Abstract

The Ammonia-Elphidium Index (IAE) allows to assess the oxygenation levels of coastal regions. Both genera used in this index are resistant to oxygen reduction conditions. The genus Ammonia has a greater resistance than the genus Elphidium to low oxic conditions, and both are abundant in the coastal zones, which makes possible the use of this index to access the impact caused by organic matter pollution in these regions. Hence, this index has been used in the literature to study polluted regions by large contribution of organic carbon. The Petrobras Polo Atalaia Production complex is responsible for the outfall of treated petrochemical effluents, called PAP-1, which introduce pollutants in the coastal area of Sergipe State, northeast Brazil. In view of the environmental complexity of the region, the objectives of this work were: 1) to analyze the dimension and diversity of living foraminifera assemblages in surface sediment samples collected in February 2014 around the Sergipe-Mar Subsea Outfall of Active Production (SMP); and 2) to apply the Ammonia-Elphidium index in this region as a proxy for environmental impact. The results allowed to evaluate the impact caused by the contribution of the effluents released by the SMP outfall of the Petrobras Polo Atalaia Production complex and their influence on the density and diversity of living benthic foraminifera in the study area.

Keywords: Benthic foraminifera. Environmental monitoring. Ammonia/Elphidium index. Petroleum hydrocarbons. Pollution.

1. Introduction

The present work is based on a study of living foraminifera assemblages of an area influenced by the supply of treated liquid effluents, located off the active submarine outfall of the Petrobras Polo Atalaia Production complex, Sergipe State (NE, Brazil). It is part of a broader biomonitroing survey of the inner continental shelf of the Polo Atalaia Production complex, entitled “Projeto de Monitoramento Ambiental do Emissário Submarino do Arvo de Produção Sergipe-Mar” (Environmental Monitoring Project for the Sergipe-Mar Subsea Outfall of the Active Production (SMP); Thalassa Project / Petrobras). The main goals of this work are to: provide technical and scientific elements to improve the biomonitroing studies that use foraminifera as environmental quality bioindicators, particularly in marine regions that are being affected by petrochemical effluents in the Petrobras Polo Atalaia Production complex area, Sergipe and; to infer about the environmental impact assessment of these effluents in the area around the SMP, based on foraminiferal response to organic matter increase and the presence of petroleum products.

According to the Group of Experts on the Scientific Aspect of Marine Environmental Protection/United Nations (GESAMP/UN), marine pollution can be defined as the direct or indirect introduction of substances or energy into the marine and estuarine environment by man, which results in: (1) negative impacts on living resources; (2) risks to human health and hindrance to marine activities, including fishing; (3) impairment of seawater quality; (4) loss of aesthetic beauty and; (5) impacts on sensitive habitats (GESAMP, 2015).
The term environmental contamination, which is often used in the sense of environmental pollution, is related to the increase of normal concentrations of chemical elements or microorganisms (bacteria), in water or sediments, due to anthropic activities. These concentrations can be harmful to living beings (GESAMP, 2015).

Pollution and/or contamination related to coastal and marine waters is a complex environmental and political problem that can be the center of conflict between countries, states, social groups and companies. These disturbances occur because national and state boundaries are not natural barriers to the dispersion of pollutants through water bodies or through the atmosphere. For this reason, political and legislative mechanisms should be created and monitored by governmental agencies that regulate the quality of the environment. However, this is not an easy task to carry out, since certain decisions should be based on studies that require a deep understanding of the interaction between pollutants and the environmental compartments they affect (Shortle and Horan, 2013).

Oil and gas industries and terminals generate large volumes of effluent and solid waste disposal, encompassing large amounts of aromatic and polycyclic hydrocarbons, phenols, metal and metalloid elements, sulfides, naphthalene acids and several other types of substances (Israel et al., 2008). Particularly, the metallic elements (Pb, Cd, Cu, Hg, Zn, Cr and Ni) are industrial contaminants that require greater environmental control because they are toxic and, of some of them, have bioaccumulative properties (Lacerda and Marins, 2006). In aquatic environments, factors such as pH and oxidation reduction potential can make bioavailable such elements which should be integrated into the trophic chain, reaching different communities, including man (Martins et al., 2015 a).

In Brazil, before launching into oceanic waters, the effluents of petrochemical terminals must be treated in Sewage Treatment Stations (STS), which requires extensive inspection by the Brazilian government. The environmental control is usually reduced to the analysis of waters and effluents, allowing the description of the impact degree only for the time of data collection (Rodrigues Filho et al., 2002). Thus, other sources of diffuse pollution are not detected and it is not possible to evaluate the impact of emissions with great security (Rodrigues Filho et al., 2002).

