.212
Cu	0.190	0.538	0.328	-0.053	0.551
Fe	**0.952**	0.090	-0.023	0.158	0.082
Hg	**0.774**	0.311	0.214	-0.180	0.421
Mn	**0.972**	0.081	-0.020	0.132	0.013
Ni	0.340	0.120	0.342	**0.724**	-0.109
Se	0.368	0.165	-0.171	0.520	0.460
Sr	0.036	**0.862**	0.102	-0.001	0.044
Zn	-0.030	0.147	**0.854**	0.147	0.255
Ca^{2+}	**0.870**	0.351	-0.020	0.119	-0.068
Cl^-	0.053	0.765	0.157	0.316	-0.128
HCO_3^-	0.120	**0.932**	0.146	0.033	0.016
K^+	0.010	**0.904**	0.175	0.070	-0.013
Mg^{2+}	**0.941**	0.035	0.016	0.231	-0.053
Na^+	0.276	**0.859**	0.098	0.192	-0.061
SO_4^{2-}	**0.956**	0.056	-0.026	0.189	-0.064
Ph	-0.365	-0.235	0.055	**-0.750**	-0.085
EC	**0.814**	0.393	0.080	0.132	0.135
Eh	0.248	0.478	-0.098	0.356	0.040
Eigenvalues	6.387	4.508	2.707	1.735	1.482
Cumulative Variance (%)	37.415	51.882	64.774	73.035	80.093

Previous paragraphs showed the strong influence of the components related to mine sources in the surface water hydrochemistry. The amount of the total variance explained by the mine factors (50.31%) put in relevance the significate importance of the mine inputs from the decommissioned liabilities in the water quality.

3.3 Environmental and human risk assessment

To assess the potential pollution to biota, as risk reference values for the dissolved metals analyzed, Long term values from the Canadian Environmental Quality Guidelines (CEQG) for the protection of the aquatic life were selected. In this work, analysis have been computed using the values of the concentrations of Al, As, Cu, Fe, Hg, Ni, Se and Zn; Co, Mn and Sr were not computed in this case due to the CEQG do not report risk reference values for these elements.

As shown in Table 3, waters of the mine district present a global value of 11.64 for the ERI index. This value corresponded to a significant metal load impact in waters where a very high risk to transfer dissolved metals to biota exist. This fact presents a stressor environment where biota must live with a continuous intake of pollutants that may produce in long terms severe impacts in the communities degrading the environmental quality, and in consequence affecting other ecosystems that depend of this. By elements, following in order of their risk, were Hg, Se, Fe, Cu, As, Zn, Al and Ni. Hg was the element with higher risk to transfer with an individual value of 5.97. Other elements susceptible to transfer were Se and Fe but much lesser than Hg. Following their individual risk classification, the elements that show the highest risk are Hg, Se, Fe and Cu, which can be related to mercury mines, put in relevance the impact of these areas in the district environment and in consequence on the water quality. The other elements, associated with other types of mineralization (As and Zn) as well as soil and lithological materials (Al and Ni), have the lowest rates of risk at general scale due to their less dispersion only located in small areas near the sources.

For Humans, the risk assessment has been computed according to USEPA (2004) [22]. Table 3 displays the HQ and risk values for the oral pathways for waters relating to adult, child and baby respectively. HQsingestion (hazard quotient by ingestion) of Al, Cu, Fe, Ni, Se, Sr and Zn for adult, child and baby were very smaller than 1, suggesting that these elements posed little hazard and their long term consumption is not likely to generate a significant risk. However, HQsingestion of Co, Hg, Mn and particularly As showed values nearly or up to 1, implying potential health risk from these elements. Within these group, As was the main health concern, especially for babies where HQsingestion>1 suggests that may cause real adverse health by the ingestion of water over a long time. The HQsingestion of this element decreases with the age of the individuals as consequence of the body weight, being adults the group of lowest risk. But, despite of the decreasing in their values, the computed results near to 1 for adults can be considered dangerous as consequence of being a carcinogenic element [27-28] that may produce adverse health effects over a long life time.

