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Macroinvertebrates as Indicators of Water Quality in Running Waters: 10 Years of Research in Rivers with Different Degrees of Anthropogenic Impacts

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1. Introduction

The management of running waters is of great importance for the life of our society and one of the challenges to be met by future generations. The sustainable use of water resources for their exploitation in different aspects is essential. Also, the maintenance of good water quality, both sanitary and environmental, is essential, since it depends largely on the conservation of biodiversity (Fernández-Díaz, 2003).

Rivers are ecosystems of great ecological value with a rich fauna that consists of communities with a complex structure and high biological value. However, their special typology makes them fragile and vulnerable to environmental changes, especially those related to disturbances of anthropogenic origin, which often imply irreversible degradation of their biota (Beasley & Kneale, 2003; Dahl et al., 2004).

The vulnerability of these habitats is also evident in relation to the possible effects of climate change. Among the most affected ecosystems are rivers and streams. One of the predictable effects could be that some of these systems will be transformed from permanent to seasonal and some of them will even disappear. In consequence, the biodiversity of many of them will be reduced and their biogeochemical cycles will be altered (Jenkins et al., 1993).

One of the major impacts that affect rivers is the pollution of their waters by both domestic and industrial waste (Benetti & Garrido, 2010). Also agriculture, with intensive use of fertilizers and pesticides, has contributed significantly to eutrophication and contamination of aquatic ecosystems (García-Criado et al., 1999; Paz, 1993). Another important impact on running waters is the deliberate modification of streams by building dams and reservoirs which alter the ecological characteristics of their basins (Richter et al., 1997).

Fluvial ecosystems support very rich and diverse assemblages, with developed adaptations that allow them to prosper in these environments, and which, at the same time, make them very vulnerable to possible alterations in the habitat. In this sense, human activity often causes severe ecological damage to river systems. These disturbances produce alterations in

the chemical composition of water and in the structure of the communities of organisms living in this environment (Oller & Goitia, 2005; Smolders et al., 1999, 2003).

Among the fauna of rivers that should be highlighted are macroinvertebrates. This group of great diversity and ecological importance consists of invertebrates of macroscopic size, normally more than 1mm, living permanently or during certain periods of their life cycle linked to the aquatic environment. They include insects, crustaceans, annelids, molluscs, leeches, etc.

Different groups of macroinvertebrates are excellent indicators of human impacts, especially contamination. Most of them have quite narrow ecological requirements and are very useful as bioindicators in determining the characteristics of aquatic environments (Benetti & Garrido, 2010; Fernández-Díaz et al., 2008; Pérez-Bilbao & Garrido, 2009), to identify the segments of a polluted river where self-purification of organic inputs is under process (Chatzinikolaou & Lazaridou 2007).

The Water Framework Directive (WFD 2000/60/EC) establishes common principles to coordinate the efforts of Member States to improve the protection of the European Community aquatic systems, to promote sustainable water use and to protect ecosystems. This directive is intended to prevent the deterioration of all types of water bodies and to ensure that these environments achieve good quality status. The WFD specifies several quality elements necessary for assessing the ecological state of a river. These elements are hydromorphological, physical, chemical and biological. For the latter the composition and abundance of benthic fauna, including invertebrates, are used. The presence/absence of certain taxa defines the quality state of a watercourse. This directive requires that member states of the European Union achieve good quality of all their water bodies by 2015.

So far, the European intercalibration process has produced class boundaries for four out of five types of Mediterranean rivers (R-M1, R-M2, R-M4 and R-M5) (European Commission, 2008) using benthic macroinvertebrates. The officially selected multimetric index for the intercalibration (European Commission, 2008) of the MedGIG rivers is the STAR_ICMi (Buffagni et al., 2006), which is also used by the Central European and Baltic GIG.

Biological monitoring has been established to control water quality. These studies are often based on the sampling of an area and the subsequent analysis of collected specimens that are suitable for monitoring the area and provide information on pollution trends. The structure of the community of one or more of these specimens (Cheimenopoulou et al., 2011) is used in classifying the watercourse ecological quality in a five-class system by using the ecological quality ratio between the observed to the reference conditions or biotic indices/scores.

Among these indices, diversity indices such as Shannon-Wiener or Margalef have been used or indices or scores based on aquatic macroinvertebrates. In Spain it is used the IBMWP score, which uses the presence of taxa and scores for their tolerance to pollution.

The purpose of this chapter is to study macroinvertebrate fauna as bioindicators of water quality in rivers. The questions are if invertebrates are good indicators of water quality in rivers and which are the effects of the impacts of human activities on invertebrate assemblages living in these habitats.

This chapter presents results from studies conducted over 10 years (1998-2008) in 10 rivers in the Autonomous Region of Galicia (North-western Spain) located in areas with different degrees of anthropogenic impacts. The selection of sampling sites was based on land uses near the river banks (woodlands, agriculture, transport system, urban areas, and industrial activities) in connection to some other habitat parameters. Several abiotic variables were also recorded at the same time as fauna was sampled. Benthic macroinvertebrates and their indices were used for the quality assessment. We also analyzed the biotic indices-environment and assemblage composition-environment relationships in order to study responses to structural characteristics of the habitat (natural or artificial) and variations in water quality parameters.



Fig. 1. Site LI3 of the Limia River (Ourense, NW Spain), before the central power station.

2. Water quality and anthropogenic impacts

Freshwater biodiversity provides a broad variety of valuable goods and services for human societies, some of them irreplaceable (Dudgeon et al., 2006), but human activities have always affected aquatic ecosystems. Rivers are highly vulnerable to change caused by anthropogenic impacts, and their flow is often manipulated to provide water for human use (Bredenhand & Samways, 2009). Globally, the biodiversity of freshwater ecosystems is rapidly deteriorating as a result of human activities (Dahl et al., 2004). According to

Dudgeon et al. (2006), there are five major threat categories to freshwater biodiversity: overexploitation, water pollution, flow modification, destruction or degradation of habitat, and invasion by exotic species.

It is possible that in future decades human pressure on water resources will further endangering aquatic biodiversity present in these systems (Strayer, 2006). Overexploitation of rivers and aquifers for irrigation is already a severe problem in many places, especially in the Mediterranean region. In most countries in the south of Europe, irrigation accounts for over 60% of water (Abellán et al., 2006). This activity can lead to drought and the disappearance of inland aquatic habitats (Abellán et al., 2006; Belmar et al., 2010) or changes in physical and chemical characteristics (Velasco et al., 2006).

