

Characterization of phytoplankton biodiversity in tropical shipwrecks off the coast of Pernambuco, Brazil

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RESUMO – (Caracterização da biodiversidade do fitoplâncton em naufrágios tropicais da costa de Pernambuco, Brasil). A comunidade fitoplanctônica foi estudada nas adjacências de dois recifes artificiais (naufrágios Servemar-X e Servemar-I), na costa de Pernambuco, nordeste do Brasil, com o intuito de identificá-la e melhor caracterizar esses ambientes. Amostras de água foram coletadas com garrafa de Nansen em três profundidades (superfície, meio e fundo) para a análise da clorofila *a* e salinidade, obtendo-se ainda dados de temperatura e transparência da água. Para coleta do fitoplâncton, um mergulhador utilizando equipamento SCUBA realizou arrastos com uma rede de 20 µm ao redor dos naufrágios a 1,5 m do fundo, por cerca de 3 minutos. A concentração de clorofila *a* foi mais elevada no fundo variando entre 0,61 e 5,97 mg.m⁻³, com a média indicando ambiente mesotrófico. A temperatura e a salinidade apresentaram uma pequena variação sazonal e a transparência da água mostrou um padrão sazonal e espacial estando positivamente relacionada aos teores de clorofila *a*. Em relação à comunidade fitoplanctônica, o grupo das diatomáceas apareceu em maior representatividade nas amostras, seguido dos dinoflagelados, e entre as espécies sobressaiu em termos quantitativos a cianobactéria *Trichodesmium thiebautii* Gomont ex Gomont. A importância ecológica desses ecossistemas ficou comprovada através dos altos índices de diversidade e equitabilidade, sendo a influência das águas costeiras fator determinante da estrutura e diversidade da comunidade fitoplanctônica.

Palavras-chave: clorofila *a*, recife artificial, *Trichodesmium*, mergulho

ABSTRACT – (Characterization of phytoplankton biodiversity in tropical shipwrecks off the coast of Pernambuco – Brazil). The phytoplankton community was studied around two artificial reefs (shipwrecks Servemar-X and Servemar-I), located off the coast of Pernambuco, northeastern Brazil, aiming to identify and thus, better describe these environments. Water samples were collected with Nansen bottle at three depths (surface, mid-column and bottom) for chlorophyll *a* analysis and salinity; temperature and water transparency were also measured. To collect phytoplankton samples, a diver using SCUBA equipment carried out phytoplankton hauls with a 20 µm net around the shipwrecks, about 1.5 m from the bottom for approximately 3 minutes. Chlorophyll *a* concentrations at the bottom varied between 0.61 and 5.97 mg.m⁻³, with an average that indicates a mesotrophic environment. Temperature and salinity registered small seasonal variation, while water transparency showed a seasonal spatial pattern positively related to Chl-*a* rates. As regards the phytoplankton community, diatoms were the most representative group in the samples, followed by dinoflagellates, and among the species, the cyanobacteria *Trichodesmium thiebautii* Gomont ex Gomont prevailed quantitatively. The ecological importance of these ecosystems was confirmed by the high diversity and evenness indexes, with the influence of coastal waters playing an essential role in phytoplankton structure and diversity.

Key words: chlorophyll *a*, artificial reef, *Trichodesmium*, diving

Introduction

On the northeastern coast of Brazil, where oligotrophic waters prevail, there is growing interest in creating artificial reefs. Studies have revealed that such structures are indicated for the improvement of the ocean environment, even in areas of low productivity (White *et al.* 1990). Further aims of such actions are to create protected areas, increase fish stocks and offer leisure activities, such as diving and sport fishing (Seaman & Seaman 2000).

As the Brazilian state with the largest number of shipwrecks (Santos & Passavante 2007) and calm, warm, transparent waters throughout most of the year, the sinking of ships as artificial reefs has helped to strengthen the diving and ecotourism industries in Pernambuco. Within a few months on the ocean floor, these sunken vessels become coated with a variety of marine organisms (from barnacles to algae) that attract small fish, which, in turn, lure larger predators (Grossman *et al.* 1997).

Despite indications of environmental degradation in some shipwrecks, there have been no studies of these artificial reefs detailing the impact such structures have on the marine environment. Moreover, surveys regarding the

biomass, ecology and taxonomy of phytoplankton in artificial reefs are scarce in the world as a whole. In Brazil, studies have focused on investigating fish assemblages and the development of sessile biota associated with these structures (Jardewski & Almeida 2005; Krohling *et al.* 2006; Brotto & Zalmon 2007).

The phytoplankton community is extremely important for the open sea environment, as it contributes most of the organic carbon available to pelagic food chains (Reynolds 2006). The continual documentation of phytoplankton biomass and species composition can provide an invaluable record of water quality and useful information for better understanding of other types of communities and ecosystems in general (Harris 1986). In this context, chlorophyll *a* (Chl-*a*) has proven to be an excellent indicator of trophic states and a useful tool in the environmental management of coastal areas (Häkanson 1994; Passavante & Feitosa 2004).

As investigations into the phytoplankton community of artificial reefs are rare, the aim of the present study was to contribute to understanding the role of these structures in the marine environment and obtain a better characterization of phytoplankton biodiversity in tropical shipwrecks.

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Materials and methods

The two shipwrecks studied (both tugboat vessels) have the following structural characteristics: Servemar-X – 19 m in length, sunk in January 2002, 12.5 km from the coast (08°07'19"S/034°45'46"W) at a depth of 25 m; Servemar-I – approximately 20 m in length, sunk in June 2004, 9.7 km from the coast (08°06'28"S/034°46'79"W) at a depth of 23 m. The distance between the two wrecks is 2.9 km (Fig. 1). Before sinking, both vessels underwent thorough preparation, which strictly followed the guidelines of Brazilian Navy norm NORMAM-07 (Diretoria de Portos e Costas 1998).