Moreover, Hg was the second significant health risk with values near to 1. This element shows a significant character as carcinogenic and teratogen [29] and in consequence, values near the break point should be taken in consideration. In this case, Hg values were lower than As values despite its high dispersion in the aquatic ecosystem [13, 30] due to its low solubility in waters in comparing with As. Similarity to As, HQsingestion of this element decrease with the body weight, but due to its mostly bioaccumulation compared with As and the demonstrated presence in the ecosystem of organic forms such as methylmercury [11], allow to rate this form of mercury as the most potential toxic to human.

Finally HQsingestion of Co and Mn showed a potential risk lower than the presented by Hg and As. Values for both elements were near 1 and in consequence they should be taken into consideration from the viewpoint of a potential human risk. However, both elements do not show the harmful potential of As nor the bioaccumulative properties of Hg.

4 Conclusions

The aquatic ecosystem of Almadén district shows pollution with high amounts of potential harmful elements in dissolved phases. PCA demonstrated that the main source of dissolved toxic elements in the environment may be attributed to the historic mine activities, and they have been abandoned almost ten years ago, environmental liabilities of the several decommissioned mines in the area remains input great amounts of toxic elements to the environment. The high contents in dissolved Hg and other potential harmful elements, mainly As, Co, Mn and Se attributed to these decommissioned mines, may involve worse conditions where dissolved metals may be bioaccumulated in the trophic chain and consequently these elements may be uptaked by the resident population through the aquatic biota consumption. In addition, the assessment of the water quality as supply source for human consumption showed that the dissolved levels recorded for Hg and As may produce severe health effects for a long term consumption for the resident population, particularly for sensitive populations.

References:
[1] Cukrov, N., Cmuk, P., Mlakar, M., & Omanović, D. Spatial distribution of trace metals in the Krka River, Croatia: an example of the self-purification. Chemosphere, Vol. 72, No. 10, 2008, pp. 1559-1566.
[2] Li, Y., Liu, J., Cao, Z., Lin, C., & Yang, Z. Spatial distribution and health risk of heavy metals and polycyclic aromatic hydrocarbons (PAHs) in the water of the Luanhe River Basin, China. Environ. Monit. Assess., Vol. 163, No. 1-4, 2010, pp. 1-13.
[3] Varol, M. Dissolved heavy metal concentrations of the Kralkızı, Dicle and Batman dam reservoirs in the Tigris River basin, Turkey. Chemosphere, Vol. 93, No. 6, 2013, pp. 954-962.
[4] Squadrone, S., Burioli, E., Monaco, G., Koya, M. K., Prearo, M., Gennero, S., ... & Abete, M. C. Human exposure to metals due to consumption of fish from an artificial lake basin close to an active mining area in Katanga (DR Congo). Sci. Tot. Environ., 2016, Doi:10.1016/j.scitotenv.2016.02.167.
[5] Alpers, C. N., Yee, J. L., Ackerman, J. T., Orlando, J. L., Slotton, D. G., Marvin-DiPasquale, M. C. Prediction of fish and sediment mercury in streams using landscape variables and historical mining. Sci. Tot. Environ., 2016, doi:10.1016/j.scitotenv.2016.05.088

Table 3: Canadian Environmental Quality Guidelines Long term levels, Environmental Risk Index, reference dose and Hazard quotient for each element of the waters from the Almadén mine district.

