

Multiple impact assessment and water quality based on diatom, benthic invertebrate and fish communities in the Arieș River catchment area (Transylvania, Romania)

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SUMMARY. The present paper represents an assessment of human impacts affecting the Arieș River catchment area, a region heavily affected by the mining industry documented in the middle river course (Roșia Montană, Abrud, Roșia Poieni) since the Roman period. Other important impacts in the study area were: eutrophication / organic pollution due to discharges of untreated domestic wastes of villages and towns from the region; river regularization works, wood exploitation and processing facilities and industrial wastes downstream Turda and Câmpia Turzii localities. Water quality evaluation was carried out using river biotic communities recommended by the European legislation (Water Framework Directive, WFD): diatoms, benthic invertebrates and fish. Twenty-three sampling sites were considered along the Arieș River main course and its main tributaries, and standardized methods were employed for sampling and processing of biological data. Benthic invertebrates proved to be the most sensitive community, indicating disturbed ecological status downstream the mining-affected region mainly due to high contamination of river sediments. While ichthyofauna responses were moderate (with water quality classes usually ranging from high to moderate), diatoms reflected better the effects of eutrophication / organic pollution caused by human settlements.

Keywords: biotic indices, physico-chemical parameters, trade-off analysis, Water Framework Directive.

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Introduction

Compared to terrestrial systems, freshwaters are more susceptible to degradation, due to synergic effects of multiple human pressures: pollution from agriculture and industrial areas, domestic wastes, hydromorphological alterations, overexploitation of resources etc. (Allan and Castillo, 2007). All these activities severely affect freshwater biota, which is currently facing a biodiversity crisis (Revenge and Kura, 2003). Streams are among the most affected ecosystems, due mainly to changes in water chemistry (organic pollution, nutrients, acidification), habitat alteration and destruction, and species removal or addition (Malmqvist and Rundle, 2002).

In this context, the development of impact assessment methodologies and techniques are imperative (Anjaneyulu and Manickam, 2007). Monitoring programs for river water quality should include hydrological, hydromorphological, physico-chemical and biological parameters (Chapman (ed.), 1996). Current European water legislation, the Water Framework Directive WFD (Directive 2000/60/EC) stressed the importance of bioassessment in surface water quality assessment and monitoring, using indicator communities like algae, macroinvertebrates and fish, sensitive to habitat degradation, land-use effects or toxicity (Heiskanen *et al.*, 2004; Hering *et al.*, 2006; Solimini *et al.* (eds.), 2006).

Benthic algae, especially diatoms, are considered to be the most important primary producers in streams, because they are found in nearly all running waters and fluvial food webs (Allan and Castillo, 2007). Numerous algal biological indices are based on diatoms, due to their high ecological diversity, short life cycles, or prompt answer to short and long term changes of water (Dokulil, 2003; Wang *et al.*, 2014; Bellinger and Sigeo, 2015). Since benthic organisms have limited mobility, their presence or absence is most likely to be associated with alterations in their environmental conditions (Chapman (ed.), 1996; Malmqvist, 2002). This is the reason why benthic invertebrates are commonly used in water quality assessment studies. They exhibit a wide diversity of form, tolerance to habitat parameters and adaptation to survival in different conditions (Resh and Rosenberg (eds.), 1993; Kenney *et al.*, 2009). Fish on the other hand are associated with certain river habitats or areas, and they are very sensitive to changes in water physical, chemical and biological quality, so they are extensively used in water quality assessment studies (Chapman (ed.), 1996; Hering *et al.*, 2006; Trautwein *et al.*, 2013).

The Arieş River catchment area was considered for the present study for two reasons. Firstly, it represents the largest right tributary of the Mureş River, with a total course length of 167 km and a catchment area of almost 3000 sqkm (Ujvari, 1972; Băţinaş, 2010). Secondly, various human activities cause a wide range of impacts in different reaches of the river: human settlements and hydromorphological alterations are present from the headwaters to mouth, similar to industrial operations like mining or wood processing, that have severe consequences on the environment (Băţinaş, 2010).