Sampling was carried out through dives with SCUBA equipment in the morning shift in both the rainy season (May, June, July 2005) and dry season (November 2005, December 2005 and January 2006), with a total of 12 dives (6 for each shipwreck). Water samples were collected with Nansen bottles at three depths in the water column at each site (surface, mid-column and bottom – alongside the shipwreck). Up to one L of water from the samples was filtered through glass fiber filters (Whatman, 47 mm diameter and 0.45 μm pore size) and analyzed by spectrophotometry for determining Chl-*a* concentration (UNESCO 1966).

For the evaluation of hydrological parameters, water samples were also collected from each of the aforementioned depths for determining temperature and salinity. The Aladdin-Pro dive computer was used to determine temperature ($^{\circ}\text{C}$), with readings at each sampling depth. Salinity was determined using a refractometer (Hanna Instruments). A Secchi disk was used to determine water transparency. Data regarding precipitation, wind direction and wind intensity were obtained from the Center for Weather Forecasting and Climatic Studies through the Data Collection Platform of Recife, located about 15 km from the area of the shipwrecks. Tide height was obtained from the Directorate of Hydrography and Navigation of the Brazilian Navy.

Phytoplankton was collected from around the shipwrecks using a net with a 20- μm mesh (Fig. 2). Due to the physical exertion of the sampling method, hauls lasted less than three minutes, starting at the prow at about 1.5 m from the bottom and 0.5 m from the shipwreck. The collected material was placed in a recipient containing 4% buffered formaldehyde.

Species identification was based on morphological characters, using specific literature on phytoplankton systematics, biology and ecology. Frequency of occurrence was calculated based on the method described by Mateucci & Colma (1982). Relative abundance was determined using the method described by Lobo & Leighton (1986). Diversity (H') and evenness (J') were calculated using the Shannon-Weaver (1963) and Pielou (1975) indexes. Species richness was determined as the number of taxa per sample.

For the principal component analysis, hydrological and precipitation data were added to the Chl-*a* matrix. Species with over 50% frequency were

submitted to unweighted pair group method (UPGMA) cluster analysis using the Bray-Curtis index with the NTSYS 2.2 program. Analysis of variance (ANOVA) was applied using the BioEstat 3.0 program to determine significant differences ($p < 0.05$) between shipwrecks and sampling depths.

Results

In the rainy season, precipitation 48 h prior to all sampling days was recorded [maximal volume of 90 mm in June (326 mm for the entire month), 5.25 mm in May and 3 mm in July]. In the dry season, rainfall 48 h prior to sampling only occurred in December [maximal volume of 90 mm (100 mm for the entire month)] (Fig. 3). Wind direction exhibited a slight tendency toward southeasterly winds in the rainy season (average velocity = 5.4 m/s) and a predominance of northeasterly winds in the dry season (average velocity = 3.9 m/s).

Secchi vanishing-point depths ranged from six m in May to 17 m in January around Servemar-I and from 10 m in June to 19 m in January around Servemar-X. The difference between the two shipwrecks was non-significant ($p = 0.5625$). However, there was a significant difference between seasons ($p = 0.0102$), with an increase of water transparency in the dry season (Fig. 4).

There was no difference in thermal stability of the water column between seasons ($p = 0.1660$). Seasonal variation in temperature was 2.2 $^{\circ}\text{C}$ at Servemar-I and 1.7 $^{\circ}\text{C}$ at Servemar-X. Mean salinity was approximately 35‰, ranging from 33 to 38‰ at Servemar-I, with no statistically significant differences between seasons ($p = 0.5144$) or shipwrecks ($p = 0.3210$) (Fig. 5).

Chl-*a* concentration at the bottom (alongside the shipwrecks) ranged from 0.61 to 5.97 $\text{mg}\cdot\text{m}^{-3}$ at Servemar-X, with the minimum recorded in November, maximum recorded in July and January and a mean value of 3.67 $\text{mg}\cdot\text{m}^{-3}$. At Servemar-I, Chl-*a* values were lower than those found

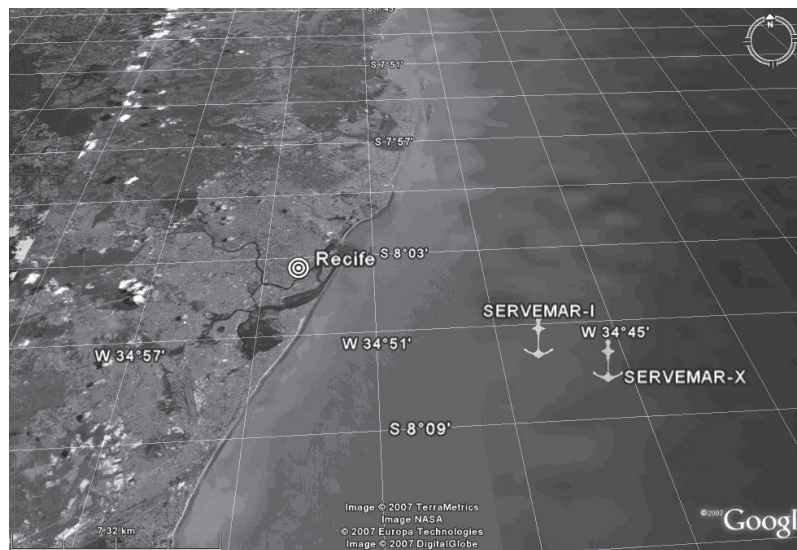


Figure 1. Map of the study area, continental shelf of Pernambuco (Northeast Coast of Brazil) and location of the shipwrecks Servemar-X and Servemar-I (Source: GoogleEarth).



