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Defining soft bottom habitats and potential indicator species as tools for monitoring coastal systems: A case study in a subtropical bay

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ABSTRACT

The definition of habitats and indicator species is a prerequisite for monitoring and conservation programs. Nonetheless, defining habitats in marine soft-bottom environments is challenging given their spatiotemporal dynamics and apparent homogeneity. The selection of indicator species is also complicated given the large number of occasional species usually presented in benthic communities. This study aims to elaborate a framework based on well-established analytical methodologies to identify soft-bottom habitats and select indicator species to support monitoring and conservation programs. The proposed framework consists of four steps: 1) perform a Redundancy Analysis (RDA) on the community data to identify the community structure response to environmental gradients; 2) conduct a kernel density analysis on the RDA biplot to determine the habitats; 3) use the indicator values analyses (IndVal) to select indicator species of each habitat; 4) run polynomial quantile regression analysis to find the optimum distribution of each indicator species. Such framework allows the determination of habitats based on the association of environmental and biological datasets, instead of relying solely on abiotic surrogates. As a case study, we used data of macro and meiofauna of a biodiverse coastal ecosystem in Southeast Brazil which is under anthropogenic pressure. Three main habitats were identified in the bay, and macro and meiofaunal assemblages were influenced by similar environmental variables. Nevertheless, macrofauna was more sensitive to changes in sediment composition, whereas meiofauna responded strongly to changes in total organic content and water depth. Macro- and meiofauna indicator taxa showed high specificity and fidelity values to each habitat, supporting their use in monitoring and conservation programs. The spatio-temporal organization of each habitat and the optimum distribution of each indicator species provide baseline knowledge to be used to monitor environmental changes in the study area and help in its conservation.

1. Introduction

All marine ecosystems are to some extent currently affected by human activities (Halpern et al., 2008), and it is estimated that almost half of the marine environment is already impacted by a combination of stressors such as ocean acidification, coastal hypoxia, and pollution (Gray, 2002; Defeo et al., 2009; Halpern et al., 2015). This unprecedented level of anthropogenic threats to marine systems has increased the need for biomonitoring and conservation programs (Crain et al., 2008; Halpern et al., 2008; Stelzenmüller et al., 2010).

The success of conservation efforts is highly dependent on the

identification and protection of natural habitats which can act as biodiversity reservoirs and are important to ecosystem functioning and stability (Stevens and Connolly, 2004; Cogan et al., 2009). Once defined, a habitat can be used to plan monitoring programs. So far, the definition of habitats in marine benthic ecosystems usually relies on physical attributes and biogenic structures such as seagrass, rocky shores, and mussel beds (Banks and Skilleter, 2002; Seitz et al., 2014). Defining such habitats, however, is particularly challenging in highly dynamic and apparently homogeneous systems such as marine soft-bottoms (McArthur et al., 2010).

The definition of habitats in marine soft-bottoms is usually linked to

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the less conspicuous variation in sediment properties (e.g., mud content, pebbles, and sorting coefficient) (Gray and Elliot, 2009). Such classification is normally done *a posteriori* to the data acquisition and based only on the environmental conditions. Nevertheless, the use of abiotic surrogates alone to map coastal habitats may generate unreliable results (Diaz et al., 2004; Stevens and Connolly, 2004). The organisms inhabiting the matrix of sediments exhibit complex interactions with the environmental characteristics and greatly influence the habitats conditions (McArthur et al., 2010). The consideration of such complex species-environment interaction is therefore crucial for properly delimiting a habitat (Diaz et al., 2004).

A complementary method to the habitat-based approach in conservation programs is the selection of indicator species (Carignan and Villard, 2002; De Cáceres et al., 2010; Siddig et al., 2016). These species show predictable responses to environmental conditions and can be used to assess the habitat conditions (Dufrene and Legendre, 1997; Carignan and Villard, 2002; Niemi and McDonald, 2004; Siddig et al., 2016). An appropriate indicator species has to respond strongly to a particular group of conditions, to which it will serve as an indicator (De Cáceres et al., 2010; Fonseca and Gallucci, 2016). The selection of indicator species for soft benthic communities is particularly important since the sedimentary habitat is dynamic, the number of species is high, and the identification of benthic biodiversity to species level is a major time-consuming activity (Warwick, 1993). On top of that, it is important to understand the optimum environmental conditions of each indicator species, in an attempt to facilitate monitoring programs (Anderson, 2008; Fonseca and Gallucci, 2016).