This is why the impacts of mining industry in the middle reach of the Arieș River represented the topic of numerous previous studies conducted in the area, focusing on the influence of polluted right tributaries draining the Roșia Montană - Roșia Poieni regions (Forray, 2002; Senila *et al.*, 2007; Whitehead *et al.*, 2009; Băținaș, 2010; Levei *et al.*, 2013; Voica *et al.*, 2013; Levei *et al.*, 2014). Pathogenic germs (Bodoczi, 2009) and algae (Péterfi and Momeu, 1984, 1985; Momeu and Péterfi, 2007; Butiuc-Keul *et al.*, 2012; Olenici *et al.*, 2017) were also considered. Water quality was only assessed from the hyporheic zone (Moldovan *et al.*, 2011; 2013), or using the saprobial system (data from the Romanian Waters National Administration, cited in Băținaș, 2010). Diatom, invertebrate and fish communities considered for the present study were described in Momeu *et al.* (2007) and Momeu *et al.* (2009).

In this context, the aim of the present paper was to assess the major impacts from the Arieș River and its main tributaries, related to the water quality based on three biotic communities indicated by the WFD. Diatoms, benthic invertebrates and fish communities yielded comparable water quality classes, correlated with the total impact score, but benthic invertebrates were more susceptible to degradation.

Materials and methods

A number of 23 sampling sites was considered for diatoms and benthic invertebrates: 15 on the main river course (AR1-The Arieș source; AR2-Arieșeni: ski track; AR3-Arieșeni: village; AR4-Gârda; AR5-Upstream Albac; AR6-Downstream Albac; AR7-Upstream Câmpeni; AR8-Downstream junction with the Abrud; AR9-Valea Lupșii; AR10-Brăzești; AR11-Upstream junction with the Valea Ocoliș; AR12-Moldovenesti; AR13-Downstream junction with the Hășdate; AR14-Upstream junction with the Racoșa; AR15-Luncani) and 8 on several tributaries (T1-The Gârda Seacă; T2-The Albac; T3-The Arieșul Mic; T4-The Abrud; T5-The Pârâul Șesii; T6-The Valea Ocoliș; T7-The Hășdate; T8-The Racoșa). Fish communities were analyzed in 15 sites (Fig. 1). Standardized methods were used in sampling and analyzing biotic communities (Momeu *et al.*, 2009).

Water quality was assessed using data sets collected in 2007, with the exception of diatoms (where 2006 samples were also included), and fish (where samples from 2005 for AR4 and 2006 for T7 were used, from Ureche *et al.*, 2007 and Pricope *et al.*, 2009). Several biotic indices were considered for water quality assessment (Table 1). Water quality was ranked using classes defined in the WFD: 1(high), 2(good), 3(moderate), 4(poor), 5(bad). Class 6 was added (no organisms found at site) (Fig. 1).

Table 1.

List of biotic indices used for water quality assessment in the present study

Index	Description
DBI: Diatom Biological Index (IBD)	- based on diatoms; references: Lenoir and Coste (1996); Prygiel and Coste (eds.) (2000); - quantitative, counts > 400 individuals; - identifications to species level; - output: water quality classes from 1 (high) to 5 (bad).
SI: Saprobity Index	- based on diatoms; references: Zelinka and Marvan (1961); - semi-quantitative; frequency ranging from 1 (not frequent) to 5 (dominant species); - identifications to species level; - output: water quality classes based on saprobity: from xenosaprobic (very clean) to polysaprobic waters (heavily polluted, very high loads of organic matter).
BMWP: Biological Monitoring Working Party	- based on benthic invertebrates; references: Hawkes (1998) for UK; Dumnicka <i>et al.</i> (2006) for BMWP-PL (adapted for Poland); - identifications to family level for all taxa; - output: water quality classes from 1 (high) to 5 (bad).
ASPT: Average Score Per Taxon	- based on benthic invertebrates; references: Armitage <i>et al.</i> (1983); - identifications to family level for all taxa; - calculated as BMWP divided by the total number of families per sample; - output: water quality classes from 1 (high) to 5 (bad).
EBI: Extensive Biotic Index (IBE)	- based on benthic invertebrates; references: Ghetti (1997); - identifications to family level for all taxa, except for Plecoptera, Ephemeroptera, Turbellaria and Hirudinea, identified to genus level; - output: water quality classes from 1 (high) to 5 (bad).
NGBI: Normalized Global Biotic Index (IBGN)	- based on benthic invertebrates; references: AFNOR (1992); - identifications to family level for all taxa; - output: water quality classes from 1 (high) to 5 (bad).
IBI: Index of Biological Integrity	- based on fish; references: Karr (1981); - parameters used: species composition and richness, trophic structure, fish stock and biomass; - output: integrity classes from 1 (unchanged gene pool of native ichthyofauna) to 5 (originally 9) (fish population disappeared entirely, mostly due to long-term alterations).
EFI+: European Fish Index	- based on fish; references: EFI+ Consortium (2009); - parameters used: species richness and number of individuals; species guilds with respect to habitat and oxygen depletion; - salmonid and cyprinid river types, according to percentage of intolerant species belonging to salmonid dominated fish communities; - output: water quality classes from 1 (high) to 5 (bad).

