Chapter

Response of Benthic Foraminifera to Environmental Variability: Importance of Benthic Foraminifera in Monitoring Studies

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Abstract

Foraminifera are eukaryotic unicellular microorganisms inhabiting all marine environments. The study of these protists has huge potential implications and benefits. They are good indicators of global change and are also promising indicators of the environmental health of marine ecosystems. Nevertheless, much remains to be learned about foraminiferal ecology. The goals of this chapter are (1) to provide a few examples from foraminifera studies, presenting possible use of foraminifera as bioindicators for the monitoring of transitional and marine ecosystems and (2) to highlight the importance of applying these organisms in environmental monitoring studies. A semienclosed coastal lagoon (Aveiro Lagoon; Portugal), an estuarine system (São Sebastião Channel; SE Brazil), a continental shelf sector (Campos Basin; SE Brazil), and a segment of continental slope (Campos Basin; SE Brazil) are used as examples.

Keywords: foraminifera, marine pollution, marine ecosystems, environmental stress, bioindicators

1. Introduction

The anthropic activity has been causing more and more negative effects on nature, among which includes the discharge of several types of pollutants from either domestic or industrial sources. The final destination for a majority of the pollutants are frequently the coastal areas where the pollutants may cause deleterious effects as harm to living resources and marine life, hazards to human health, hindrance to marine activities, including fishing and other legitimate uses of the sea [1]. Persistent pollutants like polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), pesticides (dichloro diphenyl trichloroethane), toxic metals (Cd, Hg, Ag, Co, Cr, Ni, Pb, Zn, and Cu), and others may cause deleterious effect in marine life. The pollutants can enter the food chain and may even reach the highest trophic levels [2]. As microorganisms are among the lowest levels of the food chains, they are the first to denounce the negative effects of pollution. Among the vast variety of microorganisms, foraminifera have shown potential for effective pollution biomonitoring, apart from many other applications [1].

These unicellular microorganisms are effective environmental indicators as they respond quickly to small environmental changes. Foraminifera are abundant and preserve the changes in their tests, making possible to study even the past environmental standing. Foraminifera are widely distributed in marine environments. In fact, they have been successful inhabitants from deep oceans to brackish water lagoons, estuaries, and even rarely in freshwater streams, lakes, and so on [3].

Foraminifera are excellent bioindicators of environmental changes resulting from both natural processes and by human interference. Most of the recent studies carried out with foraminifera are focused on the application of these organisms as environmental bioindicators. Most baseline studies arrived from natural distribution patterns like those reported in the classical literature by [4–6]. After this first step, the detection of patterns associated with natural disturbances or pollution related to human activities can be carried on [7–10]. The environmental studies using foraminifera were started by the end of the 1950s. In a study performed in Santa Monica Bay, California, Zalesny [11] stated that environmental factors such as currents activity, nutrients, salinity, characteristics of bottom sediments, and especially temperature should control the distribution patterns of the living benthic species in that bay. Since then, the number of works aiming to study the response of benthic foraminifera to environmental changing, namely to pollution, increased significantly [12].

Foraminifera have been applied to study areas with different kind of pollutants and in different kind of environments including heavily polluted harbors, such as French coast, Rio de Janeiro, Montevideo, and Eastern Sicily. Armynot du Châtelet et al. [10], Debenay et al. [13], Vilela et al. [14], and Burone et al. [15] observed that foraminifera distribution in the harbor of Port Joinville, at the Atlantic French coast, was influenced by the significant increase of pollution by metals such as Cu, Pb, and Zn and sediment texture. In the most polluted areas, they observed the increasing abundance of pollution-tolerant species. Armynot du Châtelet et al. [10] observed that foraminifera density and diversity were negatively correlated with heavy metal and PAHs in four moderately polluted harbors of the French coast.

Romano et al. [16] studying the effect of heavy pollution mainly due to Hg, PAHs, and PCBs on the foraminiferal assemblages from the Augusta harbor (Eastern Sicily, Italy) observed that the clearest response of foraminifera to environmental degradation was the increased percentages of abnormal specimens exceeding the background, the increase of pollution-tolerant species, and the reduced size of the specimens.

Furthermore, they have been used as suitable bioindicators owing to their immediate response to the environmental changes such as hydrodynamic [17, 18], salinity [19, 20], pH [21], Eh [22], heavy metals [13, 15, 18, 22, 23], hydrocarbon pollution [10, 23], and organic matter [8, 24].

The main goal of this chapter is to provide information about environmental factors that may influence the patterns of distribution of living benthic foraminifera in transitional and marine ecosystems. A semienclosed coastal lagoon (Aveiro Lagoon, Portugal), an estuarine system (São Sebastião Channel, SE Brazil), a continental shelf sector (Campos Basin, SE Brazil), and a segment of continental slope (Campos Basin, SE Brazil) are used as examples. Each area has different particularities that condition the type of living foraminifera associations that occur in them, being the first two areas highly anthropized. This work also emphasizes the importance of these organisms as environmental bioindicators and their application in biomonitoring studies.

2. Case study: Aveiro Lagoon (Portugal)

The Aveiro Lagoon is located on the Western Atlantic coast of Portugal (40°38′N, 8°45′W, **Figure 1**). It is 45 km long and 10 km wide and covers an area between 66 and 83 km^2 at low and high spring tides, respectively [25]. It is composed by a series of channels among intertidal areas such as mudflats, salt marsh, and old saltpans. It is connected to the ocean through a single artificial inlet (~350 m wide and 2 km long) fixed by two jetties (**Figure 1**). Four main channels radiate of the lagoon mouth: Mira, S. Jacinto/Ovar, Ílhavo, and Espinheiro channels (**Figure 1**), with lengths of 20, 29, 15, and 17 km, respectively. The inner lagoonal area receives the contribution of several rivers. The freshwater is supplied mainly by Vouga and Antuã rivers with average flows over 50 and 5 $m^3\ s^{-1}$, respectively [26] and in less way by small rivers such as Boco river, which flows into the south Ílhavo channel, and the Caster river, which flows into the north of Ovar channel, with an