For an assessment of the aquatic environmental quality as a whole, it is also necessary to continuously assess the sediment quality and the amount of suspended particulate matter. Several factors may influence the distribution of metals, such as sediment texture, grain size, organic matter concentration, oxides, iron and manganese oxides and hydroxides and calcium carbonate (Vilela et al., 2004; Martins et al., 2015b, 2016a). The study of organic matter quality, and metals concentrations and their relation with benthic fauna are of fundamental importance to evaluate the environmental health of coastal ecosystems (Martins et al., 2016 b, c; Delavy et al., 2016; Raposo et al., 2018).

Foraminifera are free-living protozoa, protected by carbonated and/or secret-agglutinated tests, consisting of sedimentary particles from the environment (Loeblich and Tappan, 1964). Recently, species lacking shells have been discovered (Pawlowski et al., 2003; Lejzerowicz et al., 2010). They have been widely used in paleoceanographic studies as proxies for paleotemperature and/or paleoproductivity, because their tests are commonly well preserved in the sediments and retain many of their chemical and isotopic properties (Lisiecki and Raymo, 2005). It is stated that a proxy can be understood as an instrument to measure a variable no longer present in the environment (in paleoenvironmental studies) or not directly accessible (as in environmental studies).

Foraminifera have been used in environmental studies mainly since the 1980s (Scott et al., 2001), as they are able to metabolize chemical compounds and organic matter from the environment. These processes are later reflected in their morphology and assemblages size and composition (Ernst et al., 2006). Therefore, these organisms can help to detect what is happening in the environment (Lançone et al., 2005).

The analysis of living foraminifera assemblages is quite useful in environmental control studies since: the laboratory treatment of the samples is easy; a small sample volume provides, in general, a suitable number of individuals for several statistical analyses; foraminiferal tests are easily preserved; these organisms are sensitive to minimal environmental changes and have short life cycles tracing environmental changes on short time scales (Leorri et al., 2008).

In areas surrounding domestic sewage outfalls or petrochemical effluents we can find two distribution zones of benthic fauna well characterized by the influence of effluents. First, there is an "abiotic zone" or "dead zone" with no living foraminifera, and surrounding of the first areas, a "hypertrophic zone", where a significant increase in the density of the specimens is observed in general (Alve, 1995). After these zones, typical assemblages of the non-impacted environment can be found (Alve, 1995). This behavior was also described for polychaetas and other benthic invertebrates (Ellison et al., 1986).

In impacted environments foraminifera assemblages are in general dominated by species that are more resistant to pollutants (Yanko et al., 1999; Cearreta et al., 2002; Burone et al., 2006a; Le Cadre and Debenay, 2006; Frontalini and Coccioni, 2008; Eichler et al., 2012). Among them Ammonia tepida is considered a bioindicator of environmental stress (Bradshaw, 1961).

It is known that sediments are the final destination of most marine pollutants, but these can be captured and transformed by benthic fauna or other chemical processes...
managed, its waters cannot be used for industrial, human and irrigation purposes, because they are salted (Subsecretaria de Estado dos Recursos Energéticos e Sustentáveis, 2014).

Regarding the oil industry, between 2009 and 2012 there was an increase of 25.97% in the number of wells drilled in Sergipe. In 2012, 1882 wells were drilled. According to the National Petroleum Agency (ANP), Sergipe Basin has a reserve of 433 million barrels of oil and approximately 7 billion cubic meters of gas (Subsecretaria de Estado dos Recursos Energéticos e Sustentáveis, 2014).

Petrobras Polo Atalaia complex answer for the called PAP-1 outfall and also houses the Nitrogen Fertilizer Factory emissary (FaFen-SE), whose riser is located southwest of the studied stations. The water terminal of Aracaju, which is also part of the Polo Atalaia complex, is being operated by the subsidiary company Transpetro. It stores the oil extracted from Ceará, Espírito Santo, Alagoas, Rio Grande do Norte and Amazonas and is also used to store and ship all oil that is produced on the continental shelf and in the land fields of Siriri, Riachuelo and Carmópolis (Petrobras, 2017). To northeast there is another Petrobras facility, the Inácio Barbosa Maritime Terminal (TMIB), and the effluent outfall operated by the Companhia Vale. The emissary PAP-1, with 2.7 km in length, has no risers. The effluents resulting from the produced water at the Pole Atalaia complex are treated and thrown into the sea, with a flow of 70 m³/h (Petrobras, 2017).