An adaptation of the *trade-off analysis* was employed to calculate impact scores (Anjaneyulu and Manickam, 2007). Six impact categories were identified in the Arieș catchment area, ranked according to their importance on a scale from 0 (minimum) to 5 (maximum): mining industry: extraction and processing (5); human settlements: local houses and touristic facilities (4); agriculture (3); river regularization (3); wood processing points (3); and industry, other than wood processing and mining (3). A total impact score was then calculated for every sampling site (see Table 2), by multiplying impact importance with impact intensity, also ranked from 0 to 5. The following classes were used: 0=no impact, 1=very low intensity, 2=low intensity, 3=moderate intensity, 4=strong impact, 5=severe impact. For ranking mining industry impacts, only sites located < 10 km downstream from extraction or processing facilities were considered, while for human settlements the following threshold values were used: <5000 inhabitants; 5000-10000 inhabitants; and >10000 inhabitants.

Principal Component Analysis (PCA) was used to observe aggregation trends in the sampling sites based on abiotic characteristics, impact score, number of taxa and water quality classes indicated by biotic communities. Xlstat software 2018.6 was used (Addinsoft, 2018).

Results and discussion

Sampling sites: impact characteristics

Variations of physico-chemical parameters usually reflect the negative effects of human activities in/near rivers. In the Arieș catchment area, temperature and dissolved oxygen values carried little information in this respect, since they recorded annual means that followed an expected pattern (Momeu *et al.*, 2009). The mean temperature increased from headwaters to mouth, as described for most temperate rivers (Lampert and Sommer, 2007), while dissolved oxygen recorded constant mean values from headwaters to mouth.

Water conductivity and pH, however, were more sensitive to human pressures. Low pH values were recorded in T4 and T5, two right tributaries that collect waters from Roșia Montană - Roșia Poieni mining area (Momeu *et al.*, 2009). Acid waters (pH <5.5, acidity generated through oxidation of Fe-rich sulfides, Lottermoser, 2007) have significant impacts on river systems, because mine effluents and acid rock drainage are associated with high concentrations of metals (Whitehead *et al.*, 2009). Heavy metals (Cu, Pb, Zn etc.) and cyanides were reported in the literature, often in concentrations exceeding the legal limits stipulated in M.O. 161/2006 (Table 2).

Table 2.

List and characteristics of impacts for the 23 sampling sites from the Arieş catchment area;
 S/R - sources/references: *a* - Bătinaş (2010), *b* - Bird *et al.* (2005), *c* - Bodoczi (2009),
d - Butiuc-Keul *et al.* (2012), *e* - Costan (2010), *f* - Forray (2002), *g* - Levei *et al.* (2013),
h - Levei *et al.* (2014), *i* - Luca *et al.* (2006), *j* - Senila *et al.* (2007), *k* - Voica *et al.* (2013),
l - *in situ* observations; for site codes: see text.