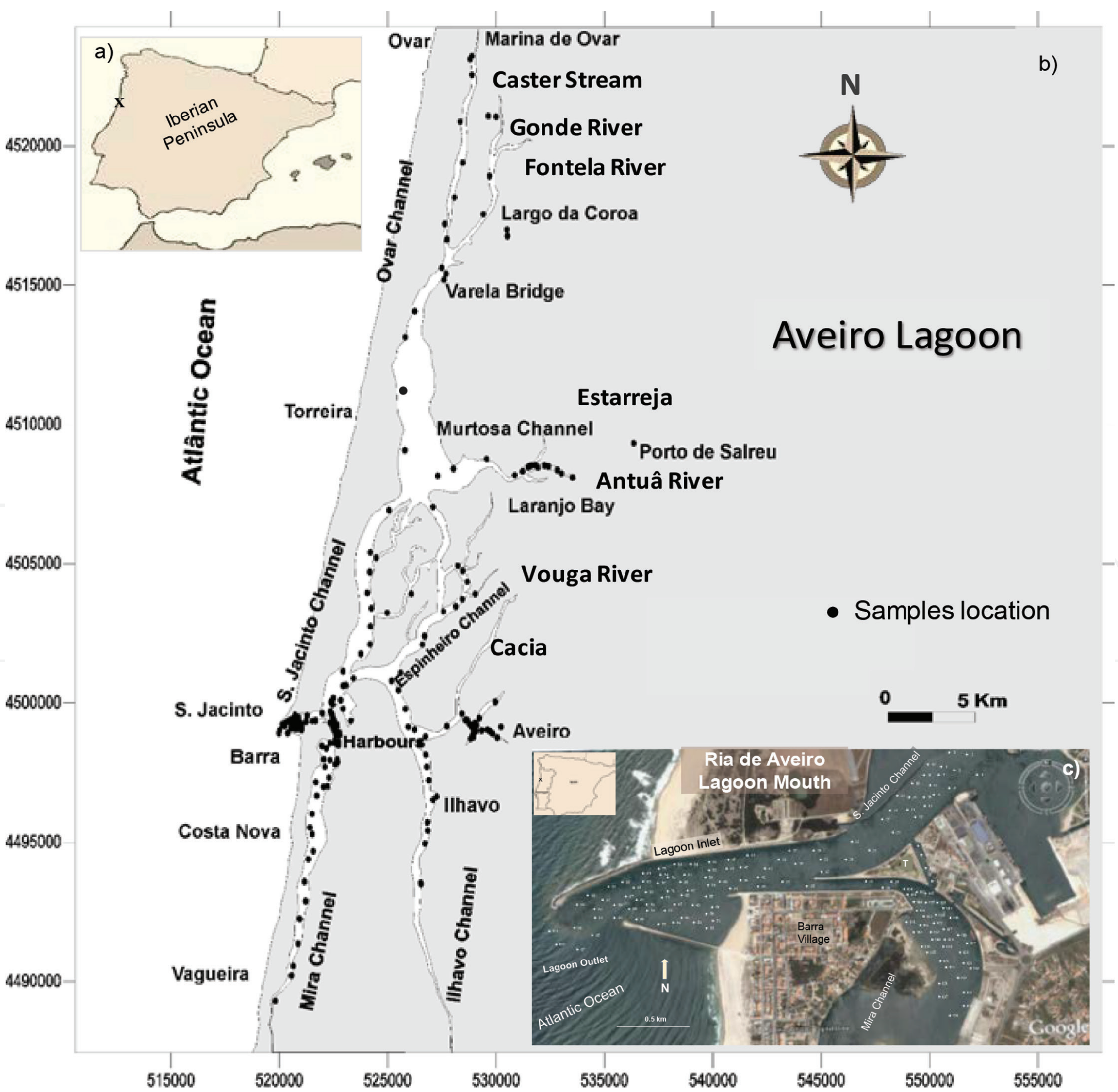

Figure 1.
(a) Aveiro Lagoon location; (b) samples location in Aveiro Lagoon; (c) a detail of the lagoon mouth (adapted from Google Earth).

average flow less than 1 $m^3 s^{-1}$ [25]. According to Dias et al. [27], each of the main channels may be considered as presenting features of separate estuaries, owing to typical estuarine longitudinal gradients of water salinity and temperature, with values close to the characteristics of the oceanic water near the inlet and close to freshwater furthest upstream.

The average depth of the lagoon is approximately 1 m relative to the local chart datum (2.0 m below mean sea level) [27] although there are deeper areas close to the lagoon mouth, where depths may reach values of ≈30 m. Nevertheless, the bathymetry in this zone has changed over time due to both natural and anthropogenic causes [28].

The Ria de Aveiro is a tide-dominated lagoon with minimum tidal range of 0.6 m (neap tides) and maximum tidal range of 3.2 m (spring tides). Near the lagoon entrance, tidal current velocities are strong [29], but they weaken in more internal lagoon areas. Furthermore, the noticeable wind effect can have an important influence on the lagoon circulation. Particular circulation patterns mainly in shallow areas and wide channels can be induced by strong winds [25].

Regarding the environmental quality of the lagoon, in internal areas near the river mouth, there are high concentrations of metals and organic matter [18]. The Laranjo Bay area (**Figure 1**) is also polluted owing to release wastewaters of plants of the chemical complex of Estarreja [30].

2.1 Methods applied in Aveiro Lagoon

The case study of Aveiro Lagoon, commonly known as Ria de Aveiro, was based on the analysis of geochemical, sedimentological, and environmental parameters combined with living benthic foraminifera. Environmental parameters and some foraminiferal data were based on Martins et al. [18, 31, 32]. The sediments were collected, in Aveiro Lagoon channels, in 255 stations, in 2006/2007 with an adapted Petit Ponar Grab sampler (opening at both extremities), and using a ZOE I boat (**Figure 1**), they were analyzed. Water depth was measured with the boat sonar, and the stations were located with a GPS. The upper sediment layer (about 1 cm) was scraped for textural, geochemical, and microfaunal (living benthic foraminifera) analyses at each site. The sediments sampled for geochemical analysis were immediately cool preserved on board. The samples collected for foraminifera studies were kept in alcohol (90%) stained with Rose Bengal (2 g of Rose Bengal in 1000 ml of alcohol). Rose Bengal was used to differentiate between living and dead foraminifera [33]. Temperature, salinity, pH, and potential redox (Eh) were measured in water and sediments in each site.

Samples for grain size and geochemical analysis were dried to constant weight in an oven for about 72 h, at 45°C, and stored for subsequent analysis. The procedures used for sedimentological and geochemical analyses are described in detail in Martins et al. [18, 31, 32]. The description of the foraminiferal analysis can be observed in Martins et al. [18, 31, 32]. The number of species per sample (S) and the diversity index of Shannon (H′) [34] were determined. The equitability also was determined according to Pielou [35] and S is the total number of species in a sample [36].

2.2 Results obtained in Aveiro Lagoon

2.2.1 Abiotic data

Sedimentary samples were collected at water depths varying between 0.5 and 30 m. Water temperatures varied between 10.5 and 26.0°C and salinity from 6.2 to 33.7. Higher temperatures were recorded in the innermost part of the main channels and salinities near the lagoon mouth and in the channels with strong marine influence. In sediments, Eh values ranged from 134 to −222 mv, and pH between 4.2 and 10.9. The lower Eh values were found in Aveiro City canals and Murtosa channel. Some sites of Murtosa channel also have low pH values.

Sediment mean grain size (SMGS) varied between 19.7 and 3660.2 μm and fine fraction (fines; <63 μm) between 0 and 97.7%. Total organic matter (TOC) content in dry sediments ranged from 0.1 to 7.7% (**Figure 1**). Concentrations of potentially toxic elements (PTEs) varied for Zn 2-684 mg kg^{-1}; Pb 7-851 mg kg^{-1}; Cu 0.03-121 mg kg^{-1}; As 03-119 mg kg^{-1}, and Cr 78-0.03 mg kg^{-1}. The highest TOC and PTEs contents were found in protected areas.

2.2.2 Foraminiferal assemblages

Living specimens density (no per gram of sediment fraction 63–500 μm) were < 2300 n°/g. Higher densities were found in protected areas of channels with a good connection with marine waters. Ninety species of living foraminifera were found in the Aveiro Lagoon. Number of species per sample (SR) varied from 0 to 28 and Shannon index values (H) were < 2.8. The most frequent species in living foraminiferal assemblages of Aveiro Lagoon are *Ammonia tepida* (<40%) and *Haynesina germânica* (<40%), which were found in all of the sites. Other species also reach relatively high relative abundance, at least locally, such as *Elphidium margaritaceum* (<54%), *Lepidodeuterammina ochracea* (<52%), *Lobatula lobatula* (<45%), *Rotaliammina concava* (<32%), *Bolivina ordinaria* (<31%), *Cibicides ungerianus* (<19%), *Planorbulina mediterranensis* (<17%), *Cribroelphidium excavatum*, *Elphidium gerthi* (<14%), *Elphidium complanatum* (<14%), *Bolivina pseudoplicata* (<13%), *Remaneica helgolandica* (<13%), *Bulimina elongata/B. gibba* (<10%), *Elphidium williamsoni* (<6%), *Gavelinopsis praegeri* (<6%), *Trochammina inflata (<5%)*, *Elphidium crispum* (<6%), *Cribroelphidium excavatum* (<5%), *Quinqueloculina seminula* (<5%), and *Cribrostomoides jeffreysii* (<5%). Other species, such as *Buliminella elegantissima*, *Miliammina fusca*, *Haplophragmoides manilaensis*, *Entzia macrescens*, *Tiphotrocha comprimata*, *Ammoscalaria pseudospiralis*, *Arenoparrella mexicana*, *Siphotrochammina lobata*, *Ammobaculites balkwilli*, and *Eggerelloides scaber*, occur in general with percentages less than 5%.

2.3 Discussion of the results obtained in Aveiro Lagoon

The higher values of SMGS are common in samples collected along the inlet and S. Jacinto channels where the tidal currents are stronger and reach frequently velocities >2 m s^{-1} [29], and in stations of other channels due to stronger currents activity in interaction with local topographic effects. Tidal currents affect not only the sediments' texture but also their chemical composition in Aveiro Lagoon [37, 38]. The sediments are coarse-grained and have low organic matter content where the currents are strong. Under low currents activity, fine-grained sediments enriched in organic matter are accumulated. The heterotrophic activity in Aveiro Lagoon is intense [39], resulting in negative sedimentary redox potential values in many areas mostly where fine sediments and high organic matter contents are accumulated. As the region surrounding the Aveiro Lagoon is densely populated, in the most confined areas located near cities and villages or close to the rivers' mouths, higher available concentrations of PTE (such as Cr, Cu, Ni, Pb, and Zn) can be found. Highest PTE values were found, for instance, in Aveiro city and Murtosa Channel and the lowest values in the lagoon entrance.

In Aveiro Lagoon, in addition to the salinity and organic matter contents, the hydrodynamical conditions have an important influence in the pattern of distribution of benthic foraminifera assemblages. Living foraminifera density tends to increase in fine-grained sediments enriched in organic matter.