Contamination due to different types of pollutants such as fertilizers, sewage, heavy metals or pesticides, is a serious problem worldwide. Increasing urbanization and industrialization generates different non-point sources of contamination, causing impairment of water quality of rivers (Beasley & Kneale, 2003). Many studies have dealt with the negative effect of different pollutants on aquatic biota, which results in biodiversity loss and poor water quality (Beasley & Kneale, 2003, 2004; Benetti & Garrido, 2010; Fernández-Díaz et al., 2008; Garrido et al., 1998; Harper & Peckarsky, 2005; Hirst et al., 2002; Lytle & Peckarsky, 2001; Smolders et al., 2003; Song et al., 2009).

Dam construction is one of the most important modifications that rivers are subjected to (Belmar et al., 2010). The general effect is the transformation of dynamic patterns into static, relatively stable ones with reduced flows (Baeza et al., 2003; Benejam et al., 2010; Stanford & Ward, 1979). Changes in marginal vegetation and in flow velocity may produce changes in the composition of aquatic assemblages, with the replacement of some species by others due to the destruction of microhabitats and the creation of new ones (Fulan et al., 2010; Lessard & Hayes, 2003; Sarr, 2011).

Widespread introduction and invasion of exotic species constitutes another human impact on freshwaters. This usually causes the extinction of indigenous species by competition or predation and biotic homogenization (Raehl, 2002). There are many examples of exotic species invasions, for instance the Nile perch, the American signal crayfish or the zebra mussel.

3. Biological indicators

In the past, water quality was assessed using only physicochemical parameters, but these variables only reflect punctual pollution. The use of biological indicators is more adequate to detect long-term changes in water quality, since aquatic organisms are adapted to specific environmental conditions. If these conditions change, some organisms can disappear (intolerant) and be replaced by others (tolerant). Therefore, variations in the composition and structure of aquatic organism assemblages in running waters can indicate possible pollution (Alba-Tercedor, 1996).

Biomonitoring is the use of biological variables to survey the environment (Gerhardt, 2000). The first step in this type of monitoring is to find the ideal bioindicator whose presence, abundance and behavior reflects the effect of a stressor on biota (Bonada et al., 2006b).

Benthic macroinvertebrates are considered as good indicators of local scale conditions (Metcalfe, 1989; Freund & Petty, 2007). These invertebrates live on the bottom of aquatic ecosystems at least part of their life cycle and can be collected using aquatic nets of 500 μm or less (Hauer & Resh, 1996) or ISO 7828 (EN 27828, 1994). They include molluscs, crustaceans, leeches, worms, flatworms and insects (especially larval stages of some orders). Aquatic macroinvertebrates are used to bioassess aquatic ecosystem quality due to their great diversity of form and habits (Rosenberg & Resh, 1993). According to Johnson et al. (1993) a biological indicator has to fulfil different characteristics:

- to be taxonomically easy and well-known,
- to be widely distributed,
- to be abundant and easy to capture,
- to present low genetic and ecological variability,
- to be preferably big,
- to have low mobility and a long life cycle,
- to present well-known ecological characteristics,
- to have the possibility of being used in laboratory studies

Different sampling protocols and metrics are used to evaluate the water quality of rivers and streams. Among them, biotic indices are the most used because they are highly robust, sensitive, cost-effective, easy to apply, and easy to interpret (Bonada et al., 2006a; Chessman et al., 1997; Dallas, 1995, 1997). Biotic indices are tools for assessing quality based on the different response of organisms to environmental changes (Ministry of Environment, 2005). There are many biotic indices developed for different regions, for instance the TBI (Trent Biotic Index, Woodiwis, 1964), the BMWP (Biological Monitoring Working Party) and the ASPT (Average Score per Taxon) (Armitage et al., 1983; National Water Council, 1981) for the UK, the BBI - Belgian Biotic Index (De Pauw & Vanhooren, 1983; Gabriels et al., 2005) for Belgian rivers, the FBILL (Foix, Besòs i Llobregat, Prat et al., 1999) and the IBMWP (Alba-Tercedor et al., 2002) for Spain, or the HES (Hellenic Evaluation System) (Artemiadou & Lazaridou, 2005) for Greece. Many of these indices have to be adapted when they are used in different regions from where they were developed.

One of the most used biotic indices in the Iberian Peninsula is the IBMWP (Iberian Biological Monitoring Working Party) formerly BMWP' (Alba-Tercedor & Sánchez-Ortega, 1988), which is an adaptation of the British BMWP (Armitage et al., 1983) for the Iberian Peninsula. The taxonomic resolution of this index is mostly at family level, and in some cases is even considered at a higher level. Each benthic macroinvertebrate family (or higher taxa) has a score in relation to their tolerance to pollution, so the sum of the scores of the different taxa found in one site gives a total score allowing this sampling site to be classified in one of the five water quality classes (Alba-Tercedor et al., 2002):

- Class I: very good (≥ 101)
- Class II: good (61-100)
- Class III: acceptable (36-60)
- Class IV: poor (15-35)
- Class V: bad (< 15)



Fig. 2. Central power station of the DevaPO River (Pontevedra, NW Spain).

4. Case study

4.1 Introduction

The negative influence of human impacts, especially pollution, on macroinvertebrate fauna has been described in different studies (Blasco et al., 2000; Dahl et al., 2004; Elbaz-Poulichet et al., 1999; Nummelin et al., 2007; Smolders et al., 2003; Yoshimura, 2008; Artemiadou et al., 2008). In Spain, in recent years such studies have increased considerably and several papers studying these types of impacts in different regions of the country have been published. Amongst them we highlight Alonso (2006) in Madrid, Bonada et al. (2000) and Ortiz et al. (2005) in Catalonia, García Criado & Fernández Aláez (2001) in León, or Marqués et al. (2003) in the Basque Country.

Despite its importance, few studies describe the effects of the impact of hydroelectric power stations, for instance Bredenhand & Samways (2009) in South Africa, Kubecka et al. (1997) in the Czech Republic, Jesus et al. (2004) in Portugal, Lessard & Hayes (2003) in Michigan (USA), Stanley et al. (2002) in Wisconsin (USA), Thomson et al. (2005) in Pennsylvania (E.E.U.U.) or Tonkin et al. (2009) in New Zealand. In Spain we must highlight the study by Oscoz et al. (2006) in Navarre, north of the country, which explored both the impacts of pollution and the impacts of hydroelectric power stations.