Site	Impact categories and characteristics [total impact score in square brackets]	S/R
AR1	1) domestic wastes (local houses and guest houses) [12] 2) wood processing facilities [3]	<i>l</i>
AR2	1) domestic wastes (local houses and guest houses) [12] 2) wood processing facilities [6]	<i>l</i>
AR3	1) domestic wastes (local houses and guest houses) [12] 2) wood processing facilities [9]	<i>a, i, l</i>
T1	1) domestic wastes (local houses and guest houses) [8] 2) wood processing facilities [9]	<i>l</i>
AR4	1) domestic wastes (local houses and guest houses) [12] 2) wood processing facilities [9]	<i>a, i, l</i>
AR5	1) domestic wastes (local houses and guest houses) [8] 2) wood processing facilities [9]	<i>a, i, l</i>
T2	1) domestic wastes (local houses and guest houses) [8] 2) wood processing facilities [9] 3) regularization (banks) [6]	<i>a, i, l</i>
AR6	1) domestic wastes (local houses and guest houses) [8] 2) wood processing facilities [9]	<i>a, i, l</i>
T3	1) domestic wastes (local houses) [8] 2) wood processing facilities [9]	<i>l</i>
AR7	1) domestic wastes (no treatment plant); water eutrophication (high PO ₄ values) [16] 2) wood processing facilities [9] 3) regularization (banks) [9] 4) industrial facilities [12]	<i>a, d, l</i>
T4	1) domestic wastes (no treatment plant) [12] 2) mining industry (Roşia Montană mining exploitation for Au and Ag, closed in 2006: Cetate quarry, 2 ore dumps, 2 tailing ponds, preparation site in Gura Roşiei): acid waters; high concentrations of Fe, Cu, Zn, Cd, Mn (values exceeding legal limits according to M.O. 161/2006), but also As (arsenic); severe risk of Pb and As contamination from the Gura Roşiei tailing pond; high contamination with Zn in the sediments of the Abrud River [25]	<i>a, b, f, g, h, i, k, l</i>
AR8	1) domestic wastes (no treatment plant); water eutrophication (high PO ₄ values) [16] 2) mining industry (Roşia Montană mining exploitation, closed in 2006): acid waters; high concentrations of Fe, <u>Cu</u> , Zn, Cd, Mn (values exceeding legal limits according to M.O. 161/2006) [25]	<i>a, b, d, i, k, l</i>
AR9	1) domestic wastes; faecal pollution (faecal coliforms / faecal enterococi germs), predominantly animal [12] 2) mining industry (Roşia Montană mining exploitation, closed in 2006) [20] 3) cultivated lands [3]	<i>c, k, l</i>
T5	1) domestic wastes [8] 2) mining industry (Abrud mining exploitation for Cu: Roşia Poieni quarry, 3 ore dumps, 3 tailing ponds, preparation site in Roşia Poieni): acid waters; high concentrations of sulfates, Fe, Mn, Cu, Cd (values exceeding legal limits according	<i>a, b, g, h, i, j, l</i>

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Site	Impact categories and characteristics [total impact score in square brackets]	S/R
	to M.O. 161/2006); severe risk of Cu contamination from the Șesii tailing pond; high contamination with Cu and As in the sediments of the Pârâul Șesii Rivulet [25]	
AR10	1) domestic wastes (no treatment plant); faecal pollution (faecal coliforms / faecal enterococi germs), predominantly animal; water eutrophication (high PO ₄ values) [12] 2) mining industry (Baia de Arieș mining exploitation for Au, Ag, sulfides, closed in 2006: subterranean extraction, ore dumps, 4 tailing ponds, preparation plant): acid waters; high concentrations of cyanides, Fe, Cu, Pb, Zn (values exceeding legal limits according to M.O. 161/2006); severe risk of Pb and As (and also Ba) contamination from the Brăzești tailing pond [25]	<i>a, b, c, d, g, i, k, l</i>
AR11	1) domestic wastes [8] 2) regularization (exploitation of construction materials in/near the riverbed) [15]	<i>i, l</i>
T6	1) domestic wastes [8]	<i>l</i>
AR12	1) domestic wastes [12] 2) mining industry (Iara mining exploitation for Fe: mine, tailing pond, waste dump; closed in 2006) [5] 3) cultivated lands [9] 4) regularization (exploitation of construction materials in/near the riverbed) [9]	<i>b, e, h, l</i>
T7	1) domestic wastes [12] 2) cultivated lands [3] 3) regularization (exploitation of construction materials in/near the riverbed) [6]	<i>l</i>
AR13	1) domestic wastes; water eutrophication (high PO ₄ values) [12] 2) cultivated lands [9] 3) regularization (exploitation of construction materials in/near the riverbed) [6]	<i>d, l</i>
AR14	1) domestic wastes; high biochemical oxygen demand (BOD) values in effluents coming from the RAGCL Câmpia Turzii treatment plant [20] 2) cultivated lands [12] 3) regularization (exploitation of construction materials in/near the riverbed) [9] 4) industrial facilities (Turda industrial plants: S.C. Holcim S.A., S.C. Sticla S.A., S.C. Electroceramica, S.C. Uzina Chimică Turda, some closed): high concentrations of chlorides, sulfates, Fe, <u>Cu</u> , Pb, Zn, Cd etc. [15]	<i>a, b, i, l</i>
T8	1) domestic wastes [20] 2) cultivated lands [12] 3) regularization (exploitation of construction materials in/near the riverbed) [9] 4) industrial facilities (Câmpia Turzii wire production plant, Industria Sârmei Câmpia Turzii): high concentrations of chlorides, sulfates, Fe, Cu, Pb, Zn, Cd etc. [15]	<i>a, i, l</i>
AR15	1) domestic wastes; faecal pollution (faecal coliforms / faecal enterococi germs), predominantly human [16] 2) cultivated lands [15] 3) regularization (exploitation of construction materials in/near the riverbed) [9] 4) industrial facilities: high concentrations of Cu and Cd [12]	<i>b, c, l</i>

