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Water toxicity assessment of Santos Bay under different climate conditions

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Abstract

Climate conditions and the movement of water masses are among the main factors that influence the destination of pollutants, acting upon transport and dispersion affecting, thus, the quality of water. Samples from Santos Bay were collected under normal conditions (08/21/2008) and during the entrance of a frontal system (08/22/2008) and tested for chronic ecotoxicity using fertilized eggs of sea urchin *Lytechinus variegatus*. Samples were toxic for stations near Xixová-Japuí State Park, Port of Santos, Submarine Outfall and Moela's Island, sites related to pollution sources. Differences in climate conditions were observed and its contribution on toxicity seemed to be significant at the entrance of São Vicente estuary and Moela's Island. At the stations in Santos Bay under influence of contamination from multiple sources such as the presence of outfall and sediments disposal from dredging, the continuous release of compounds still plays an important role in environmental quality.

Keywords: coastal management, environmental monitoring, *Lytechinus variegatus*, marine pollution, marine ecotoxicolgy.

INTRODUCTION

Many capitals and major cities around the world are located in the Coastal Zones (CZ), and consequently with the densification of cities and migration to large cities, the CZ are increasingly subjected to face an increasing number of people. The development of: (i) cities, (ii) harbors and (iii) industrial centers, combined with (iv) use of natural resources and (v) tourism and recreation are some of the major factors of environmental impacts in CZ. As a consequence, these areas are susceptible to various impacts, including pollution, since the input of large amounts of pollutants to aquatic ecosystems is a result of human activities affecting thus the environmental quality of coastal ecosystems and ecological balance.

The distribution of contaminants in aquatic environment depends on geochemical processes which controls the partition of substances that may be dissolved in water, or sorbed on suspended matter (Hedges, 2002). These processes are also involved in the transport of contaminants and, according to the residence time, contaminants may remain in the water column or precipitate in the sediments. For both conditions, it is important to emphasize the interactions between flows of energy and matter that occur among environmental compartments. Thus, it is possible to state that the pollutants originated by punctual and diffuse sources affect the quality of water, compromising its uses and its ecological functions.

Brazil has a National Policy for Water Resources (Brazil, 1997; 2006) and one of its main regulatory instruments was established by the National Environmental Council (CONAMA) through the Resolutions 357/05 and 430/11 which "Provides the framework and classification of waters, guidelines for effluent discharge, and other provisions "(Brazil, 2005; 2011).

According to these resolutions, the coastal waters can be classified as brackish and saline (seawater) and, for each class, a set of standards and benchmarks for

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different variables is presented as well as the allowable criteria. Among these variables, chemical parameters such as levels of dissolved metals, nutrients and pesticides are required, as well as biological parameters which includes the presence of coliforms and the monitoring of levels of chlorophyll-a (in case of effluent discharge). In addition, assessment of water toxicity is recommended as an environmental quality indicator.

Regarding toxicity, classical approaches include the use of bioassays by exposure of test organisms to the sample of interest. In the case of Brazilian recommendations, bioassays are carried out in order to evaluate if the chemical and physical parameters discussed in the resolution provide evidence of biological effects on aquatic organisms, since the toxicity is produced by the interactions between chemicals (contaminants) and environmental conditions (Walker *et al.*, 1996).

In coastal waters, aspects of bioavailability of contaminants and indicators of water quality must be evaluated together with climatic conditions and oceanographic aspects such as the movement of water masses (which in turn is dependent on weather conditions), since these phenomena are directly responsible for the transportation, distribution and dispersion of contaminants. In relation to these phenomena, Brauko (2008) describes a Weather Front Interface (or system) as the boundary and transitional zone between two different air masses of kilometers thick and distinct temperatures, being the mixture of air masses formed under other reference surfaces such as sea level, forming a band called Front.

These meteorological phenomena trigger the hydrodynamic circulation, producing elevations of sea level individually or in combination with increases in wave intensity and change of wind direction. Additionally, such events are often associated with a decrease in temperature, followed by the increase of rainfall and storms. Such situations, in turn, change the status of agitation of pelagic system through the stacking or mixture of large masses of water along the water-column (Quadros *et al.,* 2007; Brauko, 2008). In CZ, these climatic conditions have a direct impact on the quality of coastal waters, acting on flows and transport of matter (including pollutants), determining, thus, the extent of their effects.