2. Study Area

The study area is located in the northern region of the inner continental shelf (between 11°00’13.9480”S and 11°01’9.7781”S longitude and 37°01’10.2614”W and 37°02’43.4078”W latitude), between Sergipe and Vaza-Barris River mouths (Fig. 1).

The coast of Sergipe has an extension of 150 km (Bittencourt et al., 2006) and is delimited by the São Francisco River mouth at north and by the Real River mouth at South. Due to the São Francisco River delta, the northern part of Sergipe continental shelf is sandiest (Subsecretaria de Estado dos Recursos Energéticos e Sustentáveis, 2014). This region also presents the following sedimentary domains (Nascimento, 2011):

- Siliciclastic, consisting of muddy and sandy sediments, which are restricted to the inner continental shelf, beginning of the outer continental shelf and head of the São Francisco and Japaratuba canyons;
- Mixed, formed by bio-siliciclastic sediments, that can vary from granule to sandy mud, and can be found in the outer continental shelf;
- Bioelastic, formed by carbonated sand and mud, concentrated in the peripheral and central portions of the outer continental shelf.

With semidiurnal tides and two peaks of high and low tides in 24 hours and 50 minutes, the Sergipe coast is characterized by a meso tidal regime (amplitude between 2 and 4m) with the maximum amplitudes occurring in the equinoxes of March and September (Carvalho and Fontes, 2006).

The Sergipe River basin houses several economic activities. The main ones, by descending order of the occupied area, are ceramics, dairy, cleaning and confectionery industries, coffee production, tanning, spinning, weaving and chemical products, food, cement, textile processing, grinding, metal production, plastics, pulp production and animals slaughtering (Rocha, 2004).

In the Vaza-Barris river basin there are leisure, touristic and nautical activities. Near this river mouth of, there are large areas of limestone and sand extraction. This river crosses a land suitable for salinization. When it is poorly

3. Materials and methods

3.1 Sampling

The samples used in the present study were provided by Petrobras and are part of the biomonitoring program entitled Environmental Monitoring Project for the Sergipe-Mar Subsea Outfall of the Active Production (“Projeto de Monitoramento Ambiental do Emissário Submarino do Ativo de Produção Sergipe-Mar”, SMP). This biomonitoring program is taking place since 2013 following Brazilian environmental protection laws and is carried out twice a year during the dry and rainy seasons around the submarine emissary.

The sampling network was conceived by Petrobras and approved by the Brazilian Institute for the Environment and Renewable Natural Resources (“Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis”; IBAMA). The sampling network design was performed considering the position of the outfall, the bathymetry, the hydrodynamic conditions of the region and the displacement of the outflow resulting from the dispersion modeling of the emissary effluent. Nineteen stations were sampled: 13 were positioned in a gradient of concentric circles, with the outlet of the emissary as a starting point and 6 stations were located outside the area of the effluent dispersion plume influence (Fig. 1). The stations location is presented in Table 1.
Fig. 1. Map of the study area, with indication of the outfall, the monitoring stations and the control area and the modeling of the effluent plume.

The stations are located at a distance from the emissary of: A (50 m), B (150 m), C (250 m), D (400 m) and CO (control - 5,000 m). The bathymetry of the sampling network varies from 5m to 10m. In the present work, the stations A, B, C and D make part of the experimental group and the stations CO are the control group. Following the IBAMA specifications, at each station, three replicates were collected, totaling 57 samples destined to the study of living benthic foraminifera.

The samples were collected on February 10 and 11, 2014 (dry season), on board the MARIMAR XII vessel. Sampling was carried out by Gardline Marine Sciences of Brazil company and followed the IBAMA sampling protocols. On board, the samples for foraminiferal analysis were preserved with alcohol with Bengal Rose (Walton, 1952).

3.2 Treatment of samples for foraminiferal analyses

Shortly after the sampling process, the samples were processed in the laboratory. From each sample of the stained sediment, 20 cm$^3$ of sediment was removed and washed in sieves with 0.500- and 0.063-mm mesh apertures (Schröder et al., 1987). The fractions retained in the sieves were oven dried at 40 ℃ and subsequently subjected to densiometric analysis by flotation-sinking in trichlorethylene (Scott et al., 2001). This procedure aimed to separate the sediment (quartz grains and other minerals with density greater than 1.46 g cm$^{-3}$) from the biological material (diatoms, thecamoebians, foraminifera, tintinnids, sponge spicules, mollusks). The stained with Rose Bengal foraminifera were considered living at the sampling time and the uncolored tests (empty tests), the dead organisms. Only living organisms were analyzed in this work.