So far, in Galicia there are few studies to assess human impact on invertebrate fauna. Some of them have analyzed the effects of anthropogenic impacts on water beetle fauna, one of the most important groups of invertebrates. These studies focused mainly on the effects of water pollution (Benetti & Garrido, 2010; Benetti et al., 2007; Pérez-Bilbao & Garrido, 2009), while Sarr (2011) explores the impact of small hydroelectric stations. The results of these studies provide a basis for conducting this study, which focuses on the impact on the entire fauna of macroinvertebrates. Additionally, this study is partly based on technical reports of different environmental monitoring programs developed by the research group at the University of Vigo and the Ingenieria y Ciencia Ambiental S.L. Company (Garrido et al., 1999, 2000a, 2000b, 2000c, 2003, 2005).

Concerning the macroinvertebrates this study (a) assesses the importance of invertebrate fauna as an indicator of water quality in these rivers; (b) identifies the response of macroinvertebrates to human activities; (c) and denotes the responsible factors for the differentiation of the studied rivers.

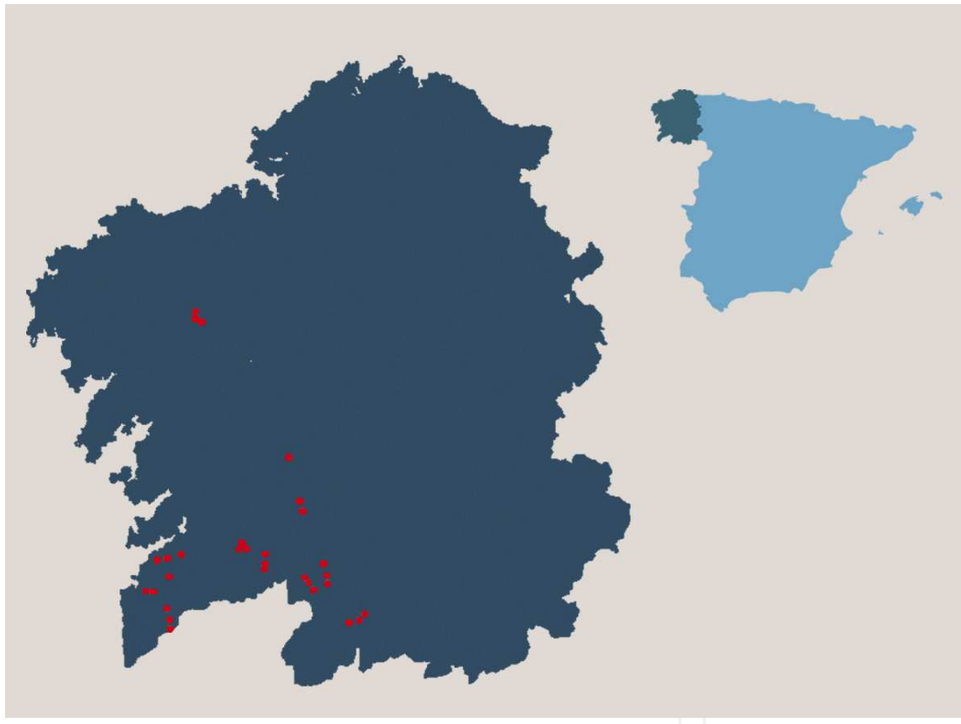


Fig. 3. Map of the study area showing the location of the sampling sites.

4.2 Material and methods

4.2.1 Study area

The study area comprised 10 rivers located in the Autonomous Region of Galicia (North-western Spain) (Figure 3). According to the Köppen-Geiger Climate Classification, the climate of the study area is warm temperate, with dry summers and mild temperature (Kottek et al., 2006). This territory belongs to the Atlantic and the Mediterranean biogeographical regions. Altitude ranges from the sea level to around 1,000 m in the

highlands. Due to its geographical location, orography and climate, this area has a large number of running waters, from large rivers to small streams (Figure 1). The landscape consists of woodlands (temperate broadleaf forest, pine and eucalyptus reforestations), farmlands, urban and industrial areas. According to the WFD (Annex II, System A), the studied rivers were classified as: Iberian-macaronesian ecoregion, siliceous/calcareous, lowland/mid-altitude and small/medium catchment area.

These rivers are located in areas with different degrees of anthropogenic impacts. The Lagares River (Figure 4) runs almost entirely through the urban area of Vigo, a city with approximately 300,000 inhabitants. This river has undergone a profound change in its structure, especially channelling, as a result of the growth of the city and rapid industrial development with the consequent establishment of industries on its banks. The rest of the rivers run mostly through rural and semi-urban areas. The course of the rivers Furnia and Miñor has not been altered very much, but the other rivers (Avia, DevaOU, DevaPO, Limia, Tambre, Tea and Tuño) have at least one small hydroelectric power station (Figure 2).

4.2.2 Sampling methods and variables measured

In a period of 10 years, between June 1998 and April 2008, 10 rivers were sampled. Each river was sampled four times, in spring, summer, autumn and winter. In each river 3 sites were selected, located in the upper, middle and low stretches (Table 1). The selection of sampling sites was based on land uses (woodlands, agriculture, transport system, urban areas, and industrial activities) and the position of the hydraulic infrastructures, for the regulated rivers. In these cases the first site was located upstream the dam, the second between the dam and the power station, and the third downstream the station.

Sampling was carried out in all types of substrate for a standardized time (one minute). Fauna was collected with an entomological water net of 30 cm diameter, 60 cm depth and 0.5 mm mesh. The specimens were stored in a 4% formaldehyde solution and taken to the laboratory, where they were sorted and identified. After being studied, they were conserved in 70° alcohol and deposited in the scientific collection of the Aquatic Entomology Laboratory at Vigo University.

The following water quality parameters were measured at each site: temperature, dissolved oxygen, pH, electrical conductivity and total dissolved solids (TDS). Additionally, we measured the altitude (meters above sea level) for each site. These above parameters are considered fundamental in the typology of rivers by the WFD.

4.2.3 Data analysis

The structure of the assemblages was assessed using different diversity indices: richness (S), rarefied richness (ES), abundance (N) and the Shannon-Wiener diversity index (H'). The IBMWP biological index was also calculated. The values of rarefied richness were calculated for 100 individuals ES (100). ES and H' were calculated using PRIMER version 6. Analysis of variance (two-way ANOVA) was used to test for significant differences between seasons in both diversity indices and environmental variables. ANOVA was run using SPSS version 19.