In this context, the Metropolitan Region of Baixada Santista, São Paulo, Brazil, constitutes one of the most industrialized coastal regions of Latin America. In its CZ, Santos Bay suffers anthropogenic pressures from different sources, including the submarine outfall, responsible for oceanic disposal of domestic effluents from the cities of Santos and São Vicente, and diffuse sources such as irregular urban occupation in estuarine sites and urban runoff. Contributions of harbor and industrial activities are characterized by emissions from the industrial complex of Cubatão city and Port of Santos, the largest port of Latin America (Lamparelli *et al.,* 2001). Currently, both harbor and industrial activities combined with urban contribution represent the main sources of pollutants to the Region and the toxic load discharged into the estuary, reaching the Bay through coastal dynamics.

Since the sources of contamination to the region are known, the aim of this study is to evaluate the quality of waters from the Bay of Santos in two distinct situations: during normal weather conditions and during a influence of frontal system, using toxicity bioassays as indicator of water quality. In this sense, the use of short-term chronic toxicity bioassays with sea urchins is recommended due its worldwide distribution, the easy collection of adults at field, the large amounts of ovules and sperm cells obtained by *in vitro* fertilization and the rapid, clear and highly successful embryonic development (Nilin *et al.,* 2008). By these means, we aimed to assess changes on larval development of sea urchin *Lytechinus variegatus* exposed to water samples collected in different climatic conditions combined with the influence of pollutant sources.

MATERIALS AND METHODS

Climatic conditions and sampling campaigns.

In the coastal region of São Paulo state (where the Bay of Santos is located), air masses suffers frequent disturbances coming from the Frontal systems in the form of cold fronts, warm fronts and stationary systems, in addition to localized phenomena such as the formation of low pressure centers (Sant'Anna-Neto, 1990).

According to the bulletin of National Institute for Space Research (INPE, 2008), in August 2008, eight frontal systems advanced over Brazil, and four of these advanced over the coast of the Southeast region (systems 1, 3, 6 and 8). The system number 5 joined the coast and interior of the southern region on the 17th, causing rainfalls and strong winds, remaining stationary during the 18th and 19th, and being intensified by the subtropical jet stream in the troposphere.

Furthermore, according to the Bulletin, this situation contributed significantly to the formation of another low pressure center adjacent to the South region on 21st, resulting in the frontal system number 6, whose cold branch moved from the South region, in Porto Alegre-RS, to the Southeast, where it remained stationary on the 23rd (Figure 1). In Santos Bay, changes in the pattern of winds were observed as effects of the passage of this cold front: NE winds and few waves on the 21st changed for intense winds from SW and higher incidence of waves and currents on the 22nd. From the weather forecast and perspective of front system passage, two sampling campaigns were established in normal condition on the 21st and on the 22nd of August, 2008, being the day 22 corresponding to the front system.

The sampling stations were distributed along the Santos Bay and Continental Shelf of Santos, São Vicente and Guaruja cities, in areas of ecological relevance and economic importance, according to their uses: stations 1 to 3, in the vicinities of the Xixová-Japuí State Park, an important Marine Protected area inserted in the Santos Bay, under the influence