Once separated from the sediment, the floated material was analyzed under stereomicroscopic Zeiss Stemi SV6 microscope. For the biocenoses characterization about 300 living foraminifera specimens were picked (Fatela and Taborda, 2002). Samples with low abundance of foraminifera were fully analyzed and those with more than 300 specimens were sub-sampled with a Green Geological microsplitter.
Tab. 1. Geographic coordinates of the studied stations.

<table>
<thead>
<tr>
<th>Stations located around the outfall</th>
<th>Geographical Coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>11°01′11.7493″S 37°02′27.085″W</td>
</tr>
<tr>
<td>A2</td>
<td>11°01′9.7781″S 37°02′25.1807″W</td>
</tr>
<tr>
<td>B1</td>
<td>11°01′14.3683″S 37°02′29.566″W</td>
</tr>
<tr>
<td>B2</td>
<td>11°01′6.1735″S 37°02′22.3957″W</td>
</tr>
<tr>
<td>B3</td>
<td>11°01′7.3379″S 37°02′29.3191″W</td>
</tr>
<tr>
<td>B4</td>
<td>11°01′13.6563″S 37°02′22.5181″W</td>
</tr>
<tr>
<td>C1</td>
<td>11°01′4.3425″S 37°02′20.5514″W</td>
</tr>
<tr>
<td>C2</td>
<td>11°01′8.575″S 37°02′26.0774″W</td>
</tr>
<tr>
<td>C3</td>
<td>11°01′8.2944″S 37°02′36.0875″W</td>
</tr>
<tr>
<td>C4</td>
<td>11°01′9.6856″S 37°02′33.4929″W</td>
</tr>
<tr>
<td>D1</td>
<td>11°01′26.4587″S 37°02′30.6689″W</td>
</tr>
<tr>
<td>D2</td>
<td>11°01′22.6081″S 37°02′37.7089″W</td>
</tr>
<tr>
<td>D3</td>
<td>11°01′8.7377″S 37°02′43.4078″W</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Stations located at control area</th>
<th>Geographical Coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td>CO1</td>
<td>11°00′24.6364″S 37°01′17.5445″W</td>
</tr>
<tr>
<td>CO2</td>
<td>11°00′15.9605″S 37°01′15.1476″W</td>
</tr>
<tr>
<td>CO3</td>
<td>11°00′13.9480″S 37°01′16.1297″W</td>
</tr>
<tr>
<td>CO4</td>
<td>11°00′14.4860″S 37°01′10.2853″W</td>
</tr>
<tr>
<td>CO5</td>
<td>11°00′30.4797″S 37°01′10.2614″W</td>
</tr>
<tr>
<td>CO6</td>
<td>11°00′21.6361″S 37°01′19.2935″W</td>
</tr>
</tbody>
</table>

Particularly in the samples with more than 300 specimens, the floated and remaining material was reexamined after picking, in order to remove any colored stained specimens that were not sorted. For the analysis of foraminiferal biocenoses (living foraminifera), several aliquots were analyzed to provide statistically feasible number of specimens (N), i.e. at least 100 stained tests (Murray, 1991). Subsequently, the absolute abundance was converted to values proportional to volumes of 20cm³.

All the picked specimens were fixed on dark cardboard slides with tragacanth gum, identified and counted. The taxonomic identification was performed by comparing the specimens found with the species described in the literature (Cushman, 1930, 1939, 1942, 1950; Ellis and Messina, 1940 and sequences; Loeblich and Tappan, 1964, 1988; Boltovskoy et al., 1980). The species density (SD) per 20 cm³ of sediment and the Shannon index values (H; Shannon, 1948; calculated considering a N of at least 100 living foraminifera) were determined to evaluate the size and diversity of living foraminiferal communities in the study area.

3.3 Ammonia-Elphidium Index

The Ammonia-Elphidium Index (IAE) was calculated to estimate the hypoxia levels throughout the study area. Both genera are resistant to hypoxic conditions, but Ammonia shows a greater resistance than Elphidium. This index has been used in the literature to study regions affected by hypoxia due to large contribution of organic matter (Gupta et al., 1996; Gupta and Platon, 2006; Eichler et al., 2015).