The upper reach of the Arieș River, from AR1 to T3 was mainly impacted by the presence of human settlements without wastewater treatment plants and by wood processing facilities like sawmills, different sawing machines, sawdust deposits etc., but total impact scores were low (< 25) in all sites, except for AR7, where river regularization and industrial impacts also occurred. High impact scores (30-40) were assigned to sites from the middle river stretch (T4 - AR10), due to severe effects of past and present mining activities in the area: discharges and

seepages of acid waters from mines, ore dumps and tailing ponds (both active and inactive), enriched in metals (Fe, Cr, Ni, Pb, Zn, Cu, Cd, As) (Forray, 2002; Bird *et al.*, 2005; Whitehead *et al.*, 2009; Băţinaş, 2010; Levei *et al.*, 2013; Voica *et al.*, 2013; Levei *et al.*, 2014). The river recovered downstream this impacted area (AR11, AR12), mainly due to the input of cleaner left tributaries (T6, T7). Thus, impact scores did not exceed 30 and included human settlements, the effects of cultivated land and river regularization. The lower river course (AR14, AR15) was characterized by high impact scores (>50) that added the effects of industrial facilities located in Turda and Câmpia Turzii to the list of impact.

Water quality assessed by biotic indices

Three classes of water quality were assessed at each site, based on diatoms, benthic invertebrates and fish (when available). When biotic indices indicated different quality classes at one sampling site, the worst evaluation was used, inspired by the WFD *one-out, all-out* rule (Fig. 1).