		UTM Coordinate			
River	Site	X	Y	Altitude	Sampling Date
Ávia	AV1	567297	4709038	569	1998-1999
Ávia	AV2	571104	4693658	113	1998-1999
Ávia	AV3	572157	4690485	108	1998-1999
Deva OU	DEOU1	575784	4662895	552	1998-1999
Deva OU	DEOU2	574167	4665277	329	1998-1999
Deva OU	DEOU3	572991	4666511	192	1998-1999
Deva PO	DEPO1	558785	4674976	508	2001-2002
Deva PO	DEPO2	558972	4671670	350	2001-2002
Deva PO	DEPO3	558650	4670559	220	2001-2002
Furnia	FU1	524677	4655852	77	2007-2008
Furnia	FU2	525442	4652027	36	2007-2008
Furnia	FU3	525603	4649199	6	2007-2008
Lagares	LA1	529266	4675118	240	2001-2002
Lagares	LA2	524323	4673627	34	2001-2002
Lagares	LA3	520918	4673068	10	2001-2002
Limia	LI1	593494	4654101	610	2003
Limia	LI2	591737	4652248	600	2003
Limia	LI3	588536	4651147	550	2003
Miñor	MI1	525456	4667235	330	2001-2002
Miñor	MI2	519199	4662061	9	2001-2002
Miñor	MI3	517156	4662213	2	2001-2002
Tambre	TA1	536133	4756871	210	1998-1999
Tambre	TA2	534947	4758037	190	1998-1999
Tambre	TA3	534574	4759534	179	1998-1999
Tea	TE1	550473	4678468	192	1999
Tea	TE2	550950	4677584	114	1999
Tea	TE3	550656	4677503	109	1999
Tuño	TU1	580854	4665249	730	1998-1999
Tuño	TU2	580506	4667324	484	1998-1999
Tuño	TU3	579643	4671803	372	1998-1999

Table 1. Sampling sites, with their code, location in UTM coordinates, altitude and sampling date.

Relationships between environmental variables with diversity indices and fauna were determined by a Pearson correlation test. Prior to this, the Kolmogorov-Smirnov test was used to verify the normal distribution of the data. Variables not following the normal distribution were logarithmically transformed ($\log_{10}$). These analyses were performed using SPSS version 19.

Canonical correspondence analysis (CCA) was used to analyze fauna-environment relationships in order to identify environmental factors potentially influencing macroinvertebrate assemblages. A Monte Carlo permutation test was performed to assess the statistical significance of the environmental parameters and the full model to arrive at the significance of the first two canonical axes (Heino, 2000). The environmental factors used were pH, water temperature, dissolved oxygen and conductivity. TDS was not considered because it was redundant with conductivity. CCA was carried out on global abundances, that is, total number of individuals collected at a site over the sampling period. Taxa with less than 10 individuals were removed from the analysis, which was carried out using the CANOCO 4.5 program (Ter Braak & Šmilauer, 2002).

Complete linkage cluster analysis with Bray-Curtis coefficient was used to cluster the rivers into groups and thus be able to verify differences in assemblage composition. The results were represented graphically by Multidimensional scaling (MDS). SIMPER analysis was used to identify which species generate the most similarity within each MDS group. For the SIMPER routine, the raw data were square root transformed and reporting was limited to species with more than 2.5% contribution to dissimilarity. This analysis was carried out with PRIMER version 6.



Fig. 4. Lagares River (Pontevedra, NW Spain) impacted by pollution.

4.3 Results

4.3.1 Diversity and biological indices

In total, 115 taxa (mostly family) of 7 phyla were collected (Table 3). The most representative groups were insects (86 families), especially the orders Diptera (20 families), Trichoptera (19 families) and Coleoptera (12 families).

Only the family Elmidae was recorded in all sites. Other very common families were Baetidae and Chironomidae (recorded in 29 sites), and Nemouridae and Simuliidae (recorded in 28 sites). Besides, 10 families (Bithyniidae, Capniidae, Hebridae, Heteroceridae, Libellulidae, Noteridae, Pyralidae, Scatophagidae, Sciomyzidae and Sisyridae) were only recorded in one site.

If we analyse these results by samples, we can see that site 3 in the Limia River (LI3) presented the highest value of invertebrate richness in autumn with 68 taxa recorded, followed by FU2 (Furnia River) in spring and DEOU1 (DevaOU River) in winter with 55 taxa each. On the other hand, the lowest richness was found in LA3 (Lagares River) in spring, where only 7 taxa were collected, followed by TA2 (Tambre River) in winter and in autumn with 8 and 11 taxa recorded respectively.

Total abundance was 217,577 individuals (185,287 insects, 14,931 Annelida, 9,430 Mollusca and 7,929 other groups). The greatest abundance was observed in the third site in the Limia River in spring, with 29,931 individuals collected. On the other hand, the lowest value was obtained in the third site of the Lagares River in spring with 59 individuals. The most abundant family of macroinvertebrates was Chironomidae with 40,584 individuals recorded. Other abundant families were Ephemerellidae (25,788 individuals), Baetidae (22,860), Elmidae (15,171) and Simuliidae (13,611).

Rarefied species richness is the expected number of species for a given number of randomly sampled individuals and facilitates comparison of areas in which densities may differ (McCabe & Gotelli, 2000). The highest values correspond to the Limia River, sites LI3 (24.18) and LI2 (24.13) in autumn, and to the Furnia River, site FU2 in autumn (23.51) and spring (23.27). On the contrary, the Lagares River had the lowest values in site LA2 (6.62) and site LA3 (6.92).

The Shannon-Wiener index (H'(log2)) revealed that most of the studied rivers presented high diversity values. The lowest diversity was recorded in DEPO3 (DevaPO River) in winter (0.86) and the highest in LI3 (Limia River) in autumn (4.40). In general, the diversity values were high, greater than 3 in 60% of the samples.

According to the IBMWP index, most samples (95%) presented good water quality (> 60, class II), even 87% of the samples presented very good quality, because the index values were above 100 (class I). The highest value (338) was obtained at site FU2 in the Furnia River and the lowest (26) at site LA3 in the Lagares River. Only 2 samples, belonging to the Lagares and Tambre rivers, obtained low values, below 35 and therefore classified as poor quality (class IV). No samples presented bad water quality (< 15, class V).

Table 2 shows the mean minimum and maximum values of the diversity indices for the studied rivers. There were no significant differences ($p < 0.05$) among seasons in any of the diversity indices, as evidenced by the ANOVA.