	Biota		Human			
	CEQG Long term ($\mu g\ l^{-1}$)	QERI	RfDingestion ($\mu g/Kg/day$)	HQingestion		
				Adult	Child	Baby
Al	100	0,218	1000	6,65E-04	1,36E-03	2,18E-03
As	5	0,390	0,3	**7,98E-01**	**9,06E-01**	**1,15E+00**
Co	-	-	0,3	7,77E-02	**1,59E-01**	**2,54E-01**
Cu	2	0,653	40	9,98E-04	2,04E-03	3,27E-03
Fe	300	0,972	700	1,27E-02	2,60E-02	4,17E-02
Hg	0,026	5,968	0,3	**1,58E-01**	**3,23E-01**	**5,17E-01**
Mn	-	-	24	**1,61E-01**	**3,30E-01**	**5,28E-01**
Ni	25	0,053	20	2,02E-03	4,14E-03	6,63E-03
Se	1	3,127	5	1,91E-02	3,91E-02	6,25E-02
Sr	-	-	600	6,67E-03	1,36E-02	2,18E-02
Zn	30	0,262	300	8,01E-04	1,64E-03	2,62E-03
	ERI	11,643	***** very high risk			

[6] Puche, O. Mecanismos estructurales de los volcanismos paleozoicos en la región Alcudiense. PhD Thesis Universidad Politecnica de Madrid (in Spanish), 1989, pp. 472.

[7] Palero, F.J. Evolución Geotectónica y Yacimientos Minerales de la Región del Valle de Alcudia (Sector Meridional de la Zona Centroibérica). PhD Thesis Universidad de Salamanca (in Spanish), 1991, pp. 827.

[8] Palero, F., Lorenzo, S. Mercury mineralization in the region of Almadén. García-Cortés, A., Agueda-Villar, J., Palacio Suárez-Valgrande, J., Salvador González, CI (Eds.), Spanish Geological Frameworks and Geosites: An Approach to Spanish Geological Heritage of International Relevance. *Ins. Geol. Min. Esp.*, 2009, pp. 65-72.

[9] Higueras, P., Oyarzun, R., Lillo, J., Morata, D. Intraplate mafic magmatism, degasification, and deposition of mercury: the giant Almadén mercury deposit (Spain) revisited. *Ore Geol. Rev.*, Vol. 51, 2013, pp. 93–102.

[10] Berzas Nevado, J.J., Bermejo, L.G., Martín-Doimeadios, R.R. Distribution of mercury in the aquatic environment at Almaden, Spain. *Environ. Pollut.,* Vol. 122, 2003, pp. 261–271.

[11] Gray, J.E., Hines, M.E., Higueras, P.L., Adatto, I., Lasorsa, B.K. Mercury speciation and microbial transformations in mine wastes, stream sediments, and surface waters at the Almadén Mining District, Spain. *Environ. Sci. Technol.*, Vol. 38, 2004, pp. 4285–4292.

[12] Millán, R., Lominchar, M.A,, Rodríguez-Alonso, J., Schmid, T., Sierra, M.J. Riparian vegetation role in mercury uptake (Valdeazogues River, Almadén, Spain). *J Geochem. Explor.*, Vol. 140, 2014, pp. 104–110.

[13] García-Ordiales, E., Loredo, J., Esbrí, J.M., Lominchar, M.A., Millán, R., Higueras, P. Stream bottom sediments as a mean to assess metal contamination in the historic mining district of Almadén (Spain). *Int J Min Reclam Environ.*, Vol. 28, No. 6, 2014, pp.377–388

[14] Hernández, A., Jébrak, M., Higueras, P., Oyarzun, R., Morata, D., Munhá, J. The Almadén mercury mining district. *Spain Miner Deposita*, Vol. 34, 1999, pp. 539–548.

[15] Schimd, T., Millán, R., Vera, R., Tallos, A., Recreo, F., Quejido, A., Sánchez, M.D., Fernández, M. The distribution of Mercury in a characterized soils affected by mining activities. 8th International FZK/TNO Conference on contaminated soil. (Consoil 2003). Conference Proccedings. 2003, pp. 328-329.

[16] Brockhoff, C. A., Creed, J. T., Martin, T. D., Martin, E. R., & Long, S. E. EPA Method 200.8, Revision 5.5: Determination of trace metals in waters and wastes by inductively coupled plasma-mass spectrometry (Vol. 61). EPA-821R-99-017, 1999.