Most of the living species found in the Ria de Aveiro are typical of European estuarine environments [40, 41], of worldwide transitional environments [42], and some are present in the nearby continental shelf [43, 44]. Species such as *H. germanica*, *A. tepida*, *C. excavatum*, and *T. inflata* are typical of coastal and transitional environments [45, 46] and are quite common in Ria de Aveiro.

In Aveiro Lagoon, the agglutinated species that are known to be well adapted to a wide range of salinities [47] predominate in different ecological niches, all of them characterized by high environmental stress. *Lepidodeuterammina ochracea* and *Rotaliammina concava* dominate in very strong hydrodynamical conditions at the lagoon entrance. Instead *Miliammina fusca*, *Haplophragmoides manilaensis*, *Entzia macrescens*, *Tiphotrocha comprimata*, *Ammoscalaria pseudospiralis*, *Arenoparrella mexicana*, *Siphotrochammina lobata*, and *Ammobaculites balkwilli* reach the highest relative abundance but have low densities in low salinity waters near the rivers' mouth and in sediments with relatively low Eh and pH values, where the abundance of calcareous species decline [23, 37]. According to Fatela et al. [21], the low pH values in sediment pore water limit the episodic presence of calcareous foraminifera. Low pH levels coupled with the reactivity of biogenic carbonates may promote dissolution and destruction of calcareous tests [48].

The diversity and species richness tend to increase in the deeper areas under greater oceanic influence where there is also an increase of, for instance, *E. margaritaceum*, *L. ochracea*, *L. lobatula*, *R. concava*, *B. ordinaria*, *C. ungerianus*, *P. mediterranensis*, *E. gerthi*, *E. complanatum*, *B. pseudoplicata*, *B. elongata*/*B. gibba*, *G. praegeri*, *E. crispum*, and *C. jeffreysii*. These species seem to prefer more saline and oxygenated waters and less impacted environments and thus are named as "marine species" [43, 44].

Excess of organic matter linked with fine-grained sediments can lead to depressed levels of oxygen in the sediment pore waters, which may cause stress to benthic foraminifera [49]. However, *H. germanica, A. tepida, Bolivina ordinaria, Bolivina pseudoplicata*, *T. inflata*, and *C. excavatum,* for instance, can occur in such conditions, which means that they tolerate better the negative effects of eutrophication than, for example, *L. ochracea*, *L. lobatula*, *R. concava*, *C. ungerianus*, *P. mediterranensis*, and *G. praegeri*. However, it is known that benthic foraminifera are very tolerant to oxygen depletion, and some species appear to be resistant to hypoxic and periodic anoxic conditions [50].

According to Armynot du Châtelet et al. [51], the relative abundance of *A. tepida* is typically favored by an increase of total organic matter, meaning food resources. *Ammonia tepida* has been invariably reported as a potential bioindicator of pollution at the majority of the coastal polluted sites [1]. In general, the sites polluted with sewage rich in toxic metals had low foraminiferal abundance, high percentages of *A. tepida*, low percentage of epiphytic species, and more deformed fauna [46]. In this work, *A. tepida* is present in the most polluted sediments of the Aveiro Lagoon, but it seems to be not firstly related to the PTE enrichment. According to Armynot du Châtelet et al. [51], the relative abundance of *A. tepida* is typically favored by an increase of total organic matter, meaning food resources. A few workers, however, suggested that the preference of *A. tepida* for fine-grained organic carbon-rich sediments may be the reason for its dominance in polluted regions [1].

In Aveiro Lagoon, *H. germanica* is mostly associated with confined lagoonal sites with high content in organic matter under low currents activity and waters with relatively high salinity. This species probably displays an opportunist behavior benefiting of the organic matter supply (food) and tolerating low levels of oxygen. *Haynesina germanica* is a mid-latitudinal, temperate, and euryhaline species that populates shallow water muddy and phytal environments of salt marshes, intertidal habitats with salinities that generally range between 1 and 30, and optimal temperatures between 12 and 22°C [52]. Armynot du Châtelet et al. [10] have also shown that *H. germanica* is a successful pioneer species in polluted estuarine environments and in rich organic matter sediments and is tolerant to heavy metals. This species seems to be quite tolerant to higher concentrations of metals, namely Zn, Pb, and Cu, in Aveiro Lagoon.

The results obtained in Aveiro Lagoon also indicate that most of the species that live in this lagoon, mainly those that drive to the most internal and confined areas of the lagoon, should tolerate the stress caused by eutrophication and relatively high concentrations of PTE, namely *H. germanica*.

In general, the density and diversity of foraminifera are low in the lagoon. However, in the most impacted zones the density and diversity of foraminifera become even smaller. In fact, the increase in pollutants has been reported in general, as being marked by a decrease in species diversity with increased abundance of stress-tolerant species and high percentage of abnormalities [45, 46].

It is known that the distribution of the living assemblages is strongly affected by the estuarine dynamic, since foraminifera react within less than 1 month to changes of environmental conditions [17]. The distribution of the living foraminifera species results in several blooms throughout the year, for this reason the abundance and diversity of foraminifera is naturally temporally variable [17]. Living assemblages of foraminifera can be quite variable over time depending on the variability of the physicochemical parameters (according to weather changes). Therefore, monitoring studies may provide data not only on the response of species to the variation of environmental parameters but also on the gradients of natural and/or anthropogenic environmental impact.

3. Case study: São Sebastião Channel, SE coast of Brazil

Deciphering the impacts of domestic and industrial pollutants is difficult because they often occur together in sheltered coastal environments (bays or estuaries). When they occur separately, it is often in environments with different natural conditions, which makes comparison problematic. São Sebastião Channel (SSC) is an open area where industrial and domestic effluents are separately disposed, but under similar natural conditions, offering the opportunity to compare their impact on benthic biota [53].

SSC, located between the latitudes 23°40′S and 23°53.5′S and the longitudes 45°19′ and 45°30′W, is a 25 km stretch, which separates the continent from São Sebastião island (**Figure 2**). SSC width ranges from 2 km in its central portion and 7 km in its southern and northern ends. Its axis, where the largest depths (30–50 m) are found, is located closer to the island, due to the erosion and/or structural conditioning of the bottom. The smaller depths (6 m) occur on the continental side of the channel. The southern and northern ends have depths of 25 and 20 m, respectively (**Figure 2**).

The water circulation in the channel is characterized by alternate northerly and southerly movements, with a periodicity of days that is not directly influenced by tidal currents [54]. Geometry and topography of the channel bottom produce more intense longitudinal currents on the insular side, with speeds of up to 1.0 m s^{-1} toward the north and 0.7 m s^{-1} toward the south.

In SSC, there are some areas where the anthropic influence is quite intensified. Among these, the central region of the channel has the largest petroliferous terminals of South America, “Dutos e Terminais Centro Sul” (DTCS) of PETROBRAS. According to Duleba et al. [53], the DTCS generates two types of