Figure 2. Diver using SCUBA equipment and plankton net carried out the hauls around the shipwrecks Servemar-X and Servemar-I.

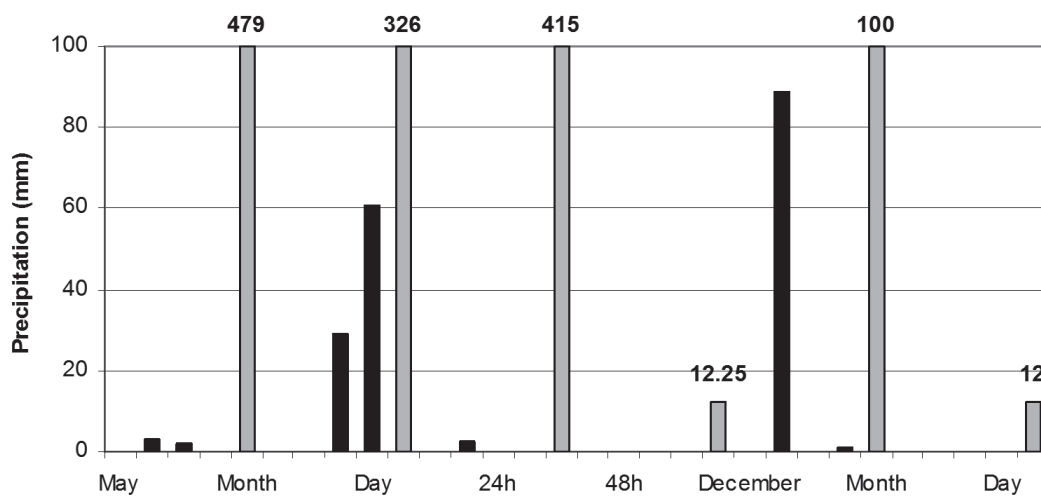


Figure 3. Pluviometric data registered 48h before sampling days (black bars) and total per month (grey bars) on the coast of Pernambuco.

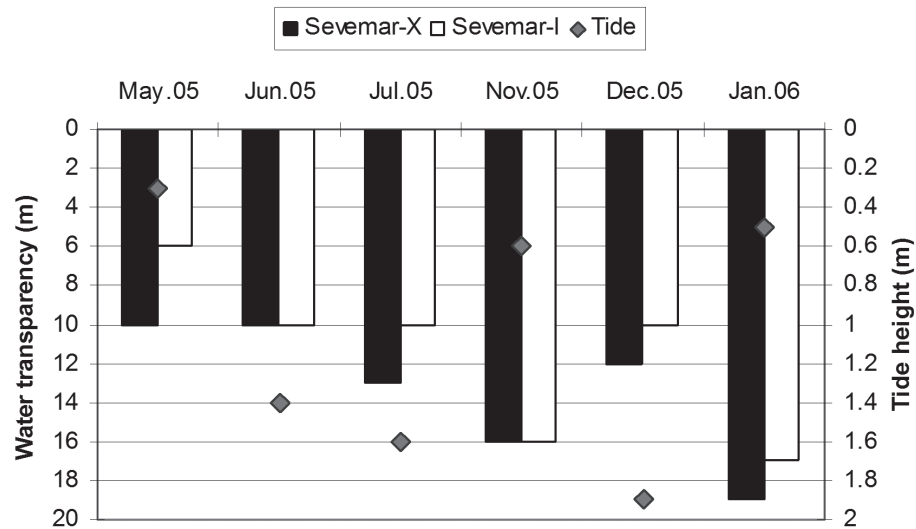


Figure 4. Water transparency and tide height recorded at the moment of sampling in the shipwrecks Servemar-X and Servemar-I.

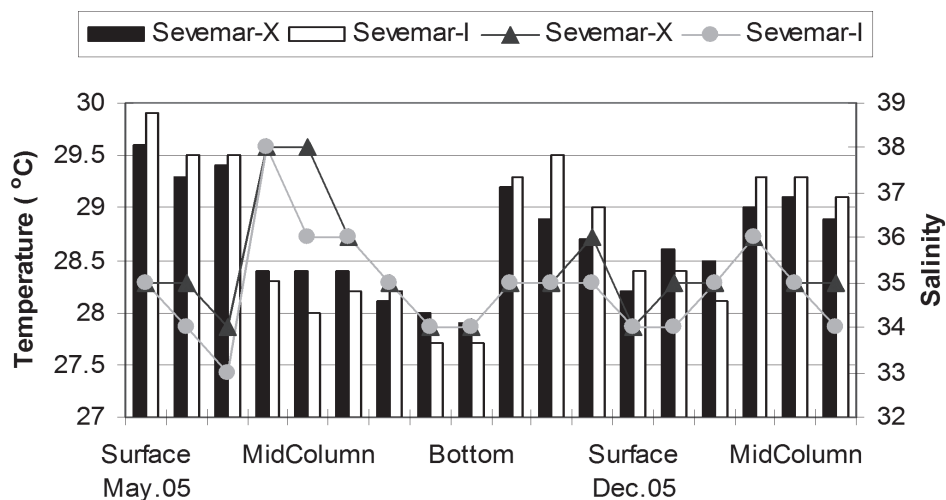


Figure 5. Temperature (bars) and salinity (lines) in the shipwrecks Servemar-X and Servemar-I.

at Servemar-X (minimum of 1.30 mg.m^{-3} in November, maximum of 5.09 mg.m^{-3} in January and mean value of 2.74 mg.m^{-3}). At the surface and mid-column, mean values were respectively 1.98 and 1.33 mg.m^{-3} at Servemar-X and 2.23 and 1.51 mg.m^{-3} at Servemar-I (Fig. 6). ANOVA revealed no significant differences in phytoplankton biomass between the two shipwrecks (bottom) ($p = 0.5869$), between surface and bottom (Servemar-X, $p = 0.1089$; Servemar-I, $p = 0.5002$) or between seasons ($p = 0.6924$).