Richness measures	Mean ± SD	Minimum	Maximum	ANOVA F	p
Richness S	30.98 ± 11.84	7	68	0.128	0.943
Rarefied Richness ES (100)	15.70 ± 4.62	6.62	24.18	0.502	0.682
Abundance N	2175.77 ± 3445.50	59	23931	1.579	0.202
Diversity H'(log2)	3.04 ± 0.75	0.86	4.40	0.535	0.660
IBMWP	174.51 ± 70.47	26	338	0.181	0.909

Table 2. Mean, SD and ranges of biological and diversity indices of the samples and ANOVA with season as factor.

4.3.2 Environmental variables

Table 4 shows mean minimum and maximum values of the environmental variables measured in the 10 studied rivers. The main result to highlight is the high value of conductivity measured in some sites, especially in the Lagares River, higher than in the other surveys. ANOVA showed no significant differences ($p<0.05$) among seasons in almost all environmental variables. This analysis only showed significant differences among seasons in temperature, as expected.

4.3.3 Influence of environmental variables on macroinvertebrate assemblages

The Pearson correlation test was performed to assess the relation between the environmental variables and the taxa and diversity indices. We found several significant correlations ($p<0.05$), but most of them were low ($r<0.5$). We only highlight those that were higher ($r>0.5$). Regarding the diversity indices, there were significant negative correlations between conductivity and rarefied richness ($r=-0.52$) and diversity ($r=-0.50$). For the taxa we found a significant negative correlation between oxygen concentration and Naididae ($r=-0.81$), and a significant positive correlation between conductivity and Hydrobiidae ($r=0.51$).

Figure 5 shows the results of the CCA. The eigenvalues for axes 1-4 were 0.400, 0.198, 0.092 and 0.074 respectively. Correlations for axes III and IV with environmental variables were low ($r<0.5$), and only axes I and II were used for data interpretation. The cumulative percentage of variance for the species-environmental relation for these two axes was 76.1%. The first two canonical axes were significant, as shown by the Monte Carlo permutation test ($p=0.004$).

The first principal axis is positively correlated with conductivity ($r=0.892$) and temperature ($r=0.788$), and negatively with altitude ($r=-0.532$). This component describes water quality and can be an indicator of contamination, since all Lagares river sites are located at the positive end of the axis. The second axis is positively correlated with temperature ($r=0.396$) and negatively with oxygen ($r=-0.786$). Rivers Limia and Miñor are located at the negative end of this axis, indicating low oxygen values. According to the CCA analysis, the family Naididae is related to sites with low oxygen values and the family Hydrobiidae to sites with high conductivity values, while Enchytraeidae prefer sites with high pH values.

Higher taxa	Taxa	Ávia	Deva OU	Deva PO	Furnia	Lagares	Limia	Miñor	Tambre	Tea	Tuño
Hydrozoa	Hydridae		5			1					
Turbellaria	Dugesiidae	16				2	159				
Turbellaria	Planariidae	37	366	256	72	43	14	27			14
Nematoda	Nematoda		11		2	82	53	3			5
Nematomorpha	Gordiidae		2								1
Hirudinea	Erpobdellidae	55	1	16	2	20	84	2	30		
Hirudinea	Glossiphoniidae	10	32			1	15		8	17	753
Hirudinea	Hirudinidae		6		1						
Oligochaeta	Enchytraeidae	1621	735	595		1957	1386	45	726	127	413
Oligochaeta	Lumbricidae	65		162			53		25		
Oligochaeta	Lumbriculidae		25			27	320			3	40
Oligochaeta	Naididae	1	7	492		646	2609	1261	25	220	
Oligochaeta	Oligochaeta unidentified				61						
Oligochaeta	Proppapidae						62				
Oligochaeta	Tubificidae	84		13			72				
Bivalvia	Sphaeriidae	10	23	1	1	11	193	9	104		11
Gastropoda	Ancylidae	20	620	11	10	15	282		33	8	111
Gastropoda	Bithyniidae		1								
Gastropoda	Hydrobiidae	6	84			6960		15	218	2	2
Gastropoda	Lymnaeidae	79	8		13		119		148		5
Gastropoda	Physidae	6				39	213		32		
Gastropoda	Planorbidae	1	1		1		1				
Gastropoda	Valvatidae		2						1		
Crustacea	Cladocera					1	45				
Crustacea	Copepoda		12			10	184				
Crustacea	Ostracoda				1	31		1			
Crustacea	Asellidae		17				233				5
Crustacea	Gammaridae	2	8	11	534	37		70	805		421
Arachnida	Hydracarina	66	1244	188	34	83	2311	65	130	187	22
Collembola	Collembola				8	20	163	3			
Odonata	Aeshnidae	24	64	17	14	2	17		44	22	28
Odonata	Calopterygidae	18	47	26	55	2	21		60	7	17
Odonata	Coenagrionidae	2					14				
Odonata	Cordulegasteridae	12	31	5	5	4	92	3	1	11	47
Odonata	Gomphidae	102	6	3	57	1	709	13	40	54	1
Odonata	Lestidae				2		20				
Odonata	Libellulidae						2				
Plecoptera	Capniidae						1				
Plecoptera	Chloroperlidae	133	7	131	41		6	32	1	6	1
Plecoptera	Leuctridae	430	623	161	272		1789	46	419	73	1458
Plecoptera	Nemouridae	670	871	301	264	11	58	368	30	102	403
Plecoptera	Perlidae	11	48	6	8		2		26	37	50
Plecoptera	Perlodidae	1	2	35	84		2	2		13	33