The index is given by $IAE = \frac{NA}{NA + NE} \times 100$, where NA and NE are the numbers of Ammonia and Elphidium individuals, respectively, in a sample of 20cm³ of sediment.

3.4 Geochemical data

The total organic carbon (TOC) values were determined by catalytic oxidation (silver sulphate and mercury sulfate) at high temperature with reflux in a strongly acidic solution according to methods 5310A and 5310B (American Public Health Association, 2005; Grasshoff et al., 1983). The petroleum hydrocarbons were determined by Gas Phase Chromatography with Flame Ionization Detection (CG/FID), according to protocols U.S. EPA 8015 D: 2003 and ISO 9377-2: 2000 (E).

3.5 Statistical analysis

Before to be submitted to statistical analysis, data were normalized by log $x+1$. Aiming to identify possible spatial conditioning factors of the variables distribution Principal Component Analysis (PCA) was performed. This analysis was based on the species density (SD), Shannon index (H), and Ammonia-Elphidium index (A-E) values and percentage of total organic carbon (TOC), residual petroleum hydrocarbons (HRP), total petroleum hydrocarbons (TPH-T) and unresolved complex mixture (MCNR).

4. Results

The range of species density (SD), Shannon index (H), Ammonia-Elphidium index (A-E) values and TOC, residual petroleum hydrocarbons (HRP), total petroleum hydrocarbons (TPH-T) and unresolved complex mixture (MCNR) contents, for the studied stations are presented in Table 2. Foraminiferal density ranged from 0.1 to 11.7 individuals per 20 cm³ (mean $3.81 \pm 2.82 / 20 \text{~cm}^3$), for the stations of the experimental group and from 0.2 to 1.46 individuals per 20 cm³ (mean $1.46 \pm 1.04 / 20 \text{~cm}^3$) for the control group samples. The Shannon index ranged from 0 to 2.0, (mean $1.3 \pm 0.4$) and Ammonia-Elphidium index from 0 to 100 (mean $81.6 \pm 24.1$).
Tab. 2. Values of species density (SD), Shannon index (H), Ammonia-Elphidium index (A-E) and percentage of total organic carbon (TOC), HRP, TPH-T and MCNR for the sampling campaign of February 2014. Legend: SD – Standard deviation.

<table>
<thead>
<tr>
<th>Variables</th>
<th>HRP</th>
<th>MCNR</th>
<th>TPH-T</th>
<th>TOC</th>
<th>SD</th>
<th>H</th>
<th>A-E</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mg.kg⁻¹</td>
<td>mg.kg⁻¹</td>
<td>mg.kg⁻³</td>
<td>mg.kg⁻¹</td>
<td>specimens.20 cm⁻³</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Maximum</td>
<td>6.0</td>
<td>12.0</td>
<td>18.0</td>
<td>4565</td>
<td>11.7</td>
<td>2.0</td>
<td>100.0</td>
</tr>
<tr>
<td>Minimum</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>1195</td>
<td>0.1</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Mean</td>
<td>2.6</td>
<td>2.0</td>
<td>3.8</td>
<td>2465</td>
<td>3.1</td>
<td>1.3</td>
<td>81.6</td>
</tr>
<tr>
<td>SD</td>
<td>1.3</td>
<td>2.9</td>
<td>4.6</td>
<td>992</td>
<td>2.6</td>
<td>0.4</td>
<td>24.1</td>
</tr>
</tbody>
</table>

The following variables varied for: TOC from 1195 to 4565 mg.kg⁻¹ (mean 2465 ± 992 mg.kg⁻¹); HRP from 0 to 6 mg.kg⁻¹ (mean 2.6 ± 1.3 mg.kg⁻¹); MCNR from 0 to 12 mg.kg⁻¹ (mean 2 ± 2.9 mg.kg⁻¹) and; TPH-T from 0 to 18 mg.kg⁻¹ (mean 3.8 ± 4.6 mg.kg⁻¹). Higher foraminiferal densities were observed in the stations near the emissary (Fig. 2, region A) compared to the most distant stations (Fig. 2, region B). Foraminiferal densities were in general lower in the stations of the control group (Fig. 2, region C). The highest values of Shannon index were observed in the stations farthest from the emissary, but the highest Ammonia-Elphidium index values were recorded close to the SMP (Fig. 3).