Principal component analysis revealed a positive relationship between phytoplankton biomass and water transparency. On the other hand, these variables had a negative relationship to precipitation and salinity (Tab. 1).

The microphytoplankton was composed of 56 infrageneric taxa, 21 generic taxa and three classes.

Bacillariophyta was the most representative group (66.25%), followed by Dinophyta (18.75%), Cyanophyta (12.50%) and Euglenophyta and Chlorophytas (1.25%) (Appendix 1).

Five percent of the species were very frequent, 7.5% were frequent, 22.5% were less frequent and 65% were sporadic. The diatoms that stood out as the most frequent species were *Bacillaria paxillifera* Müller (75%), *Chaetoceros* sp. (75%), *Amplora* sp. and *Asterionellopsis glacialis* Castracane (50%). The cyanophyte *Trichodesmium thiebautii* Gomont ex Gomont also stood out in terms of frequency (91.67%) and relative abundance (mean = 58.08).

There was a distinct seasonal pattern in abundance, with an increase in phytoplankton species in the dry season ($p = 0.0054$). The following species were restricted to dry season samples: *Ceratium declinatum*, *Ceratium extensum*,

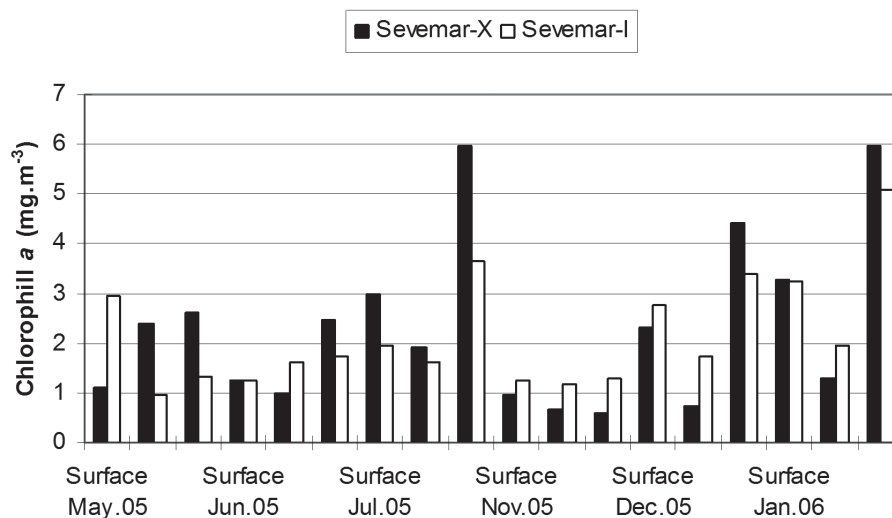


Figure 6. Chlorophyll *a* record in the shipwrecks Servemar-X and Servemar-I.

Table 1. PCA coefficient and correlation of environmental, hydrological and biological variables in the shipwrecks Servemar-X and Servemar-I.

Variables	Abbreviations	Factor 1 (31, 32%)	Factor 2 (26,79%)	Factor 3 (16,52%)
Surface Chlorophyll <i>a</i>	ChloroSurf	0,4050	0,8198	0,1138
Mid-Column Chlorophyll <i>a</i>	ChloropMidd	0,4462	0,4619	0,4639
Botton Chlorophyll <i>a</i>	ChloroBott	0,4644	0,7283	0,2746
Precipitation 48h before sampling	Pp48h	0,5495	-0,5620	0,0743
Wind Intensity 48h before sampling	WindInt48h	-0,1188	-0,1184	0,7179
Wind Direction 48h before sampling	WindDir48h	-0,3133	0,4758	-0,7602
Water transparency	WaterTransp	0,2058	0,8923	-0,1634
Light coeficient extintion	LightCoef	-0,0573	-0,4707	0,6402
Surface temperature	TempSurf	-0,9792	0,0932	0,1189
Mid-Column temperature	TempMidd	-0,9273	0,2175	-0,0788
Botton temperature	TempBott	-0,9524	0,0543	0,1595
Surface salinity	SalSurf	0,2461	-0,5045	-0,7169
Mid-Column salinity	SalMidd	0,0302	-0,6504	-0,1906
Botton salinity	SalBott	0,0984	-0,5305	0,1452
Tide	Tide	0,9291	-0,1103	-0,1515

Ceratium euarquatam, *Ceratium massiliense*, *Melchersiella hexagonalis*, *Skeletonema costatum*, *Odontella aurita*, *Rhizosolenia hebetata*, *Rhizosolenia setigera*, *Bleakeleya notata*, *Podocystis adriatica*, *Rhabdonema adriaticum*, *Striatella unipunctata*, *Gyrosigma balticum*, *Amphora egregia*, *Cylindrotheca closterium* and *Nitzschia sigma*.

Regarding species richness, Servemar-X had a higher average number of species than Servemar-I (21 species/month versus 14 species/month). Of the 80 taxa identified, 38 were found exclusively in the Servemar-X samples, 13 were found exclusively in the Servemar-I samples and 29 were found at both shipwrecks (Fig. 7).