Ephemeroptera	Baetidae	551	3765	2638	371	2411	6078	3117	1146	850	1933
Ephemeroptera	Caenidae	401				1	1094		256	4	
Ephemeroptera	Ephemerellidae	213	864	591	278	28	22547	42	578	257	390
Ephemeroptera	Ephemeridae		328	4	6		20			3	87
Ephemeroptera	Heptageniidae	180	933	57	45	35	2909	37	82	27	184
Ephemeroptera	Leptophlebiidae	1004	505	2342	258		119	132	13	112	428
Ephemeroptera	Oligoneuriidae		1				253		9		3
Ephemeroptera	Potamanthidae			532							
Hemiptera	Aphelocheiridae				1		215		112	54	
Hemiptera	Corixidae	6					33		1	4	
Hemiptera	Gerridae	13	16	54	17	25	84	11	76	40	20
Hemiptera	Hebridae						3				
Hemiptera	Hydrometridae		9	5			16		2		
Hemiptera	Mesoveliidae	2	1				1				
Hemiptera	Naucoridae				1		47				
Hemiptera	Nepidae				1		4				
Hemiptera	Notonectidae						4				
Hemiptera	Veliidae				1		2		2		
Coleoptera	Dryopidae	3	13	9	1		48	1	36	11	19
Coleoptera	Dytiscidae	9	14	79			799	1	8		4
Coleoptera	Elmidae	663	1899	3615	448	22	4474	200	1702	298	1850
Coleoptera	Gyrinidae		1	51	73		112		11	9	11
Coleoptera	Haliplidae					2	165				
Coleoptera	Helophoridae			1			6		1		
Coleoptera	Heteroceridae						40				
Coleoptera	Hydraenidae	395	500	396	126	1	414	24	15	33	239
Coleoptera	Hydrochidae		1				44				
Coleoptera	Hydrophilidae	4	1	12	1	1	139	2	23	3	
Coleoptera	Noteridae						1				
Coleoptera	Scirtidae		20	25	62		2	1		48	2
Diptera	Anthomyiidae					8	268	1	1	9	
Diptera	Athericidae	132	298	31	35	23		30	200	60	24
Diptera	Blephariceridae		3		3		17				
Diptera	Ceratopogonidae	28	106	189	21	41	2	9	10	8	27
Diptera	Chironomidae	1597	9418	13729	1326	3575	376	5287	1750	620	2906
Diptera	Culicidae						7471	1			
Diptera	Dixidae	6	30	2	5		35			3	4
Diptera	Dolichopodidae	8	123				2		5	6	5
Diptera	Empididae	22	348	221	208	94	1	74	13	81	133
Diptera	Limoniidae	62	57	18	19		1005	11		10	66
Diptera	Psychodidae	11	31	402	13	39	4	13		29	56
Diptera	Ptychopteridae						21				
Diptera	Rhagionidae	3	7	3	3	2	26				1

Diptera	Scatophagidae				2						
Diptera	Sciomyzidae	11									
Diptera	Simuliidae	787	3832	2829	1020	550	2	2740	559	168	1124
Diptera	Stratiomyidae						51				
Diptera	Tabanidae	2	7	2	1		762	3		9	2
Diptera	Thaumaleidae						3				
Diptera	Tipulidae	20	25	9	1	4	2		11	6	12
Lepidoptera	Pyralidae						9				
Megaloptera	Sialidae	29	22	2			2			1	
Neuroptera	Sisyridae						37				
Trichoptera	Beraeidae	5	4					3	19	13	1
Trichoptera	Brachycentridae	18		1496	34		651	6	21		10
Trichoptera	Calamoceratidae	5	24		5		64		175		25
Trichoptera	Ecnomidae		2				4				6
Trichoptera	Glossosomatidae	1	63	8	34	1	1	3			3
Trichoptera	Goeridae	5	148	7	1					1	1
Trichoptera	Helicopsychidae	43	107	15	24	1	1	3			17
Trichoptera	Hydropsychidae	291	837	567	466	82	4367	43	482	118	199
Trichoptera	Hydroptilidae	2		9	14		38	1			
Trichoptera	Lepidostomatidae	25	43	40	111	3		41	1	22	2
Trichoptera	Leptoceridae	49	69	71	12	5	20	3		9	5
Trichoptera	Limnephilidae	39	76	172	7		4416	1	3	5	12
Trichoptera	Philopotamidae	61	98	179	569	4	5421	63		22	9
Trichoptera	Phryganeidae		4				32				
Trichoptera	Polycentropodidae	27	45	94	11		663	4	19	5	53
Trichoptera	Psychomyiidae	5	5	73	5	1	10				
Trichoptera	Rhyacophilidae	59	235	147	56	20	200	33	58	14	124
Trichoptera	Sericostomatidae	59	463	222	124	1	8	3	2	67	173
Trichoptera	Uenoidae	20	16	2	19			20			1

Table 3. Abundance of macroinvertebrates recorded in 10 rivers between 1998 and 2008.

				ANOVA	
Variable	Mean ± SD	Minimum	Maximum	F	p
Water Temperature °C	13.90 ± 3.38	7.90	22.00	70.536	0.000
pH	6.43 ± 0.63	5.20	8.17	0.297	0.828
Conductivity µS/cm	60.95 ± 38.12	20.00	186.00	0.975	0.410
Oxygen %	97.83 ± 5.76	80.40	107.00	1.436	0.239

Table 4. Mean, SD, minimum and maximum values of the environmental variables measured in the studied rivers and ANOVA with season as factor.

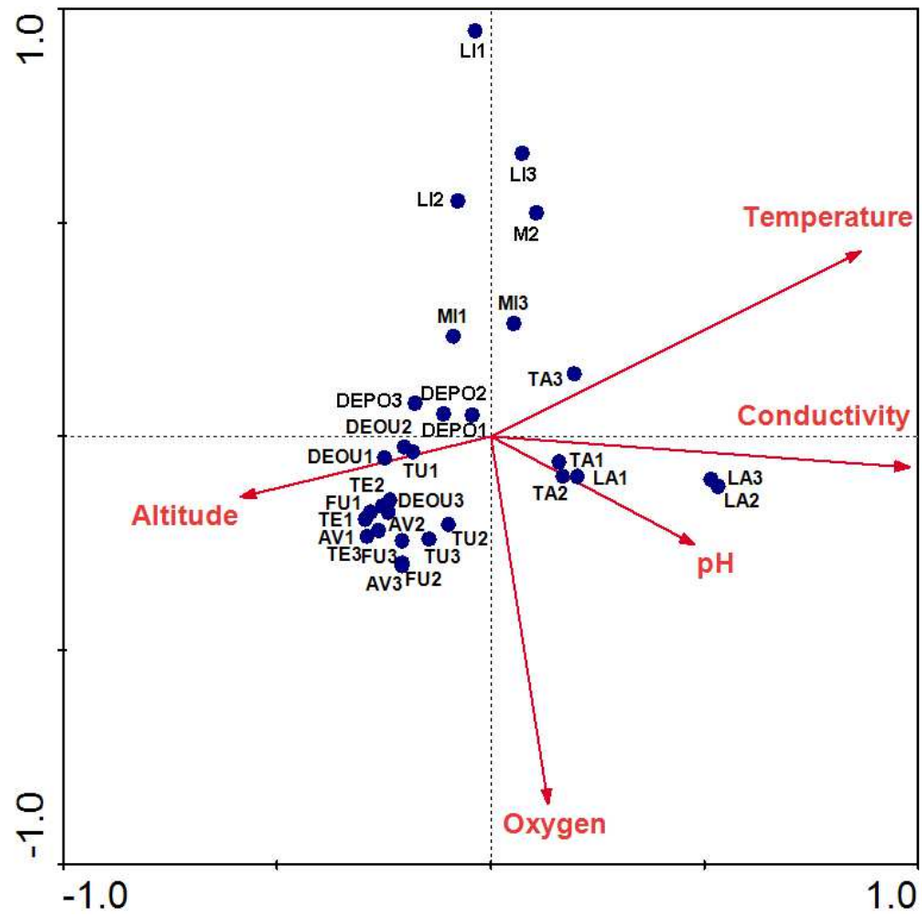


Fig. 5. Results of the canonical correspondence analysis (CCA) based on the invertebrates assemblages with respect to environmental variables. Arrows represent the environmental variables and circles the sites.