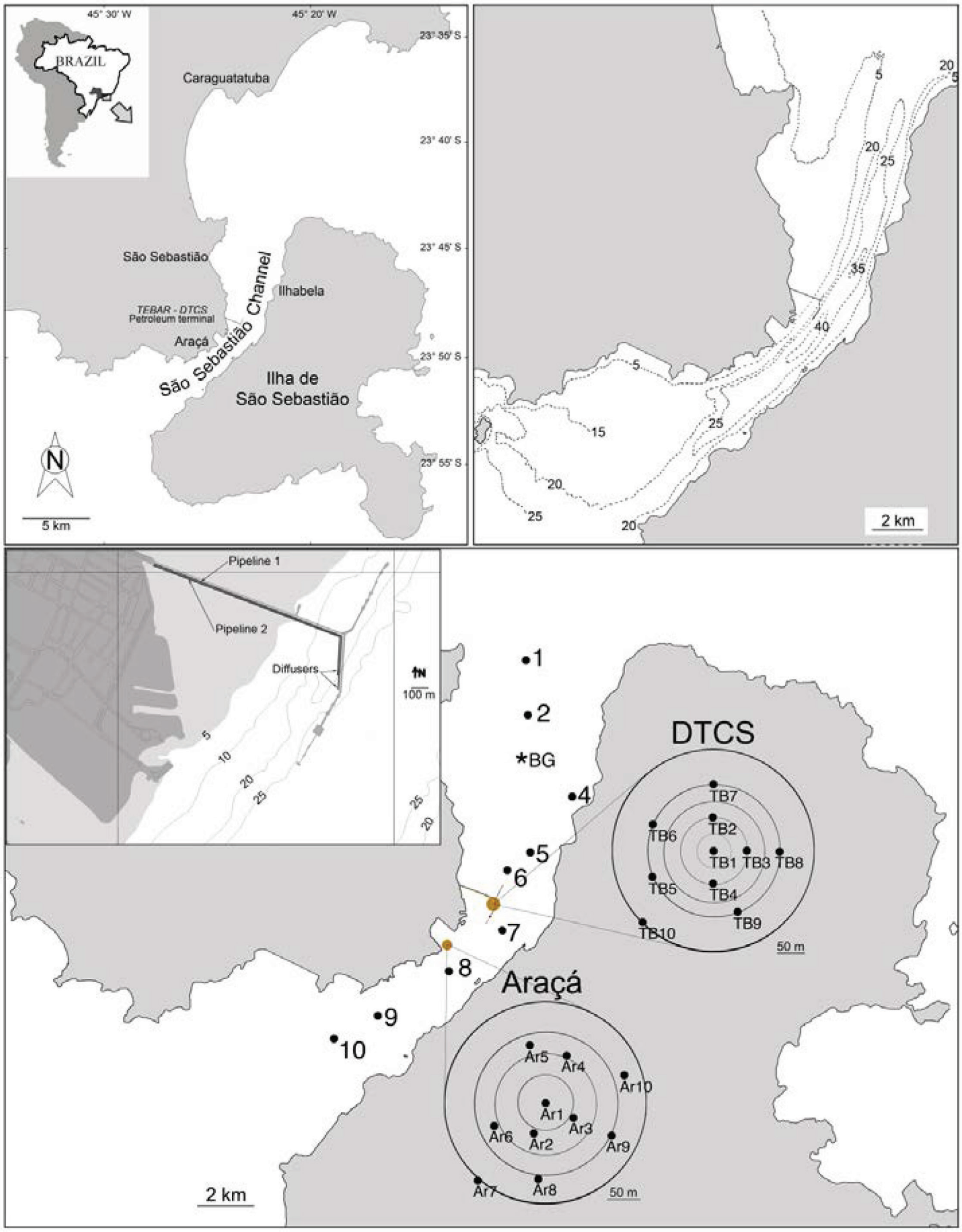

Figure 2.
Study areas and sampling grid in São Sebastião Channel (SE coast of Brazil).

liquid effluents. The first type consists mostly of water, separated from oil by density during transportation from drilling platforms in the oil tankers; the second type consists of rainwater and industrial water from the DTCS, contaminated by oil. These waters are treated in the wastewater treatment plants (ETE—Estação de Tratamento de Efluentes), where they are first separated from residual oil by adding a solution of polyelectrolyte [55]. Then, a series of treatments, using hydrogen peroxide at several pH levels, allows the oxidization of sulfides and phenol before an ultimate neutralization of the effluent. Approximately 15,000 m^3 of produced waters are treated every month. According to Fortis et al. [55], contaminated rainwater and industrial water are treated in systems that use mostly decantation to separate oil from water (SAO = Separação de Águas Oleosas). In addition, if the quality of the outflowing water does not satisfy the legal regulations, it is sent for treatment in the ETE. After treatment, effluents from both the ETE and SAO are mixed before being discharged through two submarine pipelines (with 1600 and 1400 m long), which end in diffusers, located at a depth ranging from 20 to 25 m [55].

The oil separation techniques, used in the wastewater treatment plants, primarily remove particulate matter and dispersed oil, while dissolved hydrocarbons remain in the discharged water [55]. The treated water is generally enriched with ammonia [55] and dissolved ions of sodium, potassium, magnesium, chloride, and sulfate, leading to salinity levels of up to 52.8 [55]. The treated water also has elevated levels of some heavy metals, as well as corrosion and scale inhibitors, biocides, dispersants, emulsion breakers, and other chemicals [56].

Close to DTCS, there is the submarine outfall of Araçá, which transports almost all the domestic effluents from the São Sebastião city. That emissary has a total length of 1061 m and a diameter of 400 mm. Its diffuser has a length 10.1 m and is located at Araçá point, at a depth of 8 m. The discharge speed per final discharge is 91.46 $l\ s^{-1}$ [46].

3.1 Methods applied in São Sebastião Channel

Sediment samples from DTCS (named TB) were collected near the outfall diffusers in September 2005, by the Environmental Agency of the São Paulo State, Brazil (CETESB—"Companhia Ambiental do Estado de São Paulo"). This authority evaluates the adequacy of the wastewater plants projects, the definition of environmental monitoring programs, and the regulation and enforcement of the water quality compliance. The sampling grid, consisting of 10 sampling points, was located in an area of 125,000 m^2 surrounding the diffusers. In addition, samples from 10 stations along the São Sebastião Channel (SSC) and 10 stations around the Araçá (AR) domestic sewage outfall were used for comparison. The geographical positions of the sampling stations were determined using the global positioning system (GPS), with the UTM datum SAT 69.

Surface sediment samples were collected for the following analyses: (i) grain size analyses; (ii) geochemical analyses; and (iii) determination of living benthic foraminifera. Textural, trace elements, and foraminifera data from TB, AR, and SSC areas were previously studied by Teodoro et al. [46, 53], which described in detail the methods of sedimentological and geochemical analysis.

Concerning the foraminiferal study, immediately after sampling, the samples were fixed with 70% alcohol stained with 1 g Rose Bengal, to distinguish stained (living) from unstained (dead) benthic foraminifera [57]. Aliquots of 10 cm^3 of sediment were washed through two sieves: 0.5 and 0.063 mm. The obtained fractions were dried, and the foraminifera were separated from the sediment by flotation using trichloroethylene. In samples with a low number of foraminifera, aliquots of 10 cm^3 were successively analyzed for to count of at least 100 stained individuals [33, 58]. Therefore, about 100 or more stained foraminifera were handpicked for identification and counting at each station. Foraminiferal density 1 (density 1) is expressed as the number of foraminifera per volume of sediment and density 2 is number of foraminifera per 10 cm^3 [59]. Foraminiferal assemblages structure was analyzed by using the Shannon index (H′) [34], the equitability (J′), calculated according to the Pielou index [35], and the species richness, calculated from density 1.

Canonical correspondence analysis (CCA) was used to investigate the relationship between foraminifera and sedimentological variables of the three areas: TEBAR, Araçá, and São Sebastião Channel (see details in [53]). The Monte Carlo permutation test (999 permutations) was used to assess the statistical significance of the correlations (at $p < 0.05$ and $p < 0.01$).

3.2 Results obtained in São Sebastião Channel

3.2.1 São Sebastião Channel

A total of 88 living species were identified in the São Sebastião Channel [53] belonging to the suborders of Rotaliina (63 species), Textulariina (20 species), and Miliolina (only one species). The volume of analyzed sediment needed to obtain at least 95 stained foraminifers ranged from 10 to 60 cm^3. Density 1 values ranged from 95 specimens per 60 cm^3 of sediment to 296 specimens per 10 cm^3 of sediment. Density 2 values ranged from 16 to 296 specimens per 10 cm^3 of sediment. The highest densities were identified at stations SSC3 (266 specimens) and SSC6 (296 specimens). Richness values varied from 12 to 33 species. The H′ and J′ values varied between 1.59 and 3.25 and between 0.64 and 0.93, respectively.

Ammonia tepida was the most abundant species in almost all samples (5–56.1%). The following species presented significant relative abundance: *Ammonia parkinsoniana* (2–19.6%), *Bolivina striatula* (0.8–11.6%), *Globocassidulina crassa* (<18.8%), *Globocassidulina subglobosa* (<10.2%), *Nonionella opima* (<9.1%), *Buliminella elegantissima* (2–8.5%), *Bolivina fragilis* (<8.5%), *Bulimina marginata* (<5.9%), *Pseudononion japonicum* (<5.8%), *Hopkinsina pacifica* (<5.3%), *Rosalina floridensis* (<6.1%), *Gavelinolepsis praegeri* (<5.5%), and *Hanzawaia boueana* (<5.1%). At the reference station (SSC3), *A. tepida* had the highest relative abundance (42.5%), followed by *A. parkinsoniana* (16.2%), *B. striatula* (7.9%), and *B. elegantissima* (7.5%). At this station, 32 species were recognized.

3.2.2 Dutos e Terminais Centro Sul (DTCS)

Throughout this area, 45 species were identified as belonging to Rotaliina (37 species), Textulariina (6 species), and Miliolina (2 species). Foraminiferal densities ranged from 0.5 (TB9) to 25 (TB7) specimens per 10 cm^3 of sediment. Owing to this low density, the volume of analyzed sediment needed to obtain 95 stained individuals varied from 40 to 190 cm^3. Species richness varied from 12 to 23 per 95 foraminifera. The H′ and J’ values ranged from 1.5 to 2.4 and from 0.56 to 0.71, respectively. Both indices presented low values, indicating low species diversity, due to the dominance of few species.

