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Invertebrate community responses to emerging water pollutants in Iberian river basins

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HIGHLIGHTS

- Invertebrate density changes were related with emerging compounds and conductivity.
- Catalase enzymatic activity and feeding rates in invertebrates decreased downstream.
- Emerging compounds (EDCs, PhACs) were significant predictors of catalase activity.

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ABSTRACT

Chemical pollution is one of the greatest threats to freshwater ecosystems, especially in Mediterranean watersheds, characterized by periodical low flows that may exacerbate chemical exposure. Different groups of emerging pollutants have been detected in these basins during the last decade. This study aims to identify the relationships between the presence and levels of prioritary and emerging pollutants (pesticides, pharmaceutical active compounds—PhACs, Endocrine Disrupting Compounds EDCs and Perfluorinated Compounds—PFCs) and the invertebrate community in four Mediterranean basins: the Ebro, the Llobregat, the Júcar and the Guadalquivir. Structural (species composition and density) and functional (catalase activity of the tricopteran *Hydropsyche exocellata* and the feeding activity of the cladoceran *Daphnia magna*) variables were analyzed to determine which of the pollutants would greatly influence invertebrate responses. EDCs and conductivity, followed by PhACs, were the most important variables explaining the invertebrate density changes in the studied basins, showing a gradient of urban and industrial pollutions. Despite this general pattern observed in the four studied basins – impoverishment of species diversity and abundance change with pollution – some basins maintained certain differences. In the case of the Llobregat River, analgesics and anti-inflammatories were the significant pollutants explaining the invertebrate community distribution. In the Júcar River, fungicides were the main group of pollutants that were determining the structure of the invertebrate community. Functional biomarkers tended to decrease downstream in the four basins. Two groups of pollutants appeared to be significant predictors of the catalase activity in the model: EDCs and PhACs. This study provides evidence that the information given by functional biomarkers may complement the results found for the structural community descriptors, and allowed us to detect two emerging contaminant groups that are mainly affecting the invertebrate community in these basins.

1. Introduction

Freshwater ecosystems are affected by natural factors (e.g., climatic variability) and anthropogenic activities, and are specially overstressed in regions with a marked seasonality and intense demand for these resources. Almost all major human activities act as drivers of stress for

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natural habitats and biological communities. One main threat to river ecosystems is pollution. The accumulation of diffuse pollution (e.g., from agricultural activities) and point–source pollution (e.g., from urban areas) gives rise to a mixture of toxicants in fluvial systems that makes it difficult to attribute effects to any particular toxicant. To complete this scenario, a large number of previously unrecognized pollutants have been identified over the past few decades. These pollutants, usually called emerging contaminants (ECs), enter into rivers mainly through wastewater treatment plant (WWTP) effluents (Oller et al., 2011). Secondary treatment processes are insufficient to remove them completely, because they were not specially designed for that purpose (Tauxe-Wuersch et al., 2005; Gómez et al., 2007) and these pollutants become ubiquitous in the environment (Martínez Bueno et al., 2012). Endocrine disrupting compounds (EDCs), pharmaceutical active compounds (PhACs), personal care products, flame retardants and perfluorinated compounds (PFCs) are particularly relevant ECs in rivers. It is presently unclear how the presence of mixtures of pollutants with a different mode of action influences aquatic communities after long-term exposure. To shed light on this issue would help to better assess the environmental risk of such pollutants.

Mediterranean rivers are characterized by drought periods with low discharges and low flows that reduce the dilution capacity of these rivers, during which the concentrations of pollutants may significantly increase (Osorio et al., 2014). In addition, the inputs of the WWTPs can often be more important in terms of flow than the river by itself. Hence, there is a close and evident relationship between water quantity and chemical and biological measures of water quality in Mediterranean-climate rivers (López-Doval et al., 2013) which has to be taken into account to make appropriate water management decisions. In fact, the contamination levels reported in basins of the Iberian Peninsula are often higher than in other non-Mediterranean basins and have increased in past decades as a result of human activities (Petrovic et al., 2011). Therefore, there is a real chemical exposure risk not only for the fluvial biota but also for human and environmental health (Rovira et al., 2013; Carpenter et al., 2011).

Quality indices that describe the structure of communities (e.g., taxonomic richness and diversity) have been widely used as biomonitoring tools (Hering et al., 2006) to detect the effects of general pollution in the community. However, before these structural changes occur, pollutants first interact at a subcellular level and these interactions can lead to individual changes linked with important life functions (e.g., metabolism, reproduction, mobility or feeding). These sublethal responses segregate along gradients of both toxicological and ecological relevance and can lead to future community changes (Adams, 2003). Thus, they can be used as tools for the early detection of toxicity, especially for ECs whose effects are not well known.