4.3.4 Assemblage composition

The Bray-Curtis coefficient was used to calculate the affinity between rivers and the results ranged from 0 to 100. If the value obtained is close to 100, populations will be more similar. According to this coefficient the studied rivers had a 36.32 average faunal affinity. The greatest degree of affinity was observed between the rivers DevaOU and DevaPO (68.20), and between the Tambre and Tuño (61.95). On the other hand, the rivers faunistically farthest were the Limia and Tea, with 7.66 of affinity. MDS analysis provided alternative insights into the similarity of sites with regard to macroinvertebrate assemblage composition. Figure 6 shows the formation of four clearly separated groups with low faunal affinity between them, less than 40%.

The Limia River has an affinity of less than 25% with all the other rivers, forming a group apart and completely separated from the rest. The Lagares River also forms a separate group, with a greater affinity with the Miñor River. Finally, we identified two other groups, one formed by the rivers Miñor, DevaPO and DevaOU, and another formed by the rivers Avia, Furnia, Tambre, Tea and Tuño.

Table 5 lists the species that contributed the greatest amount of similarity in groups A and B. We found a relatively high level of overall similarity in group A (59.26%) and B (50.81%) with the SIMPER analysis. The taxa that contributed most to similarity were the same in both groups. The most important taxon was Chironomidae, contributing with 42.64% similarity in the group A and 23.73% similarity in the group B. Other important taxa were Baetidae, Elmidae and Simuliidae.

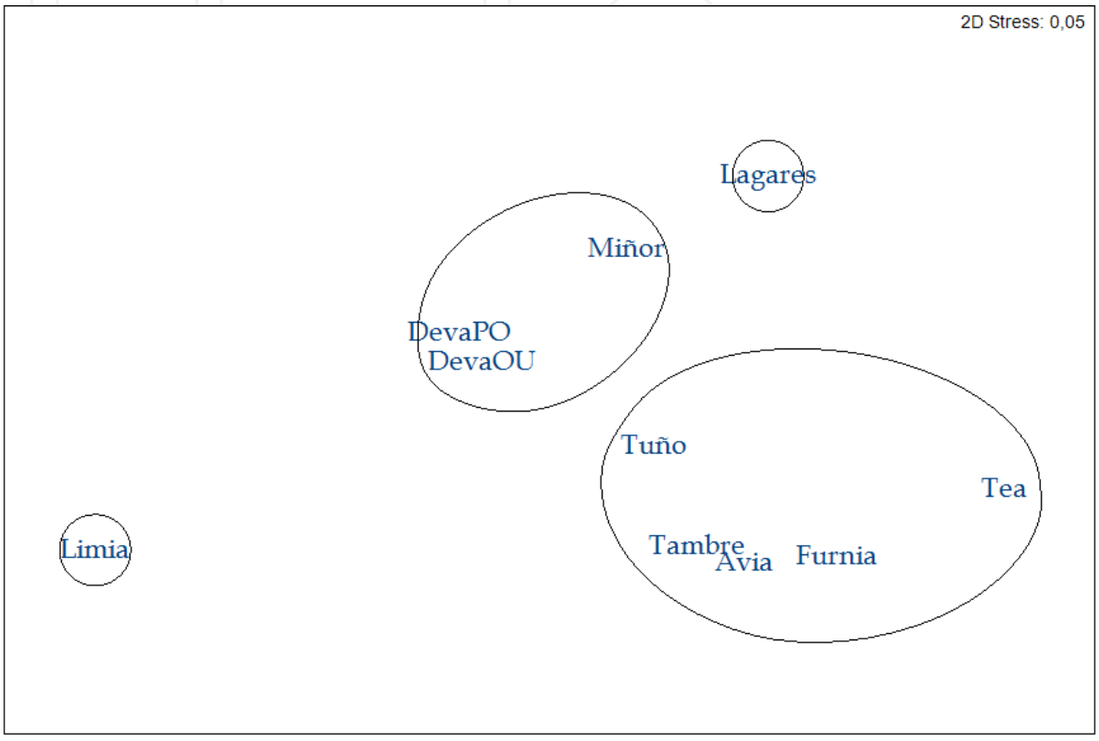


Fig. 6. Multidimensional scaling (MDS) from complete linkage clustering based on the Bray-Curtis coefficient.

4.4 Discussion

The importance of using macroinvertebrates as bioindicators of the water quality of rivers has already been highlighted by several authors (Alba-Tercedor et al., 2002; Alonso, 2006; Bonada et al., 2000; Ortiz et al., 2005; Oscoz et al., 2006). The importance of this group is also reflected in this study because we were able to evaluate the conservation status of these rivers and assess the degree of impact that they are subjected to, whether by pollution or the construction of hydroelectric power stations.

In general, the biological and diversity values observed in the studied sites were high. However, in some places, especially in the Lagares River, these indices values are considerably low in comparison with other rivers in northern Spain (Álvarez-Troncoso, 2004; Fernández-Díaz, 2003; García-Criado, 1999; Paz, 1993; Pérez-Bilbao & Garrido, 2009). We also found a negative correlation between conductivity and richness parameters with a significant decrease in rarefied richness and diversity in sites with high values of this variable, especially in the Lagares River. The reduction in macroinvertebrate richness and

abundance in stretches with high values of variables indicative of pollution reflect anthropogenic impacts and corroborates the data obtained by the biological index, which showed poor quality. In relation to this, the results obtained in this study are in concordance with others reported in other papers, which have often documented a decrease in richness and diversity in water bodies with high values of chemical variables (Heino, 2000, Prenda & Gallardo-Mayenco, 1996; Thorne & Williams, 1997).