Phytoplankton diversity and evenness were generally high, which may be related to the variability and high number of species at the shipwrecks. The lowest diversity values occurred in July at Servemar-X, which was related to the high number of *Trichodesmium thiebautii* found in the samples, accounting for 79.95% of the overall sample. At Servemar-I, *Trichodesmium thiebautii* accounted for 90.38% of the overall sample. Evenness was also low in June at Servemar-X; of the 22 species identified, three accounted for 87.73% of the overall sample: *Heliotheca thamensis* (66.52%), *Pseudonitzschia delicatissima* (13.28%) and *Trichodesmium thiebautii* (7.94%). In

July, *Trichodesmium thiebautii* accounted for 79.95% and 90.38% of relative abundance at Servemar-X and Servemar-I, respectively (Fig. 8).

The association of species revealed two distinct groups, with a cophenetic correlation coefficient of 0.88, suggesting a strong association between species (Fig. 9). The first group united coastal and oceanic species, such as *Trichodesmium thiebautii*, *Bacillaria paxillifera* and *Asterionellopsis glacialis*, with a higher average relative abundance in comparison to the second group: *T. thiebautii* (29.80%), *B. paxillifera* (16.31%) and *A. glacialis* (15.23%). The second group also comprised three oceanic dinoflagellates (*Ceratium fusus* Ehrenberg, *Ceratium pentagonum* Gourret and *Ceratium deflexum* Kofoid) and one oceanic diatom (*Amphora* sp.), with much lower average relative abundance (0.74, 1.07, 1.12 and 0.79%, respectively).

Discussion

Mean Chl-*a* concentrations in the present study were high - more than half of those reported for shelf waters off northeastern Brazil (Passavante *et al.* 1987/89; Passavante & Feitosa 1995; Ekau & Knoppers 1996; Medeiros *et al.* 1999). Based on the Håkanson (1994) classification, these values indicate mesotrophic environments near the shipwrecks. Despite the lack of a statistically significant difference between sampling depths, the greater concentration of phytoplankton biomass at the bottom may be associated with the existence of the shipwrecks, with a greater concentration of Chl-*a* at this depth.

While Chl-*a* concentration in waters of the continental shelf may be higher at the bottom (Medeiros *et al.* 1999), the comments of Seaman & Seaman (2000) regarding turbid currents are pertinent to explaining this particular situation alongside the shipwrecks. Once settled on the ocean floor,

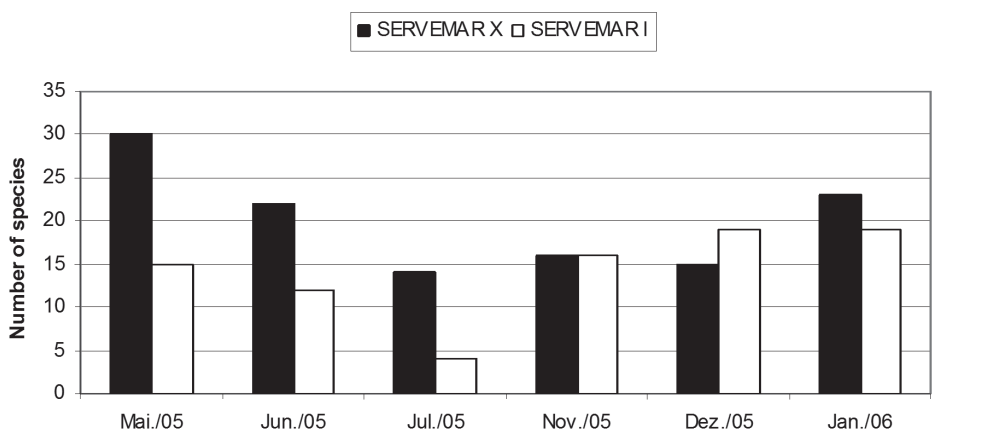


Figure 7. Total number of phytoplankton species found in the samples of shipwrecks Servemar-X and Servemar-I.

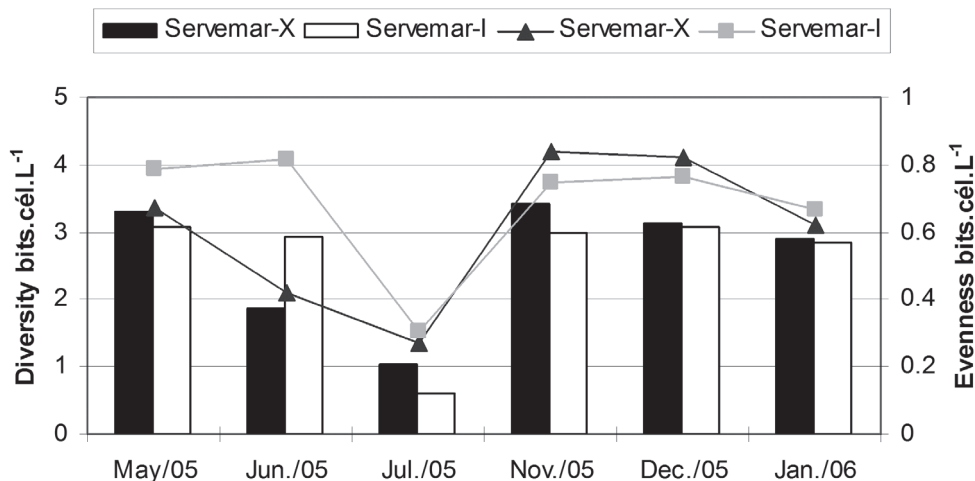


Figure 8. Diversity (bars) and evenness (lines) of phytoplankton in the shipwrecks Servemar-X and Servemar-I.

