



# Indices, multispecies and synthesis descriptors in benthic assessments: Intertidal organic enrichment from oyster farming

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## ABSTRACT

Intertidal off-bottom oyster culture is shown to cause organic enrichment of the shore and although there are two stressors of interest (the presence of a structure, the trestles, and also the sediment and organic waste from the oysters), these can be separated and their relative impacts determined using an appropriate nested experimental design and data treatments. Although no artificial food sources are involved, the oysters feeding activity and intensity of culture enhances biodeposition and significantly increases the sediment fines content and total organic matter. This in general impoverished the benthic community in culture areas rather than a species succession with the installation of opportunists or a resulting increase in the abundance and biomass of benthic species; the findings can be a direct consequence of the intertidal situation which is less-amenable recruitment of species more common to the subtidal environment. Thus the most appropriate biological descriptors to diagnose the effects associated with the organic enrichment were the multispecies abundance data as well as the primary biological variables species richness and abundance. The effects were however spatially and statistically significantly confined to the area located directly underneath the culture bags compared to the corridors located between the trestles, which do not show such enrichment effects. Synthesis biotic indices were much less effective to diagnose the benthic alterations associated with this organic enrichment. These results show that special attention must be paid when using indices in areas where the organic enrichment induces an impoverishment of the benthic community but not necessarily a species replacement with the installation of opportunists.

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## 1. Introduction

Increasing anthropogenic pressures in coastal and transitional waters have resulted in protective legislation; for example, in the European Union, the Water Framework Directive (WFD, 2000) establishes measures to protect surface waters, aiming to reach good ecological quality status. In order to distinguish levels of ecological quality and classify coastal and estuarine areas, new biotic indices have been developed. For the benthic macrofauna, these indices are usually based on the Pearson and Rosenberg (1978) paradigm of species succession in response to organic enrichment of the seabed. Indices can be valuable in simplifying the complexity of scientific data and producing results relevant for management (Quintino et al., 2006; Chainho et al., 2007; Borja and Tunberg, 2011) but for an index to be of general value as an

ecological indicator it should be applicable irrespective of the geographical area (Salas et al., 2004; Gray and Elliott, 2009). Among the possible limitations concerning such a widespread application, are the differences between biogeographic provinces, between intertidal and subtidal habitats and between hard and soft substrata (Borja and Dauer, 2008). Also, it is more difficult to detect the effects of anthropogenic stress and disturbance in naturally stressed areas, such as estuaries (Elliott and Quintino, 2007). It is hypothesised here that the same reasoning should apply to intertidal areas, under natural stress from tidal exposure. In addition, as most anthropogenic marine discharges are subtidal, very few situations exist where such indicators can be adequately tested in the intertidal area. Hence this work was developed in an intertidal estuarine area licenced for off-bottom oyster aquaculture. The organic enrichment associated with this type of aquaculture has been recently reported (Mallet et al., 2006; Bouchet and Sauriau, 2008; Forrest et al., 2009), but there is no general consensus on the severity its effects, which often are site-specific. Most studies

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on oyster culture indicate potential negative ecological impacts due to the production of faecal and pseudo-faecal biodeposits where fluxes of organic matter are enhanced beneath the cultures. The trestles that support the cultures in intertidal soft bottom areas may also contribute to sediment deposition by reducing water flow (Kervella et al., 2010). The biodeposits may alter physical and chemical properties of the water column (Dame et al., 1989) but their main effect is on the seabed due to increasing sediment fines (Crawford et al., 2003), organic enrichment (Grant et al., 1995; Chamberlain et al., 2001) and the modification of sediment geochemistry which can produce anoxic conditions (Cranford et al., 2009; Gray and Elliott, 2009). These affect the benthic communities (Forrest and Creese, 2006; Bouchet and Sauriau, 2008; Callier et al., 2009) although most benthic community studies addressing the impacts of oyster farming were performed sublittorally (e.g. Crawford et al., 2003). Fewer studies focused on impacts of intertidal oyster cultures (e.g., De Grave et al., 1998) and revealed an increased sedimentation and organic enrichment beneath the cultures (Forrest and Creese, 2006; Mallet et al., 2006; Bouchet and Sauriau, 2008). Organic enrichment, leading to hypoxia, produces severe structural changes in the benthic community, as expressed in the species-abundance-biomass (SAB) conceptual model (Pearson and Rosenberg, 1978).

This study hypothesized firstly, that intertidal off-bottom oyster culture causes detectable organic enrichment and benthic community effects, assessed at a multispecies level as well as on single biological variables and structural biotic indicators. Secondly, it tested whether a hydrodynamically more energetic and biologically more variable area such as intertidal flats had a greater resistance to stress. If this is the case then it may confer a greater resilience to recover from any imposed change (following the recovery paradigm model proposed by Elliott et al., 2007 and tested by Borja et al., 2010).