Taxa	Mean Abundance	Contribution to similarity (%)
Group A (Average similarity: 59.26)		
Chironomidae	9478	42.64
Baetidae	3173.33	18.86
Simuliidae	3133.67	18.48
Elmidae	1904.67	4.34
Group B (Average similarity: 50.81)		
Chironomidae	1639.8	23.73
Baetidae	970.2	13.15
Elmidae	992.2	11.24
Simuliidae	731.6	9.99
Ephemerellidae	343.2	5.74
Leuctridae	530.4	4.7
Hydropsychidae	311.2	4.57
Enchytraeidae	577.4	3.7
Nemouridae	293.8	2.84
Leptophlebiidae	363	2.79
Gammaridae	352.4	2.66

Table 5. SIMPER analysis of macroinvertebrates assemblages of groups A (Miñor, DevaOU and DevaPo rivers) and B (Avia, Furnia, Tambre, Tea and Tuño rivers).

IBMWP index values obtained in the samples of this study, mainly above 100, highlighted the good preservation state of the studied rivers. Besides, it is important to note that the highest value was obtained in the Furnia River, a very little impacted river with excellent water quality. On the contrary, the lowest value was obtained in the Lagares River, which is highly polluted and affected by anthropogenic pressure. The importance of using this index was also pointed out by other studies conducted in the Iberian Peninsula (Alba-Tercedor et al., 2002; Bonada et al., 2006a; Poquet et al., 2009).

In addition to the reduction in richness, abundance and biological indices, pollution also causes a change in faunal composition, as reflected by the separation of the Lagares River from the others in the faunal affinity analysis. In this sense, it is important to note that the dominance of certain taxa, e.g. Naididae and Chironomidae, and absence of others, e.g. Plecoptera, at some sites may indicate the existence of alterations in them (Oscoz et al., 2006).

According to Beasley & Kneale (2003), increasing urbanization and industrialization generates different non-point sources of contamination, causing impairment of water quality of rivers. This environmental impact can be seen in the city of Vigo and its surroundings. High anthropogenic pressure on aquatic ecosystems in this region is a consequence of the ever-increasing population and establishment of industries, especially on the banks of rivers (Benetti & Garrido, 2010).

The response of macroinvertebrates to water pollution seems to define, at least, the species typical of non-contaminated sites. In this sense, ordination analysis identified a group of sensitive taxa especially evident for those most abundant, whose numbers fall considerably in impacted sites. According to CCA, there are also tolerant taxa, for example the Hydrobiidae family, correlated with high values of conductivity, and Naididae, correlated with low values of dissolved oxygen. These results are in agreement with those found in other studies. According to Brinkhurst & Gelder (2001), several aquatic oligochaetes, including Naididae species, have red blood pigments which aid oxygen uptake and transport, thus they can live in environments with low oxygen. Pérez-Quintero (2007) documented that some Hydrobiidae species were salinity-tolerant, so possibly also conductivity-tolerant, as these variables are closely correlated.

Most studies about impacts in rivers (Benetti & Garrido, 2010; Dahl et al., 2004; Nummelin et al., 2007) are mainly focused on the impact assessment of different sources of water pollution, without considering the impacts of infrastructures that change riverbeds, such as the case of small hydro. One of the best ways to evaluate the impact produced by hydroelectric power stations on wildlife is to check changes in the faunal composition and mainly their feeding habits (Argyroudi et al., 2010). In the studied rivers, we observed that the impact produced by hydroelectrics brings about a change in the hydrological regime, mainly the damming, and consequently a change in the community of invertebrates, especially altering its faunal composition, something already noted by other authors for different groups of invertebrates (Bredenhand & Samways, 2009; Jesus et al., 2004; Sarr, 2011; Stanley et al., 2002; Yoshimura, 2008). The structure of macroinvertebrate assemblages is influenced by factors such as the hydrological regime, substrate stability, type and abundance of trophic resources, or land use in the river basin (Dessaix et al., 1995; Quinn et al., 1997; Zamora-Muñoz & Alba-Tercedor, 1996). In our study, both natural characteristics (geology, substrate, water flow) and those artificially created by the impact of hydroelectric infrastructures, determined the structure of the invertebrate assemblages.

The regulation of rivers and hydropower development changes the habitat structure (Dessaix et al., 1995; Dolédec et al., 1996; Fjellheim & Raddum, 1996; Oscoz et al., 2006) and causes the loss of more sensitive taxa and thus imbalance in community structure (Fjellheim et al., 1993). Also, this impact causes the disappearance of many species and otherwise artificially created microhabitats are colonized by other species, perhaps more tolerant to changes or perhaps better adapted to new habitats formed in the river bed and that are not characteristic of their original bed. This may be the case of the Limia River, isolated from the rest and with low faunal affinity, especially in site LI1 (Figure 6), situated upstream the power station, as damming causes the slowdown of the water flow and low levels of oxygen, similar to that observed in stagnant water environments.

In conclusion, in the studied rivers the main factors that determined the macroinvertebrate fauna were (1) water pollution, which directly affected water quality and (2) damming, a disturbance that has caused a change in water flow. In this sense, the negative effect of the anthropogenic impact, especially water contamination, on macroinvertebrates in some of the studied rivers is evident, as shown by the decrease in richness attributes and the IBMWP biological index in impacted sites. As anticipated, there is a loss of species as land use changes from rural to urban. Besides, the water quality in rivers located in natural areas and without strong impacts is better than in rivers located in areas with urban land uses. We also have demonstrated that macroinvertebrates can be used as indicators of environmental impacts in rivers. Their responses to impacts in rivers differ; the majority of taxa are not tolerant to increasing contamination and changes in river structure, but some taxa seem to have adapted to these changes and become dominant in highly disturbed sites. As expected, rare taxa appear to be unmistakeably associated with good water quality, which highlights the importance of conserving freshwater habitats.



Fig. 7. Site LI1 of the Limia River (Ourense, NW Spain), with slow flow caused by damming.

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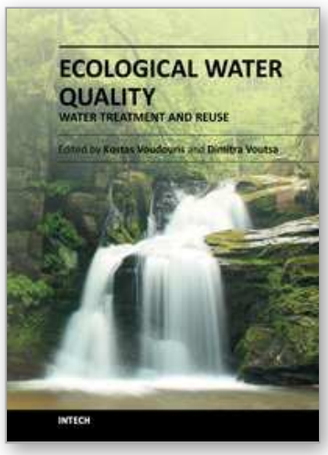
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This book attempts to cover various issues of water quality in the fields of Hydroecology and Hydrobiology and present various Water Treatment Technologies. Sustainable choices of water use that prevent water quality problems aiming at the protection of available water resources and the enhancement of the aquatic ecosystems should be our main target.

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