The aim of this study is to suggest a methodological framework to be used in monitoring and conservation programs of soft-bottom ecosystems. Using well established statistical analyses, we first identify potential habitats based on the responses of species assemblages to the environmental characteristics. Then, we select possible indicator species which can be used to assess future changes in each habitat. We apply this methodological framework to the main groups of marine benthic fauna (meio- and macrofauna) in a biodiverse benthic ecosystem which is under recent threats due to the planned expansion of the neighboring port (Amaral et al., 2010, 2016). These threats reinforce the critical need to recognize and understand the local environmental dynamics to monitor and manage the area. We use both assemblages to better comprehend the benthic environment and to test the framework on the two groups most used in benthic monitoring studies (Semprucci and Balsamo, 2012; Bessa et al., 2014; Gorman et al., 2017). The study area is also a typical example of many vulnerable parts of the Brazilian coast, and as such, the outcomes of the present study are relevant for other regions.

2. Materials and methods

2.1. Study area

This work was done at Araçá Bay (23° 49'S, 45° 24'W), a coastal ecosystem (~500.000 m²) located in the central area of the São Sebastião Channel, state of São Paulo, Southeast Brazil (Fig. 1). The area is environmentally heterogeneous, with many distinct features such as patches of different sedimentary textures, mangroves and rocky shores (Amaral et al., 2016; Checon et al., 2017). The intertidal area has a gentle slope, with a maximum depth of 5 m, while further the bay reaches 30 m deep towards the channel. Araçá Bay is located within the Marine Environmental Protection Area of the Northern Litoral (APA Marinha do Litoral Norte), a conservation unit which aims to preserve biodiversity and natural processes, and is recognized as one of the areas with the highest marine biodiversity on the Brazilian Coast (Amaral et al., 2010, 2016).

2.2. Sampling design

Sampling was performed during four periods (October 2012, February, June and September of 2013). Thirty-seven sampling stations were determined from the intertidal and shallow sublittoral area at the bay (< 5 m deep) to a depth of 25 m (São Sebastião Channel). Sampling stations were positioned to a) encompass habitat diversity (i.e., different sediment types and depths), and b) achieve a reasonable dispersion and spatial coverage (Fig. 1). The same locations (± 1 m) were sampled during each period using a GPS for orientation of sampling stations positions. Sampling was done manually at the intertidal and shallow sublittoral (< 3 m deep), and with the use of a multi-corer sampler for deeper sites. At each sampling site, four samples were collected using a corer of 10 cm diameter and 20 cm depth (0.03 m²) for the evaluation of macrofauna, and one sample of 2.5 cm in diameter and 5 cm depth (19.6 cm²) for meiofauna. Particularly for macrofauna, the total area sampled in each sampling station is smaller than the 0.1 m² commonly employed in sublittoral studies (e.g. Petersen, box-corer, vanVeen) (Eleftheriou and Moore, 2013), which may increase the effect of patchiness and sampling heterogeneity. However, we chose to collect all samples with a corer to obtain fully quantitative replicates, as well as to standardize sampling areas between the intertidal and sublittoral zones.

Additional samples were taken at each station to evaluate environmental parameters: Five samples of the top 1 cm of the sediment were taken using a corer measuring 2 cm in diameter to evaluate microphytobenthic biomass; and one sample of sediment was taken for granulometric analysis using a corer of 3 cm diameter and 20 cm depth.

2.3. Sample processing

Macrofauna samples were stored in plastic bags and posteriorly sieved with a 0.3 mm mesh. The fauna retained was sorted in taxonomic groups and fixed in 70% ethanol. All individuals were identified to the species level.

Meiofauna samples were immediately fixed in 4% formaldehyde, and posteriorly washed through a 45 μ m mesh sieve and extracted by flotation with Ludox TM 50 (specific density 1.18) (Heip et al., 1985). The retained material was stored in formaldehyde 4% and stained with Rose bengal. Meiofauna counting and identification was done under a stereomicroscope. We selected only the nematode assemblage for further study, as they were the most abundant in the area. Nematodes were identified to genus level and further separated into morphospecies. From each sample, a total of 100 nematodes were randomly chosen, evaporated slowly in anhydrous glycerol and mounted on permanent slides for identification.

Microphytobenthic biomass was estimated from phaeopigments and chlorophyll *a* concentration according to Plante-Cuny (1973). Margalef pigment diversity index (Margalef, 1968), a ratio of total green pigments, was calculated. The index ranges from 2 to 8, increasing from young microphytobenthic communities to mature, oligotrophic ones. The granulometric analysis was carried out using the routine sieving and pipetting techniques described by Suguio (1973) and sediment parameters were obtained using SysGran software, version 3.0 (Camargo, 2006) following the classifications of Folk and Ward (1957). Total organic carbon was evaluated using a modified Walkley-Black titration method, described by Gaudette et al. (1974).

2.4. Data analysis

We combined established statistical techniques to set up a framework to be used in studies that aim to characterize and define benthic habitats, and provide information to be applied in monitoring programs. The framework proposes a stepwise procedure, as shown in Fig. 2. Seven sites were excluded from analysis due to missing variables, resulting on a total of 141 sites.

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