## 2. Methods

### 2.1. Study area and experimental design

The study site in the Ria de Aveiro, a coastal lagoon in NW Portugal, includes many channels and intertidal sand and mudflats and salt marshes. Semi-diurnal tides are the main forcing water circulation agent with a mean tidal range of ca. 2 m and minimum and maximum ranges of about 0.6 m and 3.2 m, at neap and spring tide respectively (Dias et al., 2000). The benthic community distribution is strongly influenced by the hydrodynamics and salinity gradient (Rodrigues et al., 2011). The most abundant and frequent taxa are annelids but other bivalves are an important economical resource, including the production on natural banks and in licenced areas.

This study was conducted in an 8000 m<sup>2</sup> (100 × 80 m) intertidal site licenced for oyster culture in the Mira channel, a 20 km long relatively pristine, shallow channel (Castro et al., 2006), running southwards from the Ria entrance, with salinity conditions from fully marine to freshwater (Quintino et al., 2009). Juvenile oysters are grown inside plastic mesh bags, placed on top of trestles at ca. 70 cm above the ground. The experimental design set oyster exploitation as the main fixed factor, with the reference levels of bare sediment, trestles-only and trestles with oysters. The experimental design allowed us to subdivide the site into eight areas (A to H, Fig. 1): areas B and C have been used for oyster culture, areas E and F have trestles-only and areas A, D, G and H remain unused and have no trestles (reference areas). The eight areas are not located at the same beach height. Areas A and E are designated as the upper shore areas, whereas D and H are designated the lower shore areas (see Fig. 1b). Areas B and C were populated with small and large

oysters respectively, ca. 4 and 8 cm long. In each area, four sampling sites were randomly positioned and at each site four replicate sediment samples were collected, three for the study of benthic macrofauna and the other for sediment grain-size and total organic matter content. In the areas B, C, E and F, with trestles, the sediment samples were taken inside the trestles, i.e. in the cases of areas B and C directly under the oyster bags. In area B, with the smaller oysters, four extra sampling sites were also placed in the corridors between the trestles, away from the direct influence of the oyster faecal pellets and pseudo-faeces. This set of samples is identified as B<sub>0</sub> throughout the results section (see Fig. 1b and c). All samples were collected at low tide with a 0.01 m<sup>2</sup> hand held corer. The benthic fauna samples were sieved through a 1 mm mesh screen. This experimental design allowed the testing of several null hypotheses, namely there are no effects associated with the oyster culture, there are no effects associated with the trestles alone and, in the case of the area with the smaller oysters, that there are no significant differences between samples obtained beneath the oyster bags and in the corridors between the trestles.

### 2.2. Laboratory analysis

Sediment grain-size was determined by wet and dry sieving (Quintino et al., 1989) and the fines fraction is expressed as a percentage of the total sediment, dry weight <0.063 mm. Sand, particles of diameter 0.063–2 mm, and gravel, particles with diameter >2 mm, were dry sieved at 1 $\phi$  intervals ( $\phi = -\log_2$  the particle diameter expressed in mm). Total organic matter content was obtained by percentage loss-on-ignition of 1 g of dry sediment at 450 °C for 5 h (Byers et al., 1978). After hand-sorting, the macrofauna were identified to the highest possible taxonomic separation (usually species).

### 2.3. Data analysis

The sediment descriptors included 7 grain-size fractions (>2 mm; 1–2; 0.5–1; 0.25–0.5; 0.125–0.25; 0.063–0.125; <0.063 mm, used to calculate the median diameter, P<sub>50</sub>, expressed in phi ( $\phi$ ) units) and the total organic matter content. The median was used to classify the sediment, according to the Wentworth scale: gravel [(-2) – (-1) $\phi$ ]; very coarse sand [(-1) – 0 $\phi$ ]; coarse sand (0 – 1 $\phi$ ); medium sand (1 – 2 $\phi$ ); fine sand (2 – 3 $\phi$ ); very fine sand (3 – 4 $\phi$ ). Sands were classified as “clean”, “silty” or “very silty” when the fines fraction ranged from 0% to 5%, from 5% to 25% and from 25% to 50%, respectively. Samples with >50% fines content were classified as mud.

Macrofauna community descriptors included the taxon composition (as a taxon list), primary biological variables (species richness (S), abundance (A) and wet-weight biomass (B) per replicate), and derived/secondary biotic indices (Gray and Elliott, 2009) were also calculated, per replicate: Shannon–Wiener diversity (H') (Shannon and Weaver, 1949); Margalef richness (d) (Margalef, 1968); Simpson index (1- $\lambda'$ ) (Simpson, 1949), the AZTI marine biotic index (AMBI, Borja et al., 2000) and its multivariate derivative (M-AMBI, Muxika et al., 2007).

The data were analysed using principal coordinates ordination (PCO), upon inter-replicate resemblance matrices using the normalised Euclidean distance (environmental data) or the Bray–Curtis similarity upon the square-root transformed abundance (biological data). The biological and the sediment data were further submitted to hypothesis testing using permutation multivariate analysis of variance (Anderson, 2001). The biological data, multivariate for the species abundance data or univariate for the individual primary variables and derived indices, were analysed according to a three-way hierarchical design. Oyster exploitation

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