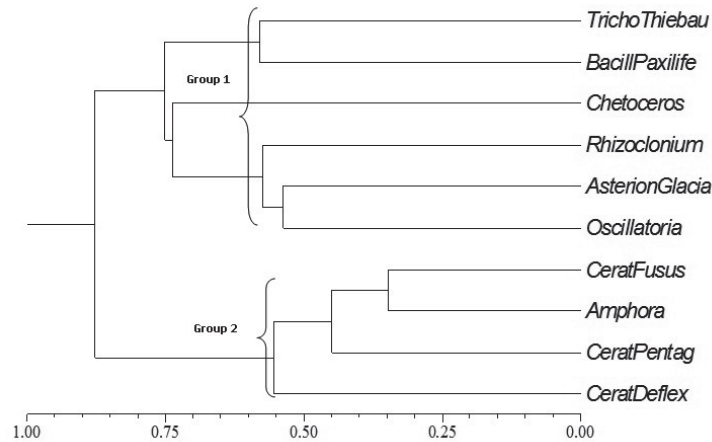


Figure 9. Dendrogram of the phytoplankton species $\geq 50\%$ frequency found in the samples of shipwrecks Servemar-X and Servemar-I. Abbreviations: TrichoThiebau = *Trichodesmium thiebautii*; BacillPaxilife = *Bacillaria paxilifera*; Chetoceros = *Chaetoceros* sp.; Rhizoconium = *Rhizoconium* sp.; AsterioGlacia = *Asterionellopsis glacialis*; Oscillatoria = *Oscillatoria* sp.; CeratFusus = *Ceratium fusus*; Amphora = *Amphora* sp.; CeratPentag = *Ceratium pentagonum*; CeratDeflex = *Ceratium deflexus*.

artificial reefs become obstacles to currents, which undergo detours, even on a small scale, with a consequent stirring up of sediment. This mechanism allows nutrient recycling and regeneration in the water column, which is necessary for phytoplankton growth (Rezende & Brandini 1997). Thus, the bottom-up effect seems to be the main factor for the high rates of phytoplankton biomass alongside the shipwrecks. As the ocean floor is moved by currents and the water is clear, allowing light to reach the bottom, these structures offer ideal conditions for phytoplankton growth. This may explain why phytoplankton diversity, biomass and production are higher in areas of natural or artificial reefs, such as sunken ships, although on a much smaller scale than that of large structures (Wilding & Sayer 2002).

Water transparency in the present study exhibited seasonal variation, with higher values in the summer. On the continental shelf off the coast of Pernambuco, previous studies have reported such seasonality, with Secchi values greater than 25 m (Resurreição *et al.* 1996). Regarding the Servemar-I, which is located closer to the coast, the variation in water transparency was 13 m, whereas the variation was seven m at Servemar-X. This clearly reveals the influence of continental runoff. Although the shipwrecks are rather distant from the coast, *in situ* observations (especially during the rainy season) indicate that discharge from the Capibaribe River can reach distances of over 9 km from the coast, which is confirmed by decrease in the photic layer around Servemar-I as well as amount of waste (plastic bags, bottles and vegetation of continental origin – e.g., *Eichornia crassipes*). Eskinazi-Leça *et al.* (1997) report the influence of this discharge on phytoplankton growth, with the waters of the Capibaribe River reaching four miles from the Port of Recife.

The slight variation in temperature and salinity values in the areas of the shipwrecks was expected and had no influence on phytoplankton growth. A salinity of 33‰

at Servemar-I in May may be associated with rain and freshwater inputs.

The phytoplankton community around the shipwrecks was characterized by a mixture of oceanic and coastal species, with considerable influence from the mainland, where water from Pina Basin meets oligotrophic oceanic waters. An indication of this are the freshwater species *Oscillatoria perornata* Skuja, *Stichosiphon sansibaricus* Geitler and *Fragilaria capucina* Desmazières, which accounted for 3.6% of the overall sample, as well as the occurrence of Euglenophyceae. Morphological analyses of these algae revealed the absence of pigment, indicating that they may have been brought by the currents and tended not to survive in the waters of the continental shelf. Estuarine species (which live in brackish water), such as *Gyrosigma balticum* and *Phormidium retizzi* Gomont, were found, possibly originating from inshore locations across the Pina Basin, and accounted for 7.2% of the taxa. Oceanic and neritic species corresponded to 69.6% of the sample and tytoplankton accounted for 19.6%.

The predominance of diatoms at both shipwrecks may be explained by the characteristics of the group, which is considered the largest of the marine phytoplankton, with wide geographic distribution, high growth rates and the ability to tolerate variations in environmental conditions (Bold & Wynne 1985). Most of the species correspond to what has been recorded for coastal waters off Brazil (Fenandes & Brandini 2004; Souza *et al.* 2008). *Asterionellopsis glacialis* is a common, widespread Atlantic near-shore and estuarine species (Johnson & Allen, 2005) that is frequent and abundant in other coastal regions as well (Videau *et al.* 1998; Varela & Prego 2003).

The metallic structures of the shipwrecks may also have a positive effect on the development of microflora. Studies have demonstrated the particular importance of iron in the

development of marine phytoplankton as a potential factor in the regulation of phytoplankton growth in coastal and oceanic regions (Martin & Gordon 1988; Martin 1994). Laboratory experiments have proven that addition of iron increases the formation of diatomic chains up to sixfold for the genera *Chaetoceros* and *Pseudonitzschia* (Graham & Wilcox 2000).

Some tychoplankton, such as *Bacillaria paxillifera* and *Nitzschia longissima* Brébisson, may also be associated with the shipwrecks, attached to the structure as a periphyton and eventually detaching and integrating the phytoplankton community. Silva (1982) reports the importance of these organisms on the continental shelf off the coast of Pernambuco. With time, the development of a shipwreck as a reef ecosystem contributes to the natural enrichment of the water, creating ideal conditions for the growth of this kind of species.