Ammonia tepida was the most abundant species in all the samples (38.5–66%). The following species also had significant relative abundance: *Pararotalia cananeiaensis* (<20%), *B. elegantissima* (0.9–11.8%), *A. parkinsoniana* (<7.3%), *C. lobatulus* (<6.6%), *B. striatula* (<6.4%), *B. marginata* (1–6.3%), *B. ordinaria* (0.9–6%), *Bolivina compacta* (<5.5%), and *Rosalina floridensis* (<5%).

3.2.3 Araçá Outfall

In this area, 51 species [53] were identified as belonging to the suborders Rotaliina (33 species), Textulariina (11 species), and Miliolina (7 species). Foraminiferal densities ranged from 28 to 98 specimens per 10 cm^3 of sediment. A volume ranging from 10 to 40 cm^3 was analyzed to obtain 95 stained individuals. Species richness values varied from 13 to 28 species. The H′ and J’ values varied from 0.70 to 2.64 and from 0.69 to 0.85, respectively.

Ammonia tepida was the most abundant species in all samples of the Araçá region, with a relative abundance ranging between 24.7 and 47.3%. The following species also had significant relative abundance: *P. cananeiaensis* (1.8–17%), *C. lobatulus* (<11.9%), *B. ordinaria* (1.1–11.3%), *R. floridensis* (<8.6%), *B. elegantissima* (<7.8%), *B. striatula* (3.2–7.7%), *P. japonicum* (<6.1%), *G. crassa* (<6.1%), and *A. parkinsoniana* (<5.4%).

3.3 Discussion of the results obtained in São Sebastião Channel

Sediments in the DTCS area were silty with high concentrations of total organic carbon (1.7–2.4%), total nitrogen (0.2–0.3%), total sulfur (0.4–0.6%), and total phosphorous (0.12–0.18%) and inorganic phosphorous (0.07–0.11%). These values were higher than those in sediments collected in the SSC and Araçá regions. The sediments' concentrations of As, Cd, Cr, Cu, Hg, Ni, Pb, and Zn in the SSC and AR regions were lower than their corresponding probable effect levels (PELs) [53]. However, sediments near the DTCS were enriched with As, Cu, and Ni, whose concentrations exceeded their corresponding threshold effect levels (TELs).

Despite potentially considerable pollution sources, mostly around DTCS, the contamination of sediment, as measured through geochemical analyses, is moderate. This probably results from the dispersion of effluents by the currents that affect the São Sebastião Channel. However, even if they are dispersed and do not accumulate within the sediment, pollutant may affect the benthos since all habitats exposed to all types of contaminants experience decreased biodiversity [60]. Indeed, the low densities of foraminifera around the DTCS diffusers illustrate the impact of environmental stress on the benthos.

In the DTCS area, it was necessary to search 50–190 cm^3 of sediment to find 100 living specimens (an average of 9 ± 6 individuals per 10 cm^3 of sediment) [53]. In the SSC and Araçá areas, a maximum of 40 cm^3 of sediment was enough to locate 100 living specimens (an average density of 62 ± 22 foraminifera per 10 cm^3 of sediment).

Organic matter may favor microfauna [7], or it may be responsible for decreasing microfauna density and richness [10, 15]. The toxic threshold depends on the nature of the organic matter and its concentration in the sediments [10]. The degradation of organic matter requires large quantities of oxygen; thus, when the flux of organic matter exceeds the degradation rate, a benthic hypoxia or even anoxia can occur. In this sense, the microfauna is compelled to change: stenobiotics can disappear and an abundance of tolerant species may be observed [10, 15, 31].

Pararotalia cananeiaensis is an herbivorous, epifaunal species characteristic of a marine environment. It is abundant in dead assemblages all over the SSC. The positive correlation of *P. cananeiaensis*, together with *D. floridana*, with TP and Cd appears to be an indication that they are tolerant to these elements and to the associated contaminants. However, the ecological preference of *P. cananeiaensis* is not yet well known in Brazil (Debenay et al., 2001c)***. Near the Araçá domestic submarine outfall, Teodoro et al. [46] reported *P. cananeiaensis* living preferentially at stations with high sulfur content ($r = 0.86$; $p < 0.001$), particulate organic matter ($r = 0.62$; $p < 0.001$), and silt ($r = 0.75$; $p < 0.01$). In the present study, no relationship between abiotic parameters and relative abundance of *P. cananeiaensis* was recorded.

The species related to reducing muddy sediment, with a moderate concentration in Cr and a noticeable content of organic matter, were *B. marginata*, *B. elegantissima*, *B. compacta*, *A. tepida*, and *A. parkinsoniana*. Most of these species are recognized in the literature as tolerant to high organic matter flux and as able to survive in low oxic conditions. Such is the case of *B. marginata* [61] and *B. elegantissima* [14]. Bandy et al. [62] noted that *B. elegantissima* and *B. marginata* tend to be abundant in areas affected by pollutants. In this study, the highest abundance of bolivinids and buliminids was observed in stations positioned in the central part of the channel: stations SSC7, SSC8, and SSC9.

Ammonia tepida is a eurybiotic species characteristic of near-shore areas and paralic environments [63]. The tolerance of *A. tepida* to adverse conditions, including organic and chemical pollution, has long been reported in both field studies and culture studies [10]. Its potential application for pollution monitoring is well established. It reached the highest relative abundance in DTCS the polluted samples and, to a lesser extent, in Araçá. This higher proportion of *A. tepida* is due to a decline of stenobiotic species. The higher abundance of tolerant species in these areas indicates that the benthos is significantly impacted by both organic (Araçá) and chemical (DTCS) pollution and suffers with a greater impact of chemical pollution.

4. Case study: continental shelf of the Campos Basin (SE, Brazil)

The continental shelf of Campos Basin is located on the southeast coast of Brazil between latitude 21° and 23°S, the southwest margin of the Atlantic Ocean, which comprises part of the area occupied by the Marine Sedimentary Campos Basin between the 25 and 150 m isobaths (**Figure 3**). The sedimentary contribution to this region is restricted to the rivers Itapemirim, Paraíba do Sul, Macaé, and São João, and most of the material derived from rivers and coastal erosion seems to be retained in coastal waters; what exceeds in this region is readily carried by oceanic currents. Detrital materials derived from the river discharge of the Paraíba do Sul River are distributed throughout the inner shelf by developing small muddy zones and large mud accumulations in an area adjacent to Búzios and Cabo Frio, where the currents have energy close to zero, 150 km south to the mouth of the river [64].

The South Atlantic Central Waters (SACW), a colder, nutrient-rich water mass, enters the continental shelf increasing local primary production and associated secondary productivity. Many studies have been carried out to investigate aspects related to the coastal upwelling of Cabo Frio, south of the Campos Basin [65].

4.1 Methods applied in Continental Shelf of the Campos Basin

Sampling methods, preparation of samples, control of quality, and methods of data analysis are described in detail in [66]. Living benthic foraminifera of the Campos Basin continental shelf were studied in 239 samples collected in nine transects (A–I) perpendicular to the coast line. The sampling was performed during the dry season of 2008 and the rainy season of 2009. The dry season, in this region, occurs in winter and corresponds to lowest precipitation and less frequent upwelling events. By contrast, the rainy season takes place in summer and corresponds to the period with higher precipitation and with more frequent upwelling events.

At each transect, five isobaths (25, 50, 75, 100, and 150 m) were sampled with a very large and modified (with an upper opening) Van Veen, which functions as a box corer. In each isobath, three independent samples successfully taken with a 10 cm × 10 cm × 2 cm, a “quadrat” yields samples with 200 cm^3. A fixative solution (4% formalin buffered with sodium borate) with Rose Bengal stain was immediately added to fix and evidence the protoplasm of living foraminifera. In the laboratory, 20 cm^3 of sediment from each sample was separated for analysis of living foraminifera.

The foraminiferal samples were washed trough a 63 μm sieve, dried in an oven (<60°C), and then picked under a stereomicroscope. Density values are equivalent to the total number of living individuals in 20 cm^3 (volume) or 10 cm^2 (area). Although the 63 μm mesh size was used as the size limit in the washing and screening of samples, many individuals smaller than 63 μm were found alive adhered to the grains; they were removed and incorporated into the slides for study. Careful quality control ensured similar patterns of screening and identification of living foraminifera, making the differences between pickers minimized. The species identification was based on [67, 68], and other specific references, as well as by the analysis of museum collections. The biomass was calculated by the volumetric method [33, 69].