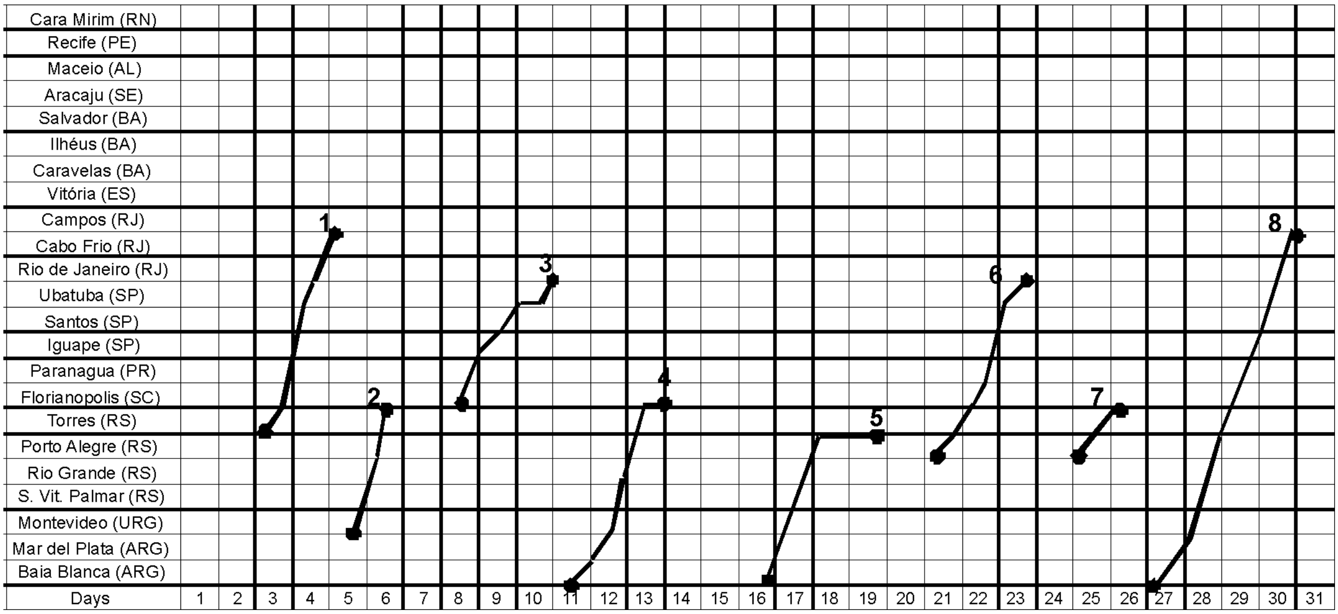

Figure 1 - Occurrence of frontal systems in Brazilian Coast in August 2008. Figure extracted and modified from INPE (2008).

of estuarine channel of São Vicente; station 4 in the area of wastewater and sewage discharges from the ocean disposal system of Santos and São Vicente; station 5 near the Guaiúba beach; and stations 6 and 7 near Moela's Island, all in Guaruja city and former areas of dredged sediments disposal from the Port of Santos; stations 8 and 9 outside the bay and current area of dredged sediments disposal. On the 21st and 22nd the stations 1 to 9 were sampled and on the 22nd, the station 10 was added and sampled at the entrance channel of the Port of Santos (figure 2).

Water sampling campaigns were conducted aboard a schooner, and the samples were collected in Van Dorn bottle. Three depths were sampled at each station (surface, middle and bottom), with the exception of stations 7, 5, 4, 3, 2 and 1