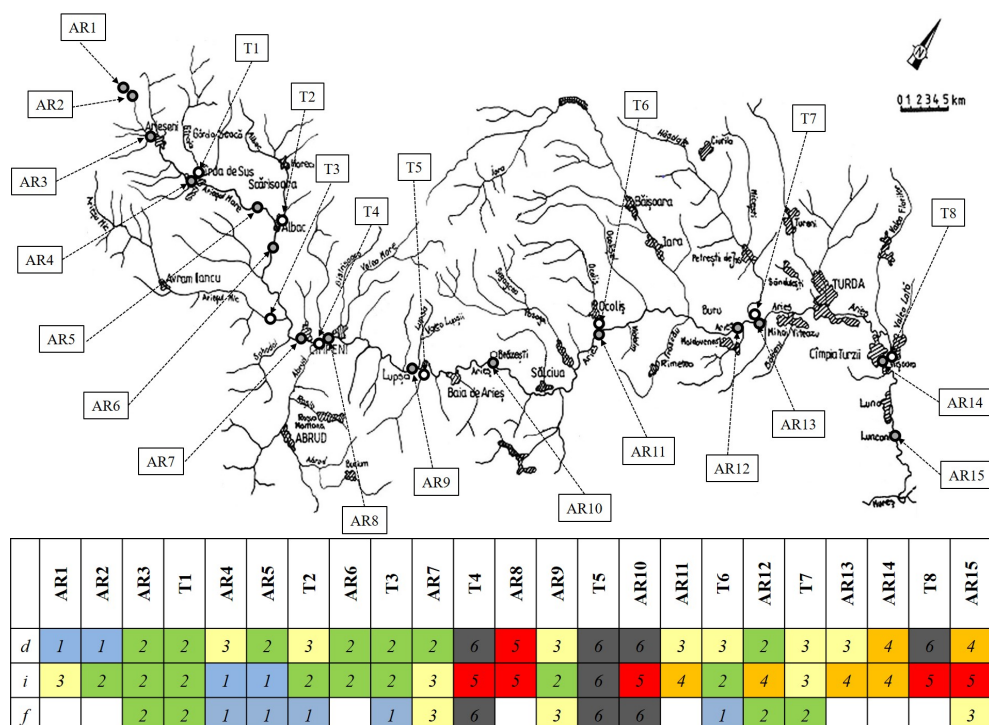


Figure 1. Water quality in the 23 sampling sites from the Arieş River catchment area: *d* - diatoms; *i* - benthic invertebrates; *f* - fish; 1 to 5: water quality classes and their colour code, according to WFD (1- high, 2- good, 3- moderate, 4- poor, 5- bad); 6 - no organisms found at site; white squares - no fish data; for site codes: see text.

Quality classes assessed by the three biotic communities were congruent, but not identical: they showed relatively good quality in the upper river reaches (AR1-T3), the worst quality in the middle river segment affected by mining activities (T4 - AR10) and moderate to bad quality in the lower stretch of the river (AR13 - AR15) (Fig. 1).

Strong negative correlations (Pearson coefficient $r > -0.576$; $p < 0.025$) were observed between water quality classes and the number of taxa, for all biotic communities. PCA biplot (Fig. 2) depicted these relationships: the higher the number of taxa, the smaller the value of the water quality class (i.e. classes 1 - 2, meaning good ecological status). This tends to be self-evident, despite the fact that pristine ecosystems are known to harbor lower number of taxa, perfectly adapted to undisturbed conditions (the *intermediate disturbance hypothesis*, Connell, 1978). The strong positive correlation between the total impact score and the water quality reflected by benthic invertebrates was also clearly represented ($r = 0.673$; $p = 0.006$), meaning that high impact scores were found in sites with water quality classes of 4 to 6 (degraded ecological status) (Fig. 2). The PCA biplot also showed that low pH and high conductivity values also correlated with inferior water quality.

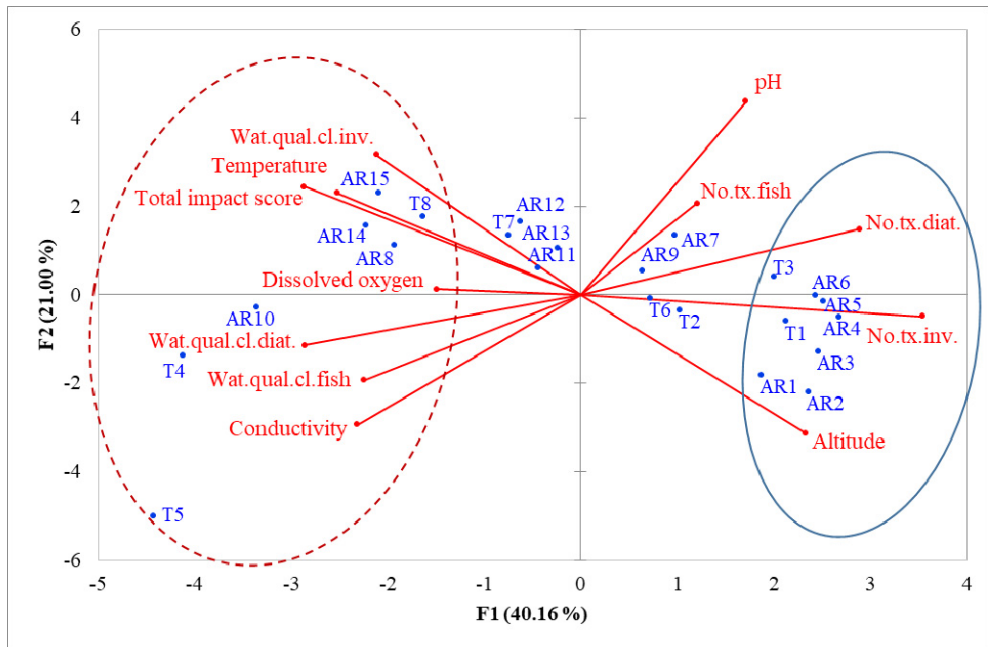


Figure 2. Principal Component Analysis PCA biplot (axes F1 and F2: 61.16 %) for the sampling sites, and their aggregation based on different biotic and abiotic parameters: altitude; pH; conductivity; temperature; dissolved oxygen; water quality classes (Wat.qual.cl.) from 1 (best) to 6 (worst) based on diatoms (diat.), benthic invertebrates (inv.) and fish; and number of taxa (No.tx.); blue circle - sites with high or good water quality; red dotted circle - sites with degraded water quality; for site codes: see text.