The abundance of dinoflagellates was greater at Servemar-X than at Servemar-I, especially the genus *Ceratium* Schrank. The species of this genus have cosmopolitan distribution from eurythermal and euryhaline areas of oceanic and neritic waters (Smalley & Coats 2002). Off Pernambuco, the coastal water is less rich in terms of *Ceratium* species (Koenig & Lira 2005). With increasing distance from the coast, the decrease in relative abundance of diatoms is followed by an increase in dinoflagellates (Zhou *et al.* 2008).

Found in 91.6% of the samples, *Trichodesmium thiebautii* is a colonial, nitrogen-fixing, filamentous cyanobacterium, responsible for a significant portion of total primary production of oceanic surface waters (Marañón *et al.* 2000). On the Brazilian coast, a number of studies have recorded *Trichodesmium erythraeum* Ehrenberg causing occasional blooms (Satô *et al.* 1963/64; Medeiros *et al.* 1999; Rosevell-Silva 2005; Rörig 1998). Clayton (2000) found an abundant population of *T. thiebautii* in samples collected from the Gulf of Mexico in an area with several artificial reefs (oil rigs). This researcher reports that colonies were commonly found in parallel bunches, which was also observed in the present study. In the Atlantic Ocean, high *Trichodesmium* abundance has been found to be correlated with shallow, mixed-layer depths and high estimated iron deposition on the ocean floor (Tyrrell *et al.* 2003).

Servemar-X had greater species richness in the rainy period, whereas Servemar-I had greater species richness in the dry season. Spatial fluctuations in species richness have been described for the coast of Pernambuco (Resurreição *et al.* 1996; Eskinazi-Leça *et al.* 1997), with phytoplankton blooms occurring in autumn and winter more than five miles offshore. Closer to the mainland, freshwater inputs decrease light penetration in the rainy season; thus, phytoplankton rates increase only in spring and summer, when the water is clear.

Diversity and evenness values confirm the ecological importance of these ecosystems. Equilibrium among

taxa indicates stable conditions in the area, which is characteristic of open sea environments. Moreover, the occurrence of coastal species reveals the connection between land and ocean environments, with strong interaction between these communities.

Conclusion

There is evidence that shipwrecks are associated with high concentrations of chlorophyll *a* on the ocean floor, with turbid currents and a bottom-up effect, acting as potential factors for phytoplankton growth alongside sunken vessels. However, the influence of enriched coastal waters is a key factor in phytoplankton structure and the local food chain, contributing to the high diversity of species found at the shipwrecks.

The shipwreck situated farther off the coast had a higher concentration of chlorophyll *a*, greater number of taxa and greater species richness and diversity than the wreck located closer to shore, which was more affected by continental runoff, as evidenced by the occurrence of freshwater species, such as *Oscillatoria perornata*, *Stichosiphon sansibaricus* and *Fragilaria capucina*. Moreover, distance from the coast led to two different seasonal patterns, with an increase in phytoplankton biomass and richness in the rainy season at Servemar-X and in the dry season at Servemar-I.

The sampling method around the shipwrecks using SCUBA equipment and plankton nets proved successful and can be extended to other, even deeper areas, including natural reefs. The use of a small mesh size (< 65 µm) will increase diver effort during hauls, thereby slowing down the collection procedure. Strong currents can also hinder sampling. Sampling with several divers at the same site is not recommended. Moreover, divers should avoid swimming close to the bottom and stirring up the sediment.

Acknowledgements

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Appendix 1. List of species, relative abundance (average), frequency, shipwreck and ecology of the phytoplankton collected at Servemar-X and Servemar-I. VF = Very Frequent ($\geq 75\%$), F = Frequent ($<75\%$ and $\geq 50\%$), FF = Few Frequent ($<50\%$ and $\geq 25\%$), S = Sporadic ($<25\%$); X = Servemar-X, I = Servemar-I; O = Oceanic/Neritic, C = Coastal, E = Estuarine, Fw = Freshwater, T = Tycho planktonic.

Species	Relative Abundance Rainy Season (Mean)	Relative Abundance Dry Season (Mean)	Frequency	Shipwreck	Ecology
CYANOPHYTA					
Cyanophyceae	-	1.73	FF	X and I	
<i>Oscillatoria perornata</i> Skuja	-	0.65	S	I	Fw
<i>Oscillatoria</i> sp.	0.90	7.40	F	X and I	
<i>Trichodesmium thiebautii</i> Gomont ex Gomont	37.31	20.77	VF	X and I	O
<i>Lyngbya</i> sp.	-	1.50	F	X and I	
<i>Phormidium anomala</i> Rao	-	0.65	S	X	E
<i>Phormidium retizzi</i> Gomont	-	3.64	S	I	E
<i>Phormidium</i> sp.	-	1.30	S	X	
<i>Spirulina</i> sp.	-	0.65	S	I	
<i>Stichosiphon sansibaricus</i> Geitler	-	1.79	S	I	Fw
EUGLENOPHYTA					
<i>Euglenophyceae</i>	0.97	10.14	FF	X and I	
DINOPHYTA					
<i>Dinophysis caudata</i> Stein	0.20	-	S	X	C
<i>Ceratium contortum</i> (Gourret) Cleve	0.13	-	S	X	O
<i>Ceratium declinatum</i> (Karsten) Jörgensen	-	3.95	S	X	O
<i>Ceratium deflexum</i> (Kofoid) Jörgensen	0.25	2.00	F	X and I	O
<i>Ceratium extensum</i> (Gourret) Cleve	-	0.65	S	X	O
<i>Ceratium euarcuratum</i> Jörgensen	-	0.65	S	X	O
<i>Ceratium furca</i> (Ehrenberg) Claparède	0.17	1.02	FF	X and I	C
<i>Ceratium fusus</i> (Ehrenberg) Dujardim	0.65	0.90	F	X and I	C
<i>Ceratium massiliense</i> (Gourret) Jorgensen	-	0.65	S	X	O
<i>Ceratium pentagonum</i> Gourret	0.60	1.53	F	X and I	O
<i>Ceratium porrectum</i> Karsten	3.85	-	S	X	O
<i>Ceratium tripos</i> (Müller) Nitzsch	0.60	-	S	X	O
<i>Ceratium trichoceros</i> (Ehrenberg) Kofoid	1.97	1.82	FF	X and I	O
<i>Protoperidinium</i> sp.	0.21	-	FF	X	
<i>Protoperidinium oblongum</i>	0.65	-	FF	X and I	O
BACILLARIOPHYTA					
Bacillariophyceae	-	14.65	FF	X and I	
Coccinodiscophyceae	0.43	-	S	X	
<i>Melchersiella hexagonalis</i> Teixeira	-	1.80	FF	X	C
<i>Skeletonema costatum</i> (Greville) Cleve	-	1.82	S	X	C
<i>Odontella aurita</i> (Lyngb.) Agardh	-	0.65	S	X	T
<i>Odontella</i> sp.	-	1.79	S	X	
<i>Dimerogramma dubium</i> (Grunow) Grunow in van Heurk	0.20	-	S	X	T
<i>Isthmia enervis</i> Ehrenberg	0.20	1.82	FF	X	T
<i>Hemiaulus membranaceus</i> Cleve	21.08	2.17	FF	X and I	C
<i>Hemiaulus</i> sp.	0.29		S	X	
<i>Lithodesmium</i> sp.	-	0.65	S	X	
<i>Heliotheca thamensis</i> Shrubsole (Ricard)	33.17	-	FF	X and I	C