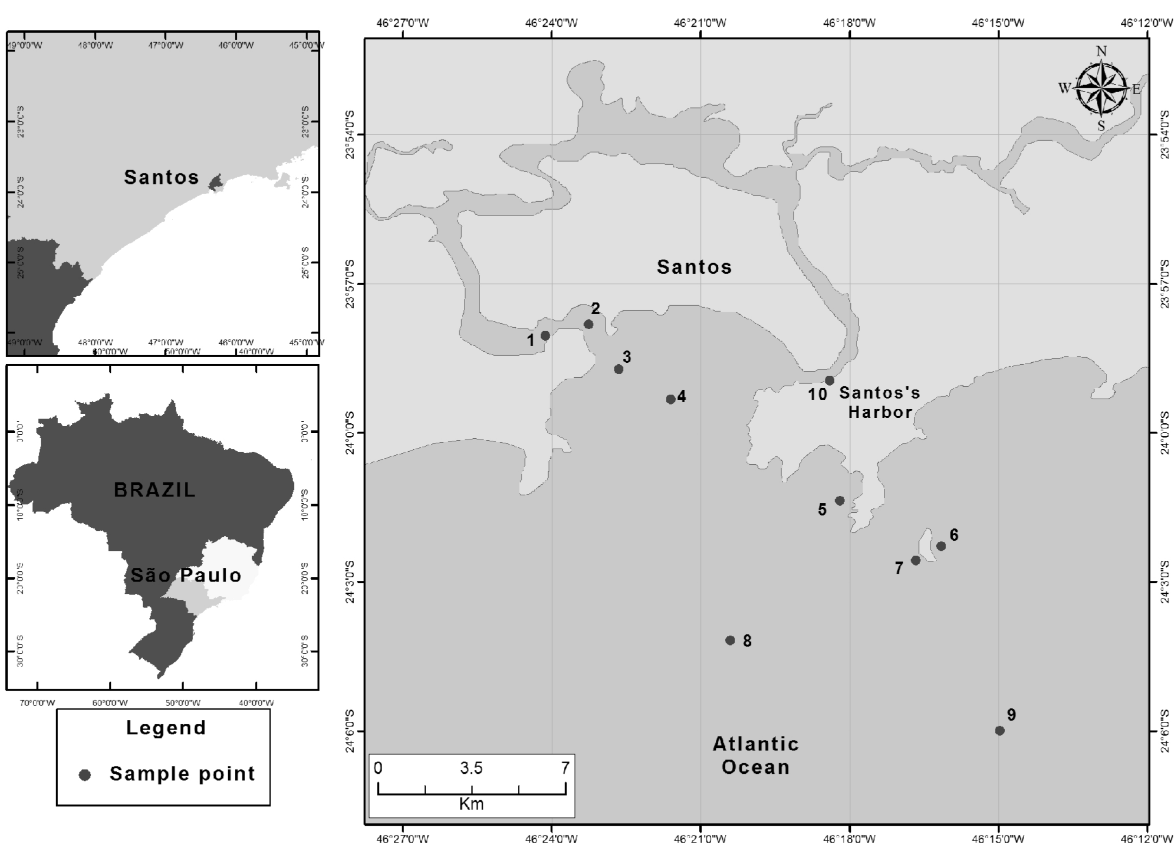

Figure 2 - Location of sampling stations in Santos Bay.

on the 21st (in normal condition), and stations 5, 4, 3, 2 and 1 on the 22nd (in frontal system), where only two depths (surface and bottom) were sampled. During sampling, temperature, salinity and pH were measured using a thermometer, portable refractometer and pH-selective electrode, respectively. After collection, samples were placed in plastic bottles and transported to the laboratory, where they were kept at 4°C until the toxicity bioassays preparation (10 days after sampling).

Toxicity Bioassays.

Samples were tested for waterborne toxicity on embryo-larval development of the sea urchin *L. variegatus* according to ABNT NBR 15350 protocol (ABNT, 2006). Adult specimens were collected in rocky shores from Palmas's Island in Santos (SP) and transported to the laboratory. After spawning, the sea urchins were tested and acclimated for 2 days and then returned to the environment. In this method, sea urchin spawning was induced and subsequently the *in vitro* fertilization was made. The test was performed by introducing approximately 400 embryos in each of four replicates, in addition to a negative control (filtered seawater). After the end of the test (24h), embryos were analyzed microscopically for morphological anomalies and delayed development. The results were expressed as percentage of development.

Negative control was prepared by using filtered and uncontaminated seawater. Positive control was prepared by testing $ZnSO_4$ at 0.5, 0.1, 0.07, 0.05 and 0.01 mg L^{-1}. The calculated value of inhibitory concentration (IC_{50}) was 0.057 ± 0.012 mg L^{-1}, in agreement with literature (Nilin *et al.,* 2008). Salinity, temperature and dissolved oxygen were controlled during the execution of tests and total ammonia concentration was measured by phenate Method 4500-NH_3C (APHA, 1999) and the unionized ammonia contents were estimated using the model proposed by Whitfield (1974). Bioequivalence hypothesis testing was used to compare responses for each sample and the control (Bertoletti *et al.,* 2007). Samples statistically different to the control were considered toxic ($p<0,05$; bioequivalence constant of 0.91).

RESULTS

The results of the variables and toxicity bioassays from water samples collected in Santos Bay at 21st and 22nd of August, 2008, are presented in Table 1. In normal condition, the salinity ranged from 27 to 37 with temperatures between 21 and 23°C, while, for pH, there was a small variation with values found between 8.4 and 8.7. Levels of total and unionized ammonia were not detected. During the entry of a front system, the variation in the salinity was smaller, ranged between 34 and 37, as well as for temperature, which was between 21 and 22 °C. A similar pattern of variation in pH was also found, and ranged from 8.2 to 8.6. Total and unionized ammonia were detected only in samples from surface of

Table 1 - Results of variables and toxicity bioassays from water samples collected in Santos Bay at 21st and 22nd of August, 2008. Toxicity results are expressed as percentage of larval development.