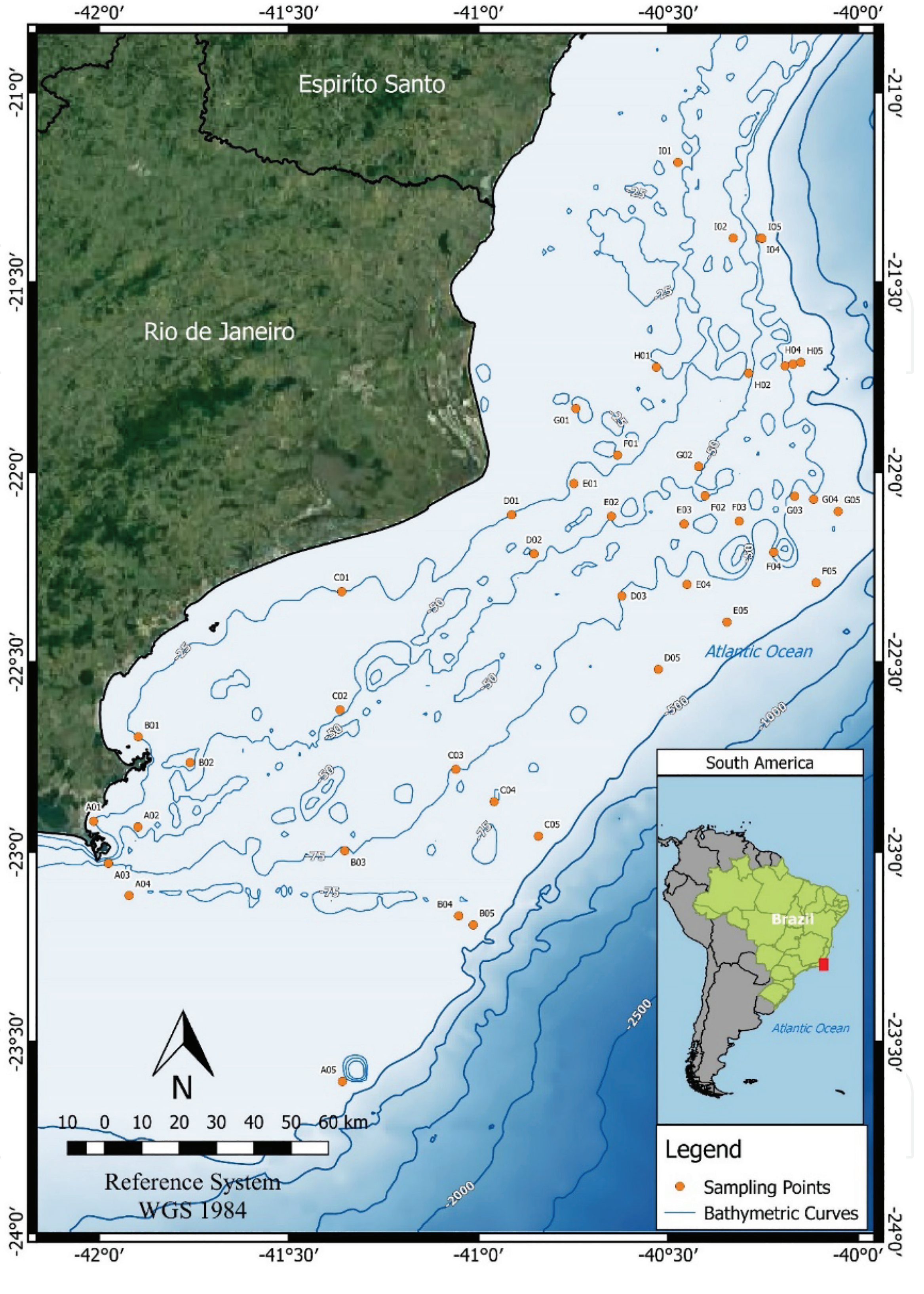

Figure 3.
Continental shelf of Campos Basin area. The transects A–I and their sampling points 1–5, according to the isobaths (25 m = 1; 50 m = 2; 75 m = 3; 100 m = 4; and 150 m = 5).

According to Pianka [70], k-strategist or conservative species have greater body size, longer life cycle, and population size largely constant in time, being close to the capacity of support of the environment; r-strategist species are known for their opportunistic behavior, small size, short life cycle, and very variable population size without adjustment balance in relation to available resources (mainly space and food). R-strategists can proliferate opportunistically and vary considerably its absolute abundance. According to these characteristics, Warwick [71] developed a method based on the abundance and biomass curves (ABC) for the detection of perturbation and to analyze the species response to environmental changes. The ABC curves are compared together with the W statistic calculation [71]. According to Magurran [36], in undisturbed environments, one or two species dominate the biomass and this has the effect of raising the biomass curve in relation to the abundance curve. On the contrary, in highly disturbed environments, a few species have very large number of individuals of small size, but since these species have small size, they do not dominate the biomass and so, the abundance curve is above the biomass curve. In intermediate conditions, the biomass and abundance curves cross each other several times. In this work, the ABC curves were generated for each group in both periods with all the species in which it was possible to calculate the biomass.

4.2 Discussion of results of continental shelf of the Campos Basin

The composition of the foraminifera assemblages allowed to detect areas under upwelling influence on the continental shelf. *Epistominella exigua*, *Adercotryma glomeratum*, *Bulimina marginata*, *Pappina compressa*, *Angulogerina angulosa* s.l., *Nonionella stella*, *Nonionella opima*, *Hopkinsina pacifica, Bolivina fragilis, Bolivinellina translucens, Fursenkoina pontoni*, and *Stainforthia complanata* can be considered as indicators of seasonality-enriched areas with phytodetrital material signaling the upwelling events in the study region of Campos Basin. The continental shelf presents many encrusting foraminifera, both large and small adults, as well as juveniles of larger species that are adhered to the sediment grains, indicating that it is a region with a predominance of high hydrodynamic energy near the bottom. Small individuals play an important role in the ecological characterization of oligotrophic areas where organic carbon is scarce and rapidly consumed, transported, or oxidized.

The increase of the continental discharge during rainy season brings more inorganic and organic nutrients to the coastal system, increasing the primary production. The high supply of continental nutrients in addition to the nutrients of the coastal upwelling of Cabo Frio and adjacent areas disturbs the natural equilibrium of the biological communities of this area.

The results of the ABC curves by isobaths evidence that in the 25 m isobath of the dry season of 2008, there was some disturbance ($W = 0.0332$), but in the rainy season of 2009, the disturbance was much high ($W = -0.0071$), so that the abundance curve overlaps with that of biomass (**Figure 4**). The ABC curves indicate the 25 m isobath as the most disturbed one within the continental shelf and the upwelling/organic enriched group as an area of moderated disturbance. During the rainy season, the 25 m isobath is disturbed by natural eutrophication phenomena and it may be significantly amplified by anthropogenic activities in the catchment area of Paraiba do Sul river and other anthropogenic disturbances, once this segment of the Brazilian coast is strongly influenced by agricultural, industrial, and urban activities as well as by fishery and many other coastal activities. In the southern region, the disturbed patterns seem to be strongly related to natural phenomena due to the presence of many species who indicate upwelling events, which are well known near the Cabo Frio area.

In his final remarks, Warwick [71] does not believe that this method can be applied to meiobenthic species, because according to him, there are no obvious size differences between k species and meiobenthic r-strategists. He mentioned Oncholaimidae nematodes and Tisbidae copepods as dominant species in polluted environments and reminds that they are often larger. However, among the foraminifers, there are species known as k-strategists that correspond to the characteristics mentioned by Pianka [70], and therefore, the application of the ABC curves to assess environmental disturbance could be successful. Although the foraminifera belong to both micro- and meiofauna, the results obtained with the ABC curves in