Species respond to water quality changes using different detoxification mechanisms, for example, by increasing biotransformation enzymes and antioxidant defenses (Gillis et al., 2014). These biochemical responses can be used as biomarkers to detect the biological impacts of pollution in rivers. Hence, biomarker analyses may provide evidence of effects that are not detected at a structural community level, and can complement the information given by conventional studies of aquatic communities. Additionally, in situ bioassays offer greater relevance with respect to natural conditions and allow detecting effects in hours or days at the individual or population level (Maltby et al., 2002). The measured responses can be different across species and include mortality, feeding rates, growth rate and reproduction. These responses are directly related with key ecological processes that sustain ecosystem functioning and may represent a fertile methodology for the investigation of exposure to a mixture of pollutants.

This study aims to identify the relationships between the presence and concentration of common, prioritary and emerging pollutants (pesticides, PhACs, EDCs and PFCs) and the invertebrate community. The study was developed in four Mediterranean basins: the Ebro, the Llobregat, the Júcar and the Guadalquivir. Structural (species composition and density) and functional variables were analyzed to determine which of the pollutants would greatly influence invertebrate responses. The two functional biomarkers studied were the antioxidant enzymatic activity (catalase) of the tricopteran *Hydropsyche exocellata* Durfour 1841 and the inhibition of feeding activity in the cladoceran *Daphnia magna* Straus 1820.

2. Materials and methods

2.1. Study area and sampling

The study area involves four rivers of the Iberian Peninsula: the Ebro, the Llobregat, the Júcar and the Guadalquivir (Fig. 1), from north to south. These watersheds are highly populated and have important agricultural areas and industrial clusters that depend on the surface and groundwater resources and water transfers. Thus, they were selected because of their economic and environmental importance. The climate in these basins is Mediterranean, characterized by mild and moderately moist winters, warm, dry summers and irregular rainfall concentrated in the Spring and Autumn. Only the upper portion of the Ebro basin has a more continental climate.

The Ebro drains an area of 85,534 km^2 from north central to northeast Spain. It has a complex hydrologic regime because it receives water from tributaries with different climates. Despite its large size, it has a relatively small population, some 2.7 million inhabitants. The Ebro watershed is one of the most heavily irrigated regions in Spain, although recently the industrial sector has become increasingly important. Consequently, a broad spectrum of emerging contaminants has been found recently in the water and sediment (Navarro-Ortega et al., 2010).

The Llobregat River drains a 4957 km^2 catchment. It is subject to heavy anthropogenic pressure (4.5 million inhabitants in the valleys), receiving extensive urban and industrial wastewater discharges as well as surface runoff from agricultural areas that cannot be diluted by its natural flow, especially in the dry seasons. Consequently, the water has a high concentration of nutrients and priority and emerging pollutants.

The Júcar River drains a 21,632 km^2 catchment. Agriculture accounts for most of the water demand, but industrial and urban demands are increasing. The Júcar is highly regulated, and the management of the system is very complex, which leads to considerable hydrologic fluctuations (Paredes-Arquiola et al., 2010).

The Guadalquivir River basin is 57,527 km^2 and approximately 7000 km^2 of its basin is devoted to agriculture (rice, olives and fruits). As a consequence of a high population (7 million inhabitants), the river receives many inputs, from both natural and anthropogenic origins.

A total of 20 sites were selected in the main channel of the rivers: 5 in the Ebro (E1, E2, E3, E4 and E5), 5 in the Llobregat (L3, L4, L5, L6 and L7), 6 in the Júcar (J1, J2, J4, J5, J6 and J7) and 4 in the Guadalquivir (G1, G2, G3 and G4) (Fig. 1). Sites were selected along the rivers to represent pollution gradients. The sampling was performed in two consecutive years at the end of the dry summer period (a. Autumn 2010 and b. Autumn 2011). At each site, composite samples from water column and sediment (jointed sample of the uppermost 10 cm layer from the two river banks) were taken for chemical analyses. Five sediment samples randomly distributed were taken to determine community composition. *H. exocellata* individuals were collected in all sites where this taxon was found to measure catalase activity. *D. magna* enclosures to assess post-exposure feeding rates were deployed in the Ebro, the Llobregat and the Júcar rivers in 2011.

2.2. Chemical analyses

2.2.1. Physicochemical parameters

Dissolved oxygen, pH, temperature and conductivity of water were measured with Hach DO meters and WTW conductivity meter in each sampling site and campaign. Water samples for nutrient analyses