Stations		Normal condition (21/08/2008)								Frontal system (22/08/2008)							
		salinity	Temperature °C	pH	NH_4 mg L^{-1}	NH_3 mg L^{-1}	Toxicity			salinity	Temperature °C	pH	NH_4 mg L^{-1}	NH_3 mg L^{-1}	toxicity		
							Mean	DP	Result						Mean	DP	result
1	surface	32	23	8,5	<0,04	<0,01	80	3	**Toxic**	34	22	8,4	<0,04	<0,01	88	3	**not toxic**
	bottom	32	21	8,5	<0,04	<0,01	96	2	**not toxic**	33	22	8,4	<0,04	<0,01	92	4	**not toxic**
2	surface	32	23	8,5	<0,04	<0,01	91	2	**not toxic**	34	22	8,3	<0,04	<0,01	88	1	**not toxic**
	bottom	31	21	8,5	<0,04	<0,01	89	1	**not toxic**	34	22	8,4	<0,04	<0,01	90	2	**not toxic**
3	surface	34	23	8,5	<0,04	<0,01	73	5	**toxic**	34	22	8,5	<0,04	<0,01	89	5	**not toxic**
	bottom	35	23	8,5	<0,04	<0,01	89	5	**not toxic**	34	22	8,5	<0,04	<0,01	88	5	**not toxic**
4	surface	32	22	8,5	<0,04	<0,01	78	2	**toxic**	35	22	8,5	<0,04	<0,01	72	5	**toxic**
	bottom	27	22	8,5	<0,04	<0,01	88	7	**not toxic**	35	22	8,2	<0,04	<0,01	83	6	**not toxic**
5	surface	34	22	8,5	<0,04	<0,01	98	0	**not toxic**	35	22	8,6	<0,04	<0,01	74	2	**toxic**
	bottom	35	21	8,5	<0,04	<0,01	86	4	**not toxic**	35	21	8,5	<0,04	<0,01	88	4	**not toxic**
6	surface	35	22	8,6	<0,04	<0,01	68	9	**toxic**	36	22	8,6	<0,04	<0,01	45	26	**toxic**
	bottom	34	22	8,5	<0,04	<0,01	62	10	**toxic**	35	21	8,6	<0,04	<0,01	63	8	**toxic**
7	surface	35	22	8,6	<0,04	<0,01	71	3	**toxic**	34	22	8,5	<0,04	<0,01	96	3	**not toxic**
	middle	36	21	8,7	<0,04	<0,01	88	4	**not toxic**	35	22	8,5	<0,04	<0,01	95	1	**not toxic**
	bottom	36	21	8,6	<0,04	<0,01	77	3	**toxic**	34	21	8,4	<0,04	<0,01	95	3	**not toxic**
8	surface	35	22	8,6	<0,04	<0,01	93	2	**not toxic**	34	22	8,5	<0,04	<0,01	96	4	**not toxic**
	middle	36	21	8,6	<0,04	<0,01	93	4	**not toxic**	34	21	8,5	<0,04	<0,01	97	3	**not toxic**
	bottom	36	21	8,6	<0,04	<0,01	95	4	**not toxic**	35	21	8,5	<0,04	<0,01	87	2	**not toxic**
9	surface	36	22	8,5	<0,04	<0,01	98	2	**not toxic**	33	22	8,6	<0,04	<0,01	96	3	**not toxic**
	middle	36	21	8,4	<0,04	<0,01	96	1	**not toxic**	36	21	8,6	<0,04	<0,01	96	4	**not toxic**
	bottom	37	21	8,4	<0,04	<0,01	84	5	**not toxic**	37	21	8,6	<0,04	<0,01	97	2	**not toxic**
10	surface	-	-	-	-	-	-	-	-	35	22	8,3	0,10	0,01	79	5	**toxic**
	middle	-	-	-	-	-	-	-	-	34	22	8,4	<0,04	<0,01	75	3	**toxic**
	bottom	-	-	-	-	-	-	-	-	35	22	8,4	<0,04	<0,01	78	4	**toxic**
Control		35	24	8,4	<0,04	<0,01	97	2	**not toxic**	35	24	8,7	<0,04	<0,01	97	2	**not tóxic**

station 10. Although the results of the variables were similar for both days, changes in the sea conditions, as the increase of wave agitation was observed on 22nd, which indicated the entrance of the frontal system.

Regarding the results for toxicity bioassays, in normal condition samples from stations 1, 3, 4, 6 and 7 were toxic in surface waters and stations 6 e 7 were toxic to surface and bottom. Stations 1 to 3 are influenced by the estuary of São Vicente, where urban occupation and its urban drainage promotes inputs of different materials, such as untreated domestic wastewater and others compounds. At station 4, toxicity can be explained by the presence of the outfall, since the station is under influence of dispersion plume generated by a sewage disposal system. In stations 6 and 7, results can be associated with indirect effects of dredging operations, where the disposal of sediments promotes the increase of suspended materials, resulting in risks of adverse effects to biota.