Continue

Appendix 1. Continuation.

Species	Relative Abundance Rainy Season (Mean)	Relative Abundance Dry Season (Mean)	Frequency	Shipwreck	Ecology
<i>Guinardia</i> sp.	-	-	S	X	
<i>Proboscia alata</i> (Brightwell) Sundström	1.72	0.60	FF	X and I	C
<i>Rhizosolenia hebetata</i> Brightwell	-	0.60	S	I	O
<i>Rhizosolenia setigera</i> Brightwell	-	0.65	S	X	O
<i>Rhizosolenia styliformis</i> Brightwell	3.22	7.25	FF	X and I	O
<i>Rhizosolenia</i> sp.	-	0.65	S	X	
<i>Chaetoceros affinis</i> Lauder	2.90	-	FF	X and I	O
<i>Chaetoceros brevis</i> Schütt	0.20	-	S	X	O
<i>Chaetoceros coartactus</i> Lauder	1.40	-	FF	X and I	O
<i>Chaetoceros didymus</i> Ehrenberg	0.20	-	S	X	O
<i>Chaetoceros</i> sp.	4.22	2.49	VF	X and I	
<i>Leptocylindrus danicus</i> Cleve	9.22	-	S	I	C
<i>Fragilaria capucina</i> Desmazières	0.13	-	S	I	Fw
<i>Asterionellopsis glacialis</i> (Castracane) Round	0.87	18.11	F	X and I	C
<i>Bleakeleya notata</i> (Grunow) Round	-	30.33	FF	I	C
<i>Podocystis adriatica</i> Kützing	-	0.60	S	I	T
<i>Licmophora</i> sp.	-	1.79	S	X	
<i>Thalassionema frauenfeldii</i> Grunow	2.20	5.00	FF	X and I	O
<i>Thalassionema nitzschioides</i> Grunow	7.55	-	FF	X and I	O
<i>Rhabdonema adriaticum</i> Kützing	-	1.96	F	X and I	T
<i>Rhabdonema punctatum</i> (Harvey & Bailey) Stodder	0.20	-	S	X	T
<i>Rhabdonema</i> sp.	-	1.79	S	X	
<i>Striatella unipunctata</i> (Lyngbye) Agardh	-	3.95	S	I	C
<i>Striatella</i> sp.	-	1.79	S	X	
<i>Lyrella lyra</i> (Ehrenberg) Karayeva	0.94	3.64	FF	X	T
<i>Diploneis bombus</i> Ehrenberg	0.14	-	S	X	T
<i>Navicula</i> sp.	-	3.64	S	X	
<i>Pleurosigma</i> sp.	0.29	0.72	FF	X and I	
<i>Pleuro/Gyrosigma</i> sp.	1.91	0.96	F	X and I	
<i>Gyrosigma balticum</i> (Ehrenberg) Cleve	-	1.95	S	X	E
<i>Amphora egregia</i> Ehrenberg	-	0.72	S	I	C
<i>Amphora ostrearia</i> Ehrenberg	0.49	-	S	I	C
<i>Amphora</i> sp.	0.50	1.07	F	X and I	
<i>Bacillaria paxillifera</i> (Müller) Hendey	14.99	17.37	VF	X and I	T
<i>Cylindrotheca closterium</i> Ehr. (Reiman & Lewis)	-	0.65	S	X	T
<i>Nitzschia longissima</i> (Brébisson) Grunow	0.40	0.65	FF	X	T
<i>Nitzschia sigma</i> (Kützing) Wm. Smith	-	0.60	S	I	T
<i>Nitzschia</i> sp.	0.74	0.65	FF	X	
<i>Pseudonitzschia delicatissima</i> Cleve	10.86	-	FF	X and I	C
<i>Pseudonitzschia pungens</i> Cleve	1.20	5.42	FF	X	C
<i>Auricula complexa</i> (Gregory) Cleve	0.13	-	S	X	T
CHLOROPHYTA					
<i>Rhizoclonium</i> sp.	2.91	6.60	VF	X and I	