Benthic invertebrates were the most sensitive to various impacts, especially mining activities. Studies from the middle Arieş course reported that river sediments were highly contaminated with heavy metals like Cd, Cu or As, coming mostly from the mine-affected right-side effluents (Levei *et al.*, 2014). Sediments were generally found to be more widely contaminated than surface waters (Bird *et al.*, 2005). Moreover, the bioavailable fraction (i.e. percentage found in labile or dissolved forms) of elements potentially toxic in elevated concentrations (Fe, Mn, Zn, Cu etc.) was reported to be extremely high (Senila *et al.*, 2015). This high contamination of sediments, habitat for benthic invertebrates, explained the severe effects shown by these communities downstream the region affected by mining activities (water quality classes were 4, 5 or 6) (Fig. 1).

The water column on the other hand, was reported by various studies to be less contaminated compared to the sediments. Bird *et al.* (2005) showed that the Arieş was much less polluted than the Abrud River, with only Cu showing concentrations above guideline values, since elevated metal levels in surface waters were confined to within approximately 10 km of point sources. The moderate influence of the polluted tributaries on the Arieş River water quality was explained by Senila *et al.* (2015) as a consequence of the tributaries low flow rate compared with that of the Arieş River. All these factors led to good water quality assessed by benthic invertebrate community at AR9. The high contamination with Cd, Pb and As coming from the Brăzeşti tailing pond, inactive at present (Levei *et al.*, 2013) caused the worst water quality at AR10, since no organisms were found most of the times (Fig. 1). Benthic invertebrates showed class 5 and not 6 in T4 and AR10, due to the presence of several Chironomid individuals, probably coming from upstream.

Diatom indices used in the present study yielded comparable water quality classifications: DBI classes 1 and 2 were reflected by SI oligosaprobic and oligo- β -mesosaprobic levels, class 3 by oligo- β -mesosaprobic to β - α -mesosaprobic levels, while class 4 by α -polysaprobic levels. Similar findings were previously reported in the Arieş catchment area (Momeu and Péterfi, 2007).

Diatoms were known to be sensitive to a wide range of stressors (Wang *et al.*, 2014), however diatom metrics were reported to be better correlated to eutrophication and organic pollution (Hering *et al.*, 2006). Similar trends were identified in the present study: in the upper Arieş reach, water quality at sites AR4 and T2 was ranked "moderate" (class 3) by diatom indices, and "high" or "good" (classes 1 and 2) by invertebrates and fish. Since the dominant impact in the area was untreated domestic wastes coming from human settlements (Table 2), diatoms seemed to be more sensitive to eutrophication/organic pollution compared to other biotic communities.

Ichthyofauna ecological characteristics in streams are greatly influenced by hydromorphology (Solimini *et al.* (eds.), 2006), so fish communities are extensively used to assess hydromorphological degradation, especially in lowland rivers and at meso-scales (Hering *et al.*, 2006). Our data however did not support these findings,

since water quality indicated by fish biotic indices ranged from high to moderate (classes 1 to 3), apart from T4, T5 and AR10, where no fish were caught (Fig. 1). The EFI+ quality classes were similar to IBI integrity classes, even though in 7 sites EFI+ was recommended to be used with caution, because the number of fish caught was under 30.

Conclusion

The present study assessed the impacts in the Arieș River catchment area, a severely affected water course due primarily to mining industry facilities from the Roșia Montană - Roșia Poieni region, but also to domestic waste discharges, river regularization works and industrial activities, other than mining (wood processing, chemical, wire production etc.). High impact scores correlated with a decrease in water quality shown by biotic communities: diatoms, benthic invertebrates and fish. Benthic invertebrates were more sensitive to degradation, showing the poorest water quality classes in most of the cases.

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