



Sensitivity of macroinvertebrate indicator taxa to metal gradients in mining areas in Northern Spain



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ABSTRACT

The study of macroinvertebrate communities using a Reference Condition Approach (RCA) is a powerful tool for detecting the impacts of mining activities due to heavy metal contamination in rivers. However, studies on this topic based on an RCA fully satisfying the Water Framework Directive (WFD) criteria for reference site selection are still scarce. Following a WFD-compliant RCA design, a total of 14 reference and 15 test sites were intensively sampled during July 2015 in the Nalón River basin (Asturias, Northern Spain). Macroinvertebrate samples, water physicochemistry, metal concentrations in sediments and habitat quality were evaluated at each selected site. To determine whether increasing heavy metal levels in sediments have significant ecological impacts in the structure of macroinvertebrate communities in the study area, we applied a combination of non-parametric multivariate and univariate procedures, as well as gradient analysis and Threshold Indicator Taxa ANalysis (TITAN) over the biotic and environmental datasets. We found significant alterations in the macroinvertebrate community structure with respect to the reference condition (REF group) for those test sites within mercury (HG group) and gold (AU group) mining areas, with Ephemeroptera, Plecoptera and Trichoptera (EPT) richness and abundance being the community descriptors showing the strongest alterations in relation to mining pressures in the studied area. Metal concentrations in sediments were significantly correlated to the metrics of community structure related to abundance, richness and biodiversity, with As and Hg being the two metals showing higher biological effects on macroinvertebrate assemblages. TITAN also allowed us to establish a set of macroinvertebrate families specifically sensitive to metal concentrations in sediments, most of which corresponded to EPT families. Our results prove that determined sensitive taxa could be considered as reliable indicators of metal pollution in the mining areas of Northern Spain. From a WFD perspective, our study clearly indicates that the responsive community descriptors found in this study are actually key parameters in the evaluation of the ecological status in the rivers of Northern Spain. These data are highly relevant for the future development of environmental quality standards for river sediments in Spain.

1. Introduction

Freshwater ecosystems are one of the most endangered ecosystems worldwide, mainly due to human dependence on them (Sala et al., 2000; Dudgeon et al., 2006). Increasing anthropic pressure on running waters has led to severe levels of environmental degradation in these ecosystems. Indeed, industrialisation, urbanisation, land-use changes and watercourse alterations are the main drivers threatening running waters (Malmqvist & Rundle, 2002). Among the existing pressures, mining activities may well be one of the most persistent impacts on rivers and streams (Marqués et al., 2001a), and metal pollution has become a major concern regarding the preservation of aquatic systems

in and downstream of mining areas (Luoma et al., 2010). In general, together with suspended particulate material, sediments represent the main storing site for heavy metals in rivers (mainly in the finer fraction, < 63 µm) (ERMITE-Consortium, 2004), and both sediment deposition and resuspension may represent potential sources of pollution at different spatial scales (Martin, 2000).

Assessing the risk of the ecological impacts from human activities is the core of an effective management of freshwater ecosystems, and in this regard, the evaluation of the relationships between biological responses and environmental pressures is crucial. The Water Framework Directive (WFD, EC, 2000) approach is focused on the development of ecological status classification systems for water bodies based on assessments of the degree of

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deviation in biological condition at particular study sites from the so-called Reference Condition (EC, 2000). In Europe, this concept is defined by the use of minimally disturbed sites (*sensu* Stoddard et al., 2006) characterised by type-specific biological conditions and the accompanying physicochemistry and hydromorphology, in which the absence of significant human disturbances has been assessed (Pardo et al., 2012). Within this context, the use of different biological quality elements (i.e., macrophytes, phytobenthos, phytoplankton, benthic macroinvertebrates and fish) is promoted by the WFD as an effective assessment tool for detecting impacts derived from anthropogenic pressures, such as eutrophication or organic pollution (Sánchez-Montoya et al., 2010; Pardo et al., 2014). Among them, metrics based on macroinvertebrate community structure are probably the most widely used indicators of water quality (Metcalf, 1989; Reynoldson & Metcalfe-Smith, 1992; Birk et al., 2012; García et al., 2014). In fact, worldwide mining-derived water and sediment contamination by metals was demonstrated to have a direct effect on different benthic community structural components, such as abundance, diversity, richness, or metrics related to sensitive species (Hirst et al., 2002; Solà et al., 2004; Doi et al., 2007; Qu et al., 2010; Poulton et al., 2010; Ruiz-García et al., 2012; Byrne et al., 2013). For all that, studies where reference conditions are designated according to WFD precepts are desirable towards creating an adequate background to allow being able to efficiently use benthic macroinvertebrates as indicators of metal pollution.