On the other hand, with changes in sea due to the entrance of frontal system, the results of toxicity also changed. Some stations such as 1, 3 and 7, which were characterized as toxic on the first day, did not presented toxicity on the next. Samples from stations 4 and 6 remained toxic. The station 5, which was not toxic in normal conditions, presented a significant reduction in the larval development on the frontal system, with toxicity observed in surface water. Station 10 showed toxicity for all depths sampled.

DISCUSSION

Considering changes observed in climate conditions from the 21st to the 22nd of August, 2008, it was possible to observe effects in the analyzed variables, especially in toxicity results. Regarding salinity, stations 1 and 2 (32 and 31) were under the influence of the estuary of São Vicente, while station 4 (27) reflects the influence of submarine outfall, which explains the lower values found, since the estuary receives the input of freshwater drainage of local rivers and the discharges of the outfall consists of effluents marked by low salinity.

The temperature in normal wheather condition presented a greater variation among stations, with higher temperatures in surface samples, which is typical of stratification, while, on the frontal system, such difference was not observed in stations near the coast (1 to 4), which can be associated to the effect of water layers mixing. The low pH variation in both climate conditions was expected, since the carbonate system of the ocean promotes this balance through buffering reactions.

Harari & Gordon (2001) studied substance dispersion modeling in the Santos Estuarine System and the Santos Bay and reported that, under normal conditions and NE winds, the influence of the tides in material transport is restricted. However, on the other hand, the increase of strong winds and currents tends to enhance the dispersion ability.

Due to its economic and ecological importance, and to environmental pollution, aquatic environments of the Metropolitan Region of Baixada Santista have been intensely studied. Such investigations reported the occurrence of different class of compounds, such as nutrients, metals, hydrocarbons and butyltin (Braga *et al.,* 2000; Lamparelli *et al.,* 2001; Godoi *et al.,* 2003; Bicego *et al.,* 2006; Martins *et al.,* 2008). Through these studies, it is possible to relate the presence of these compounds with the toxic effects found herein, as well as changes in transport ability of waters due to climatic conditions.

Abessa *et al.* (2008) and Cesar *et al.* (2007), assessed the quality of sediments from Santos Estuarine System and found an association of higher levels of Cr, Hg, n-alkanes, aliphatic hydrocarbons and anionic surfactants with sediment toxicity to the amphipod *Tiburonella viscana* (percentage of mortality) at the entrance of the São Vicente estuary, in areas surrounding stations 1, 2 and 3 of this study. Sousa *et al.* (2008) also reported the toxicity of sediments for the same species in the navigation channel of the Port of Santos and in dredged material disposal sites, near the stations 6 and 7.

For these areas, Torres *et al.* (2009) assessed the dredging impacts analyzing the material contained in the dredges and the plume formed during dredging. The authors reported a high contamination of sediments by metals and classified such operations as potential sources of adverse risks for biota. Sediments can act as the destination of many compounds, which entering aquatic environment through the water column and, in many cases, can act as a secondary source of contamination due to transfer processes in sediment-water interface (Anderson *et al.,* 2001).

At the center of Santos Bay (station 4), Abessa *et al.* (2005) evaluated the influence of ocean disposal system of domestic effluents and reported an enrichment of mud, Ni, Hg, nutrients S and N associated with discharge of materials and sediment toxicity. These results corroborate the studies of Braga *et al.* (2000) and Aguiar & Braga (2007), which demonstrates the influence of the outfall on the distribution of nitrogen and phosphorus in the waters of Santos Bay.

Moreover, Abessa *et al.* (2005) observed the transport of contaminants to the east portion of the Bay due to the entrance of frontal system and the same condition was modeled by Harari & Gordon (2001). This situation indicates that the intense action of winds may produce a significant transport of materials released in the outfall area to adjacent beaches, and is probable that this situation occurred during the entry of the frontal system.

CONCLUSIONS

Analysis of the toxicity of waters from Santos Bay in different climatic and oceanographic conditions showed that the influence of the known sources of pollution in the region is still relevant. A reduction in toxicity was expected, however, this change occurred only in some areas such as the entrance of São Vicente estuary (stations 1 to 3) and Moela's Island (station 7). On the other hand, in the stations 4 and 6, the toxic effects in samples persisted.

The presence of outfall explains the toxicity found in station 4, while for stations 5 and 6 can be associated to the influence of sediments disposal from dredging, since such operations can act as a diffuse source of contamination. These results show that, although climatic conditions may contribute to the dispersal of pollutants, the continuous release of compounds still plays an important role in environmental quality.

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