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Estuarine intertidal meiofauna and nematode communities as indicator of ecosystem's recovery following mitigation measures

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ABSTRACT

The Mondego estuary (Portugal) has been under environmental pressure since the early 1990s due to different anthropogenic stresses. The system has been studied following benthic communities' features from an impacted situation until the recovery phase, focusing mostly on macrobenthos. Following the application of mitigation measures in the estuary, this study is focused on the variability of the intertidal meiobenthic and nematode communities in a system that has recovered after different anthropogenic stresses. While at the spatial level (among areas along the eutrophication gradient) no significant differences were observed regarding the structure and function of the nematode communities, at the seasonal level some differences stood out. These results broadly suggest that the system has recovered from the early situations of pressures being, to the best of our knowledge, the first attempt to investigate the variability of intertidal meiobenthic and nematode communities in the scope of a system's recovery along an estuarine gradient of eutrophication. Even if performed in a short timeline, this study provides a good baseline analysis of conditions, being important for future comparisons.

1. Introduction

Estuaries are dynamic and productive systems (Kennish, 2002), being amongst the most valuable ecosystems in the world (Costanza et al., 1997). Besides supporting important ecological functions and services (*e.g.* biogeochemical cycling and movement of nutrients, water purification, flux regulation of water, particles and pollutants, shoreline protection) (Kennish, 2002; Meire et al., 2005; Paerl, 2006), resources provided by estuaries have been a target of human exploitation, compromising estuarine ecological integrity (Halpern et al., 2008; Borja et al., 2010). Furthermore, human induced impacts (including nutrient enrichment, chemical contamination, hydrological modification, habitat loss, among others; Kennish, 2002) and their negative effects on estuarine systems triggered the attention toward the need for monitoring, assessing and managing ecological integrity to promote the long-term sustainability of these systems (Borja et al., 2008).

Estuarine communities have to cope with the high variability in the physicochemical characteristics felt within these systems (Elliott and Quintino, 2007). However, besides this natural variability, anthropogenic pressures and their impacts are also felt in estuaries, increasing the difficulty in distinguishing a signal reflecting natural or anthropogenic change (Estuarine Quality Paradox) (Elliott and Quintino, 2007). Establishing relationships between species distribution and environmental characteristics is a major goal in the search for forces/causes driving species distribution (Peres-Neto et al., 2006) and the awareness of increasing pressures on aquatic systems enhanced the development and implementation of environmental policies worldwide, addressing the ecological quality or integrity within estuarine systems (Borja et al., 2008).

Regarding environmental assessments, good indicators are those that respond to natural gradients or disturbance at spatio-temporal scales appropriate to the study and faunal groups are deemed appropriate for this task (Schratzberger, 2012). Although macrobenthic invertebrates are favored as indicators in aquatic assessments over meiofauna (mainly due to well documented sampling protocols and taxonomic keys for macrobenthos), meiofauna are useful indicators in a variety of studies (their close association with the substrate, high diversity and importance

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in ecosystem functioning makes meiofauna a valuable tool for environmental assessments) (Heip et al., 1985; Sandulli and de Nicola, 1991; Kennedy and Jacoby, 1999; Schratzberger et al., 2000; Moreno et al., 2008). Assessment of the meiofaunal (mostly nematodes) communities of estuaries often relate their horizontal distribution at different scales (from small to global scale) with the complex interaction between biotic (food source distribution, competition among species) (Montagna et al., 1983; Gallucci et al., 2008) and abiotic factors (variations in the physicochemical properties of the sediment matrix, salinity and tidal exposure) (Heip et al., 1985; Steyaert et al., 2001, 2003; Ferrero et al., 2008). Moreover, human disturbances affecting the physical structure of the sediment and food availability, as well as pollution impacts on nematode communities have been documented (Coull and Chandler, 1992; Schratzberger and Warwick, 1999; Schratzberger et al., 2000, 2002), reinforcing nematode communities as highly informative and useful in efficiently evaluate the ecological status of aquatic bodies (Moreno et al., 2011).

The Mondego estuary (Portugal), a south-western European transitional system, underwent intense anthropogenic pressure over the last decades, contributing to an overall decline in its environmental quality (further description in Section 2). Following a management measure in the spring of 2006 (Veríssimo et al., 2012a, 2012b) designed to improve the overall system ecological condition by contributing to the total re-establishment of the upstream connection between the two arms of the estuary), it was created the opportunity to assess and compare the system new ecological quality status with the previous eutrophication state, and studies relating these conditions were especially performed for macrobenthic communities (Veríssimo et al., 2012a, 2012b; Marques et al., 2013).