[17] Rapant, S., Kordík, J. An environmental risk assessment map of the Slovak Republic: application of data from geochemical atlases. *Environ. Geo.* Vol. l, No. 44, 2003, pp. 400–407.

[18] Rapant, S., Dietzová, Z., & Cicmanová, S. Environmental and health risk assessment in abandoned mining area, Zlata Idka, Slovakia. *Environ. Geol.,* Vol. 51, No. 3, 2006, pp. 387-397.

[19] Rapant, S., Salminen, R., Tarvainen, T., Krmová, K., Cveková, V. (2008). Application of a risk assessment method to Europe-wide geochemical baseline data. Geochem. *Explor. Environ. Anal.*, Vol. 8, 2008, pp. 291–299

[20] Rapant, S., Bodiš, D., Vrana, K., Cvečková, V., Kordík, J., Krčmová, K., et al. Geochemical Atlas of Slovakia and examples of its applications to environmental problems. *Environ. Geol.* Vol. 57, 2009, pp. 99-110.

[21] Huang, X., Sillanpää, M., Gjessing, E. T., Peräniemi, S., & Vogt, R. D. Environmental impact of mining activities on the surface water quality in Tibet: Gyama valley. *Sci. Total Environ.*, Vol. 408, No. 19, 2010, pp. 4177-4184.

[22] US EPA. Risk Assessment Guidance for Superfund Volume I: Human Health Evaluation Manual (Part E, Supplemental Guidance for Dermal Risk Assessment) Final. EPA/540/R/99/005 OSWER 9285.7-02EP PB99-963312 July 2004,Office of Superfund Remediation and Technology Innovation U.S. Environmental Protection Agency Washington, DC, 2004.

[23] Rodriguez-Proteau1, R., Grant, R.L. Toxicity evaluation and human health risk assessment of surface and ground water contaminated by recycled hazardous waste materials, Handb. Environ. Chem. Vol. 5, Part F, No. 2, 2005, pp. 133–189.

[24] Wu, B., Zhao, D. Y., Jia, H. Y., Zhang, Y., Zhang, X. X., & Cheng, S. P. Preliminary risk assessment of trace metal pollution in surface water from Yangtze River in Nanjing Section, China. *B. Environ. Contam. Tox.*, Vol. 82, No. 4, 2009, pp. 405-409.

[25] U.S. EPA. Exposure Factors Handbook 2011 Edition (Final). U.S. Environmental Protection Agency, Washington, DC, EPA/600/R-09/052F, 2011.

[26] Liu, L., Hawkins,D.M., Ghosh,S. and Young,S.S. Robust singular value decomposition analysis of microarray data. Proc. Natl. Acad. Sci. U. S. A., Vol. 100, 2003, pp. 13167-13172.

[27] Smith AH, Goycolea M, Haque R, Biggs ML. Marked increase in bladder and lung cancer mortality in a region of Northern Chile due to arsenic in drinking water. *Am J Epidemiol*., Vol. 147, 1998, pp. 660–669.

[28] Morales KH, Ryan L, Kuo TL, Wu MM, Chen CJ. Risk of internal cancers from arsenic in drinking water. *Environ. Health Persp.,* Vol. 108, 2000, pp. 655–666.

[29] Kazantzis, G. Role of cobalt, iron, lead, manganese, mercury, platinum, selenium, and titanium in carcinogenesis. *Environ. Health Persp.,* Vol. 40, 1981, pp. 143.

[30] Garcia-Ordiales, E., Loredo, J., Covelli, S., Esbrí, J. M., Millán, R., & Higueras, P. Trace metal pollution in freshwater sediments of the world's largest mercury mining district: sources, spatial distribution, and environmental implications. Journal of Soils and Sediments, 2016, DOI 10.1007/s11368-016-1503-5.