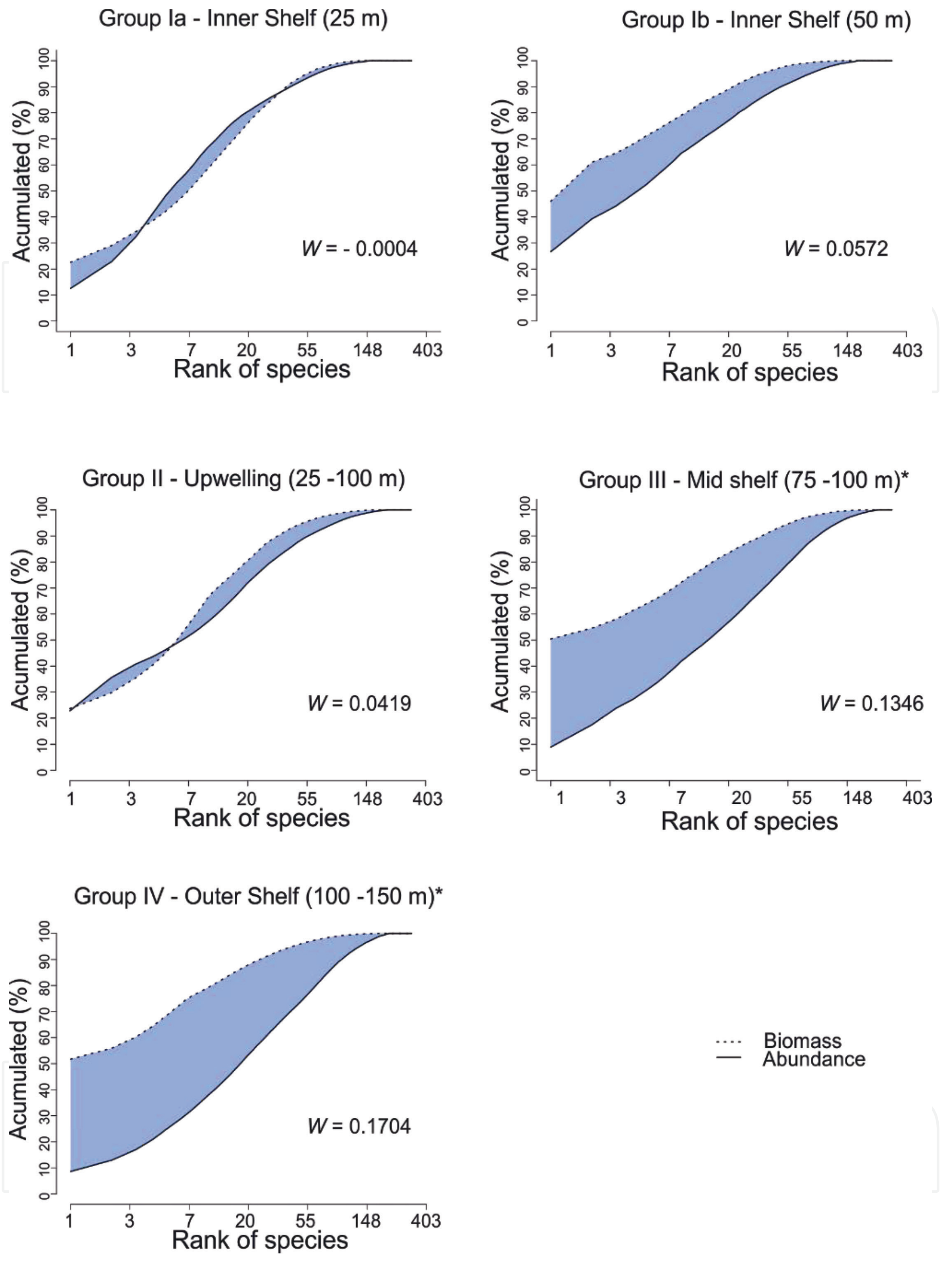

Figure 4.
ABC curves of the continental shelf assemblages of foraminifers. Groups 1–4 of the cluster analysis from rainy period (2009).

this work evidence they can be used to trace environmental disturbance. In fact, it is expected that the ABC curves can be used to identify both natural and anthropogenic stressors. However, without complementary studies that allow to qualify and quantify the disturbance origin by applying the ABC curves, we will have only the sign of the environmental disorder. On the other hand, we still have little information on the response of some species to natural and anthropogenic stressors, but this integrated study will allow the construction of a more effective monitoring plan, which can provide subsidies for preserving the integrity of the environment.

5. Case study: continental slope of the Campos Basin (SE, Brazil)

The Campos Basin is located between latitudes 21°S and 23°S in the southwestern margin of the South Atlantic Ocean (**Figure 5**). It ranges from the coastal plain to the São Paulo Plateau, the outer boundary of the salt tectonic coinciding with the boundary between continental and oceanic crusts [72]. The continental slope displays a convex profile to the north and a concave one to the south, and it is cut by several canyons, such as the São Tomé, Itapemirim, Grussaí, and Almirante Câmara [72]. The Campos Basin is considered a meso-oligotrophic system with oxygenated bottom water [73].

In Campos Basin, the oil exploration activities began in 1976, with the first maritime drilling occurring at 100 m of water depth. In 1999, exploration and production of hydrocarbons in deep waters (below 2000 m of depth) started in the Campos Basin. About 65% of hydrocarbon exploration and production activities are concentrated in marine areas deeper than 400 m [74].

The first initiative to evaluate the environmental conditions of the Campos Basin occurred in the end of 1980s, through a partnership between the Fundação de Estudos e Pesquisas Aquáticas (FUNDESPA), University of São Paulo, and Petróleo Brasileiro S/A (PETROBRAS). Regrettably, the data obtained were not widely disseminated through scientific publications [74].

From this on, other projects were executed in the Campos Basin, such as "Campos Basin Deep-sea Environmental Program" (Oceanprof) and

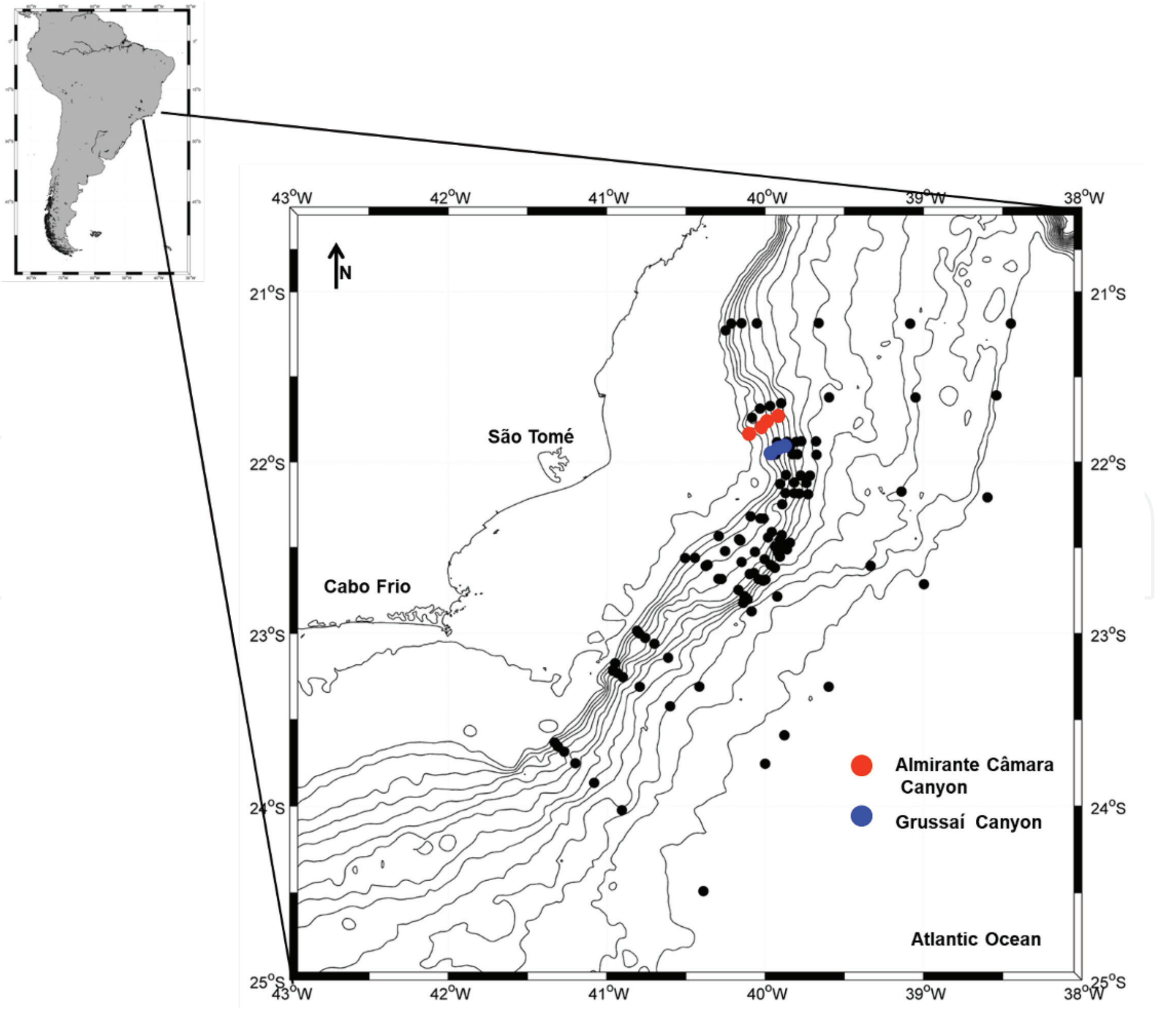

Figure 5.
Location of Campos Basin in the Southwest Atlantic Ocean and sampling sites of the study area (Oceanprof and Habitats project): red circle (station in the Almirante Câmara canyon); and blue circle (station in the Almirante Câmara canyon).