The Nalón River basin was one of the main mining-exploited areas in Northern Spain, where the Texeo copper mines (located in the Riosa district) and the mercury mines (Mieres, Pola de Lena and Somiedo districts) were still active until the early 1970s (Méndez-Fernández et al., 2015). Spoil heaps derived from past mining activities can still represent an important source of local contamination for river sediments (Loredo et al., 2006, 2010). Previous studies have reported sediment pollution in rivers affected by mine spoil heaps, as well as metal bioaccumulation and toxicity in the Nalón River basin (Ordóñez et al., 2013; Méndez-Fernández et al., 2015). The environmental quality standards (EQS) directive (EC, 2013) recognised the importance of both sediments and biota matrices for water quality policies in Europe. However, both sediment and biota EQSs for metals and metalloids (henceforth, metals) need to be developed by member states. Different proposals have attempted to relate metal tissue concentrations to metal-induced ecological impacts on different macroinvertebrate metrics (Luoma et al., 2010; Schmidt et al., 2011; Rainbow et al., 2012; De Jonge et al., 2013; Bervoets et al., 2016). However, the integration of metal levels in the sediment due to mining activities and the ecological impacts on river macroinvertebrate communities should be the object of deeper research, especially in regions like the Nalón River basin with a long history of mining activities. Thus, the evaluation of metal concentrations in sediments within a Reference Condition Approach (RCA) may be relevant not only in the development of useful tools for assessing the risk of mining activities derived from metal pollution, but also in the development of EQSs in Spain or at European level.

A key issue in biomonitoring is the level of taxonomic resolution used. Although recent studies pointed out the benefits of using finer taxonomic levels in stream assessment (Guerold, 2000; Lenat & Resh, 2001; Svitok et al., 2014), the family level continues to be preferred as it was demonstrated to provide valuable ecological information (Lenat & Barbour, 1994) and is able to respond significantly to different pressures (for example, see Pardo et al., 2014). Moreover, different studies demonstrated similar response patterns to different stressors at both the genus and family levels (i.e., Bowman & Bailey, 1997; Pond et al., 2008). Considering that family constitutes the level of resolution used in ecological quality assessment in the context of the WFD in Northern Spain (Pardo et al., 2014; METI, 2015), evaluating the ability of this taxonomic level to detect significant metal contamination is of high relevance.

The general aim of this study is to detect and estimate the possible effects of sediment metal pollution on the macroinvertebrate communities inhabiting the rivers located in mining areas of the Nalón River

basin in order to improve assessment, management and conservation efforts. We hypothesise that metal pollution plays a fundamental role in structuring macroinvertebrate communities in these areas. The specific objectives include 1) to describe the community composition of macroinvertebrates at both reference and potentially impacted sites, 2) to assess the response of a benthic community structure to a gradient of metal concentration in the sediments based on the metal levels measured at each site as a ratio with a pre-selected reference condition in the study area, 3) to detect benthic taxa that are sensitive to metal pollution using a wide exposure range of sediment concentrations, and 4) to determine whether the variation in metrics at the family level can be detected by alterations due to sediment metal concentrations. Overall, our research on the macroinvertebrate communities within the mining areas of the Cantabrian region using a reference condition approach may facilitate the identification of target-sensitive families for the development of adequate EQSs, both at regional and European scales.

2. Materials and methods

2.1. Study area

This study was conducted in the Nalón River basin (Asturias, Northern Spain), including both the Nalón and Narcea rivers and some of their tributaries (Fig. 1). This basin is the largest one within the Cantabrian water district, with a total length of 140.8 km covering a total area of 4907 km². The dominant climate is oceanic, characterised by mild winters and summers, and abundant rainfall throughout the year (a mean annual precipitation of 1296 mm). As a consequence of its lithological characteristics, the study region also witnessed historical mining activity, with more than 800 sites active between 1950 and 1975 (Águeda-Villar & Salvador-González, 2008; Ordóñez et al., 2008).

A total of stream 29 stream sites were sampled once during July 2015 (Fig. 1). The sampling design considered two main groups of sites: reference sites ($n = 14$) and test sites ($n = 15$), and covered four of the macroinvertebrate community-based river typologies (Types 1, 2, 4 and 5) of the Cantabrian region (Pardo et al., 2014). Reference sites (REF group) were selected according to WFD criteria (EC, 2000) and validated following the procedure and criteria of the absence of pressures described in Pardo et al. (2012). Eight of the fourteen reference sites actually belong to the spatial network used by the Cantabrian Hydrographical Confederation (CHC) the Nalón River Water Management Agency (unpublished data). Test sites were selected covering rivers near historic (mercury and copper mines) and active (gold mine) mining industries, and thus subject to known inputs of heavy metals. Three of them were located in a copper mine area (CU group), 8 in mercury mine areas (HG group) and 4 in a gold mine area (AU group) (Fig. 1).

2.2. Data collection and laboratory processing

2.2.1. Biological data

Collection and processing of macroinvertebrate samples was done according to the Spanish official protocol ML-Rv-I (2013) (see Pardo et al., 2014). The sampling protocol followed a multihabitat procedure adapted from Barbour et al. (1999). At each study site, a total of twenty 'sample units' of 0.125 m² each (distributed proportionately in the main habitats existing along a 100 m reach) were collected using a kick-net (500 µm mesh size), and combined into a single mixed sample (a total sampled area of 2.5 m²). After collection, the samples were preserved with 70% ethanol and stored. All were washed and separated into three fractions using a series of sieves of different mesh sizes (5, 1 and 0.5 mm). The invertebrates were sorted in each fraction and subsamples taken in the 1 and 0.5 mm fractions only when necessary (Wrona et al., 1982). Taxon richness and abundance were evaluated at the family level (except for aquatic oligochaetes and Hydracarina) under 57× magnification (Olympus SZX9).

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