4.1 Statistical analysis

The factor 1 (59%) against the factor 2 (19%) of principal component analysis (PCA) explain most of data variability (77%). The biplot of the factor 1 against the factor 2 is presented in Figure 4. Table 2 shows that the factor 1 is positively correlated to total TOC, HRP, TPH-T and MCNR and species density (SD), whereas the Factor 2 is low correlated with these geochemical variables and has positive correlation with the Shannon index (H) values.

5. Discussion

The analyzed volume of sediment ranged from 20 to 110 cm³, to obtain about 100 living specimens of foraminifera. From the outfall outlet to the stations C region, the volume

Fig. 2. Density of biocenose individuals from the experimental group (A and B) and control (C) samples from the February 2014 campaign. Distance from the outfall: from 50m to 150m (A), from 250m to 250m (B) and 5000m (C).
of 30 cm³ was used to obtain at least 100 specimens. In the regions farthest from the experimental group (C, D) and control (CO), it was necessary to analyze at least the mean volume was 90 cm³. In some samples (e.g., C3-R2, CO3-R2, CO6-R2 and CO6-R3) it was necessary to analyze a larger volume of sediment (90 to 110 cm³) to obtain about 100 living specimens of foraminifera. The density of foraminifera is low, in general, in the study area. Similar results were acquired by Duleba et al. (2007) and Teodoro et al. (2010) at the Almirante Barroso Oil Terminal.

The results of PCA suggest that foraminiferal density is related to TOC and hydrocarbon contents (HRP, TPH-T and MCNR). These results suggest that foraminifera may take advantage of organic matter as a source of food and may use also HRP, TPH-T and MCNR as food source, probably feeding on smaller molecules resulting from the degradation of these substances by bacteria. Species that manage to use the petroleum residues as food source are not being selective. *Ammonia tepida*, a frequent species in the study area, is an opportunistic species tolerating large variations of salinity, temperature, pH, among other parameters (Brashaw, 1961; Murray, 1991; Kitazato, 1994; Elshanawany et al., 2011). Due to these characteristics, this species is in general abundant in areas affected by domestic and petrochemical sewers (e.g. Seiglie, 1968; Alve, 1991, 1995; Yanko et al., 1994, 1998, 1999; Gearreta et al., 2002; Le Cadre and Debenay, 2006; Duleba et al., 2007, 2018; Teodoro et al., 2011, Delavy et al., 2016). This species is tolerant to oxygen reduction as well as the *Elphidium* genus (Gupta et al., 1996; Gupta and Platon, 2006; Eichler et al., 2015).

According to Burone et al. (2006b), the presence of *Ammonia tepida* and *Cribroelphidium excavatum* are associated with reduction of oxygen in Montevideo Bay. But these authors did not use the *Ammonia-Elphidium* index in that bay. This index was developed by studies aiming to evaluate the current and past anoxia of the Long Island Sound and Chesapeake Bay coastal regions in the United States (Gupta et al., 1996; Gupta and Platon, 2006).

In this study, the *Ammonia-Elphidium* index reached 100% in most of the A and B stations of the experimental group, which should mean that these areas present the highest number of oxygen-depleted stations. Some C and D stations have high values of *Ammonia-Elphidium* index which should have low oxic conditions (Fig. 3, region B). The *Ammonia-Elphidium* index values tended to decrease successively from the C and D stations to that of the control (CO) group (Fig. 3). These results indicate that the region closer to the emissary has a larger number of stations with oxygen deficiency than the other stations.

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**Fig. 3. Ammonia-Elphidium** index for the stations of the February of 2014 campaign. Distance from the outfall: from 50m to 150m (A), from 250m to 250m (B) and 5000m (C).
The obtained results indicate that foraminiferal assemblages are dominated by Ammonia in the most impacted zones by organic matter and organic compounds (hydrocarbons) of the study area. The most impacted areas are marked by a deep decline of foraminiferal diversity. This decay in biodiversity is not always followed by the specimens density drop. This should be a consequence of the opportunistic behavior of some species, which can tolerate adverse conditions in the presence of abundant food, such as Ammonia species.

In order to allow a more accurate analysis of the region that is affected by the emissary of the Polo Atalaia complex, larger volumes of sediments are required, since the density of living foraminifera were quite low in the region. It is suggested that the sediment volume for the study of living foraminifera is increased to at least 500 cm$^3$. The analyzes of other contaminants, such as heavy metals, should be also considered.

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