"Environmental Heterogeneity of the Campos Basin" (Habitats) projects. These projects had a multidisciplinary goal: geology and meteoceanography; organic and inorganic components of water and sediment; and the distribution and composition of biota [73, 74].

5.1 Methods applied in continental slope of the Campos Basin (SE, Brazil)

In the Oceanprof project, 41 surface sediment samples were collected by using a box corer, along transects ranging from 750 to 1950 m of water depth on the continental slope (**Figure 5**), in the austral winter of 2003. The core top (0–2 cm interval) sediment sampled in each site was used to understand the living and dead benthic foraminifera distribution patterns and ecological preferences. For this purpose, identification and quantitative foraminifera analysis were performed using the 63 μm size fraction. The variables considered for the statistical analysis included percentages of sand and mud, calcium carbonate and total organic carbon contents in the sediment, and total phosphate in the water (see details in [73]).

In the same project (Oceanprof), 20 surface sediment samples were chosen along 1050 and 1950 m depth (**Figure 5**). In this case, only living (rose Bengal stained) benthic foraminifera were analyzed in combined samples collected in four different slices of the cores (0–2, 2–5, 5–10, and 10–15 cm) to understand the distribution patterns and their ecology. In this study, the variables considered for statistical analysis included the sand and mud contents, particulate organic matter flux to the sea floor, bottom water dissolved oxygen concentrations, calcium carbonate, total organic carbon, total nitrogen, and total lipid contents in the sediment (see details in [75]).

In the Habitats project, an ecological study of living (rose Bengal stained) benthic foraminifera was performed in samples collected with a box corer, on the continental slope, Plateau of São Paulo, and canyons, during two campaigns (austral winter of 2008 and summer of 2009). The stations followed nine transects from 400 to 3000 m deep (**Figure 5**). In the Grussaí and Almirante Câmara canyons, the stations were located in four isobaths (400, 700, 1000, and 1300 m deep) and the obtained data were compared with adjacent transects on the open slope (**Figure 5**). Changes in the density, diversity, and composition of benthic foraminifera were analyzed in response to environmental factors (i.e., sand and mud contents, calcium carbonate, total organic carbon and chlorophyll-*a* concentrations, and phytopigment concentrations in the sediment) (see details in [76]).

5.2 Results obtained in continental slope of the Campos Basin and discussion

The middle slope is characterized by the dominance of different species of the genus *Bolivina, Cassidulina laevigata*, and *Globocassidulina subglobosa*. The occurrence of these species in association with *Cibicidoides kullenbergi, Epistominella exigua*, and *Uvigerina proboscidea* seems to be related to seasonal organic matter fluxes, relatively oxic bottom waters, strong bottom currents, and sandy sediments. The lower slope is inhabited by a microfauna with different characteristics, preferentially composed of epifaunal or shallow infaunal deposit feeding species (e.g., *Bolivina* spp., *Eponides weddellensis,* and *Lenticulina cultrata*) and suspension feeders that are adapted to oligotrophic conditions and high dissolved oxygen levels in the bottom waters, for example, *Rhabdammina* spp., *Rhizammina* sp. [73].

Yamashita et al. [75] concluded that besides the sediment grain size, the vertical flux of particulate organic matter seems to be the main factor controlling the spatial distribution of benthic foraminifera species in the slope of Campos Basin. The middle slope (1050 m of water depth) was characterized by relatively high foraminiferal density and a predominance of phytodetritus-feeding foraminifera such as *Epistominella exigua* and *Globocassidulina subglobosa*. The occurrence of these species seems to reflect the Brazil Current System (BCS). The abovementioned currents are associated with the relatively high vertical flux of particulate organic matter and the prevalence of sandy sediments, respectively. The lower slope (between 1350 and 1950 m of water depth) was marked by low foraminiferal density and assemblages composed of *Bolivina* spp. and *Brizalina* spp., with low particulate organic matter flux values, muddy sediments, and more refractory organic matter. The distribution of this group seems to be related to episodic fluxes of food particles to the seafloor, which are influenced by the BCS at the surface and are deposited under low deep current activity (Intermediate Western Boundary Current; [75]).

According to Sousa et al. [76], the availability and quality of the food, the energy state (stability) at the benthic/pelagic boundary, and the grain size of the substrate seem to be the most important environmental factors determining the distribution pattern of the benthic foraminiferal assemblages in the Campos Basin slope. The highest values of density, diversity, and richness, as well as the predominance of hyaline calcareous foraminifera and infaunal species, reflect a higher contribution of food received continuously in the shallower stations (400 m depth). At 700–1000 m of water depth, the density of foraminifera decreased and there was a larger presence of opportunistic species, possibly reflecting the pulse of phytodetritus. The considerable increase in the agglutinated foraminifera, the continuous decrease in the density of foraminifera, and also decrease in the values of the Benthic Foraminifera High Productivity Index (BFHP [61]) as depth increases indicate typical oligotrophic conditions in this sector of the Campos Basin (lower slope and São Paulo Plateau).

The comparison of density and species composition data in the austral winter of 2008 and austral summer of 2009 periods allows us to infer that during the winter, the food input was higher. The values of density and biomass of living benthic foraminifera allow us to suggest that the Almirante Câmara canyon is a greater entrapment site of organic matter between 400 and 1000 m isobaths in comparison to the open areas at the same isobaths [76].

Even though the stations analyzed were the same or spatially very close to each other, the response of benthic foraminifera of the Campos Basin seems to be slightly different due to the seasonality. Because of these dissimilar times, the local hydrodynamics can change and, consequently, variations can occur in the particulate organic matter flux, and quantity and quality of the organic matter on the sea floor [73, 75, 76]. The benthic foraminifera and the geochemical results of these projects showed that, despite the intense oil exploration and production activities in the Campos Basin by PETROBRAS, these specific study areas (**Figure 5**) did not suffer anthropic impact in terms of pollutants [77].

6. Conclusion

This work presents results of living foraminifera used to analyze these meiofaunal organism responses to different types of environmental disturbance in different transitional and marine settings: a semienclosed coastal lagoon (Aveiro Lagoon), an estuarine system (São Sebastião Channel), a continental shelf (Campos Basin), and continental slope environments (Campos Basin). Each area has different particularities, conditioning the type of living foraminifera associations that inhabit them, being the first two areas highly anthropized.

The dynamics of tidal currents, in interaction with the configuration of channels and local topography, generates different sedimentary facies in the Aveiro Lagoon, influencing the abundance of living benthic foraminifera. Salinity is a key factor for governing the structure (diversity) of foraminifera assemblages, as well as the concentrations of PTE, whereas the sensitive species ("marine species") avoid the inner lagoonal environments. Species such as *H. germanica*, *A. tepida*, *T. inflata*, and *C. excavatum* increase their frequencies in the most confined places and are impacted by high concentrations of PTE.

In São Sebastião Channel, the sediments near the "Dutos e Terminais Centro Sul" (DTCS) of PETROBRAS were enriched by As, Cu, and Ni, with concentrations exceeding TEL; these levels are associated with adverse biological effects. Comparatively, foraminiferal parameters (density and diversity) at the DTCS were lower than those observed in neighboring areas, even near the Araçá submarine outfall less than 3 km away. These findings lead us to conclude that wastewater treatment in DTCS is not effective in removing some chemical elements from petrochemical waste liquid. Moreover, it may negatively impact benthic fauna around the DTCS.

The distribution pattern of the living foraminiferal assemblages in the continental shelf of the Campos Basin changes depending on the bathymetry, sediment characteristics, and the supply of organic matter. The abundance of benthic foraminifera populations is strongly enlarged by the seasonal supply of phytobenthos and phytoplankton. The application of the ABC curves method in foraminiferal assemblages is a promising alternative to evaluate the environmental conditions and to access specific areas over time, and thus, they can be applied in environmental monitoring studies.

The benthic foraminifera species that occur in the deep marine system of the Southwestern Atlantic (continental slope of the Campos Basin) are mainly controlled by local hydrodynamics, which mainly controls changes in the particulate organic matter flux, quantity, and quality of the organic matter in the seafloor. It should be also considered that these parameters are affected by seasonality.

The data presented here show the importance of understanding the ecology of the benthic foraminifera species for environmental assessment of the ecosystems, and therefore for the establishment of biomonitoring procedures.

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Conflict of interest

There are no "conflicts of interest."

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