Regarding meiofauna and nematode communities, data previous to the intervention are not available. However, due to the extensive knowledge regarding the system evolution in the south arm of the Mondego estuary (spatial gradient of eutrophication – Marques et al. (1997); see Section 2), the analysis of meiofauna communities' succession can give new insights about the system recovery. Following a gradient of *Zostera noltii* coverage, this study has as main goals (i) the analysis of changes in intertidal meiofaunal communities, especially free-living nematodes, along an eutrophication/recovery gradient, (ii) the identification of relations between the obtained distribution patterns and the physicochemical environment, and (iii) the interpretation and integration of the results considering the evolution (recovery) of the system, in order to understand how nematode communities reflect the impacts. We hypothesized that (i) meiofauna and nematode communities will have higher diversity and abundance in the area dominated by *Z. noltii*, and that (ii) the differences between areas can be attributed to the different pressures suffered during time at the different areas.

2. Materials and methods

2.1. Study area

The Mondego estuary, located in the western coast of Portugal (40°08′ N, 8°50′ W) (Fig. 1), is a mesotidal system influenced by a warm-temperate climate. In its terminal part this 21 km long estuary consists of two arms – north and south – separated by an alluvial island, and join again in the estuary mouth. The two arms present different hydrological characteristics (Marques et al., 1993, 2003): the south arm is shallower (2–4 m during high tide), covered by large areas of intertidal mudflats (almost 75% of the area) exposed during low tide (Neto et al., 2008); the north arm is deeper (5–10 m during high tide), receives most of the system's freshwater input and constitutes the main navigation channel supporting the Figueira da Foz harbor. The estuary supports several industries, salt-works, agricultural areas, mercantile and fishing harbors, thus having various anthropogenic pressures (Marques et al., 1993; Flindt et al., 1997).

The estuary has suffered several physical modifications over the years (see Neto et al., 2010, for a complete description of the estuary's modifications) and both the river bed topography and the system hydrodynamics were altered, leading to the interruption of the communication between the two arms in the early 1990s (Marques et al., 1997, 2003; Neto et al., 2010) with severe impacts on the south arm. In this subsystem, the increase in water residence time and nutrient concentration promoted eutrophication symptoms and the deterioration of the environmental quality (Marques et al., 2003). A gradual shift in primary producers from a community dominated by rooted macrophytes (*Z. noltii*) to a community dominated by green macroalgae (mostly *Ulva* spp.) was observed (Marques et al., 2003), leading to a reduction in the *Z. noltii* coverage area (Martins et al., 2005) and to a shift in benthic primary producers, affecting the structure and functioning of the biological communities (Marques et al., 1997, 2003; Martins et al., 2005; Patrício and Marques, 2006).

After the mitigation measures implemented to improve the system ecological condition in 1998 (the discharge of freshwater from the Pranto River decreased and the communication between the two arms was re-established) the system underwent partial improvements in its environmental quality (Teixeira et al., 2008; Cardoso et al., 2010), with a recovery of the *Z. noltii* meadow and a cessation of the macroalgae blooms (Martins et al., 2005; Dolbeth et al., 2007; Patrício et al., 2009).

The recovery of the system allowed the identification of the high residence time as a cause for the ecological degradation in the south arm and suggested that the efficient renewal of water in this subsystem would increase the flow and load capacity of the water mass, which encouraged a complete re-establishment of the communication between both arms by the spring of 2006, decided at the Portuguese government level (Veríssimo et al., 2012a). The upstream connection between the two arms was enlarged and the hydraulic regime fully re-established (Veríssimo et al., 2012b). This investigation focuses on periods after the intervention.

2.2. Sampling strategy and laboratory procedures

Sampling was conducted during low tide on three campaigns (September 2009, December 2009 and March 2010) in four intertidal areas of the south arm of the Mondego estuary, representing different environmental situations along a spatial gradient of eutrophication (Marques et al., 1997, 2003; Patrício and Marques, 2006) and with a gradient of coverage by *Z. noltii*: (a) a non-eutrophic area located downstream, where *Z. noltii* predominates, and considered the richest area of the estuary in terms of productivity and biodiversity (Marques et al., 1993; Dolbeth et al., 2007); (b) an intermediate eutrophic area (*Z. noltii* absent, although residual roots can be found and campaignal formation of macroalgae mats observed); (c) a bare sediment area in the inner part of the estuary where eutrophication processes occurred in the estuary (macrophyte community absent, regularly occurring blooms of *Ulva* spp.), currently characterized by a few, small and irregularly distributed *Z. noltii* patches (Veríssimo et al., 2013); and (d) a bare sediment area located further upstream adjacent to the intervention area, with higher freshwater influence; hereafter referred as "Zostera", "Intermedia", "Armazens" and "Montante", respectively (Fig. 1).

2.2.1. Environmental variables

Bottom water variables were measured *in situ* at each area using an YSI Data Sonde Survey 4: salinity, pH, temperature (°C) and dissolved oxygen (mg L^{-1}). Additionally, water samples were collected