

Tracking of spatial changes in the structure of the zooplankton community according to multiple abiotic factors along a hypersaline lagoon

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ABSTRACT

In this study we evaluated the effects of changes in salinity, temperature, pH and dissolved oxygen on the spatiotemporal variation of zooplankton. Samples were collected in January–March, May; October–December 2010; and January–March 2011 in the Araruama lagoon, state of Rio de Janeiro, Brazil. There were eight fixed stations sampled using a WP2 net equipped with a flow meter. The zooplankton diversity of the Araruama lagoon was low and dominated by Cirripedia larvae and by the copepod *Acartia tonsa*, which is an indicator species of eutrophication. In general, a few species from the Cabo Frio region were able to adapt to the conditions of this hypersaline lagoon. In addition, a specimen of *Monstrilla bahiana* (Monstrilloidea) was found at a salinity of 46‰. This is the first record of the order Monstrilloidea in the region of Cabo Frio. Temperature, salinity and pH were shown to be limiting factors for the species present in the lagoon since Cirripedia seemed to avoid releasing their larvae under unfavorable environmental conditions. The abundance of *A. tonsa* was always associated with that of Cirripedia larvae, and it was higher at cooler temperatures.

KEYWORDS

Acartia tonsa, Araruama lagoon, bioindicator species, temperature.

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INTRODUCTION

Hypersaline marine ecosystems are very limited in extent in Brazil and worldwide and have been poorly studied (Cotner, 2004). Brazil has a few hypersaline water bodies such as the Araruama lagoon, Pernambuco lagoon and Vermelha lagoon, Mossoró river (Silva *et al.*, 2004a; Silva *et al.*, 2004b; Serpe *et al.*, 2010). A striking feature of Araruama lagoon is its high salinity, which is twice that of seawater. The high salinity allows the extraction of natural salt, a traditional activity in the region (Coutinho *et al.*, 1999; Carvalho *et al.*, 2014).

In Brazil, many aquatic environments are polluted with untreated sewage (Aslan *et al.*, 2017), bays and estuaries are some of the most affected environments (Coelho-Botelho *et al.*, 1999; Pereira, 2007; Carvalho *et al.*, 2014; Costa, 2015). Since the 1980s, the Araruama lagoon has been subjected to sewage contamination (Pereira, 2007) due to an increased number of summer tourists and growth of the resident populations of Araruama and neighboring towns (Coutinho *et al.*, 1999; Pereira, 2007; Carvalho *et al.*, 2014). The continuous discharge of sewage in the lagoon has decreased the salinity and increased the nutrient loading (Souza *et al.*, 2003; Rosa *et al.*, 2016a). After many years of sewage discharge, Araruama is now considered as an eutrophic lagoon (Carvalho *et al.*, 2014). Moreover, its average salinity is currently 41‰, which is higher than that of the sea of Cabo Frio where the lagoon water exchange occurs (Rosa *et al.*, 2016a).

Eutrophication influences the planktonic community (Costa *et al.*, 2018), and globally, the zooplankton community of hypersaline environments is usually poor in diversity and abundance. Indeed, in a previous study at the Araruama lagoon the copepod assembly was represented by three taxa only (Coutinho *et al.*, 1999), whereas the copepod assembly of the adjacent Cabo Frio region has 39 taxa (Rosa *et al.*, 2016b). The present study aimed to evaluate whether abiotic factors influence the spatial and temporal variation of zooplankton in the Araruama lagoon, and to compare the zooplankton diversity with the assembly of copepods of Cabo Frio, to understand which species of the region can adapt to the extreme conditions of the lagoon.

MATERIALS AND METHODS

Study area

The Araruama lagoon occupies 210 km² and is located on the coast of the state of Rio de Janeiro, Brazil, between latitudes 22°40'S and 22°57'S and longitudes 42°00'W and 42°23'W (Castro *et al.*, 1999). The lagoon is bound by the following cities: Araruama, Arraial do Cabo, Cabo Frio, São Pedro da Aldeia and Iguaba (Carvalho *et al.*, 2014). One of the main characteristics of the lagoon is its high salinity, due to its size (around 200 km²).

Sampling

Abundance and zooplankton composition was obtained in eight sites chosen according to their proximity to sewage discharges: Boqueirão (station 1), Monte Alto (station 2), Centro de São Pedro da Aldeia (station 3), Enseada de Iguaba (station 4), Ponta do Acaíra (station 5), Barbudo (station 6), Centro de Araruama (station 7) and Ponta dos Excursionistas (station 8) (Fig. 1). The highest discharge of untreated sewage occurs at stations 2, 3 and 7.

In 2010, samples were taken monthly from January to June, October to December, and in 2011, from January to March. During the data sampling, the state of the tide (ebb or flood) could not be standardized at the time of sampling, but previous data reveal that the tide has no significant influence on salinity from the Boqueirão sampling site (station 1) to Ponta dos Excursionistas (station 8) (Rosa *et al.*, 2016a). However, the general scenario of tidal trends ranged between ebb and flood (January 2010 ebb; February 2010 ebb; March 2010 ebb; May 2010 flood; October 2010 ebb; November 2010 ebb; December 2010 flood; January 2011 ebb; February 2011 ebb; March 2011 flood). The tide data was obtained from the site of the Brazilian Navy (<https://www.marinha.mil.br/chm/>) (Centro de Hidrografia da Marinha, Marinha do Brasil).

Eight samples were taken per site at each month, giving a total of 80 samples collected by means of horizontal surface hauls with a net with a 60 cm diameter, 200 µm mesh and coupled flowmeter. The samples were fixed in a 4% formalin solution diluted with water from the lagoon previously neutralized with sodium tetraborate.

The abundance of zooplankton (individuals.m⁻³) was estimated using Stempel pipette subsampling

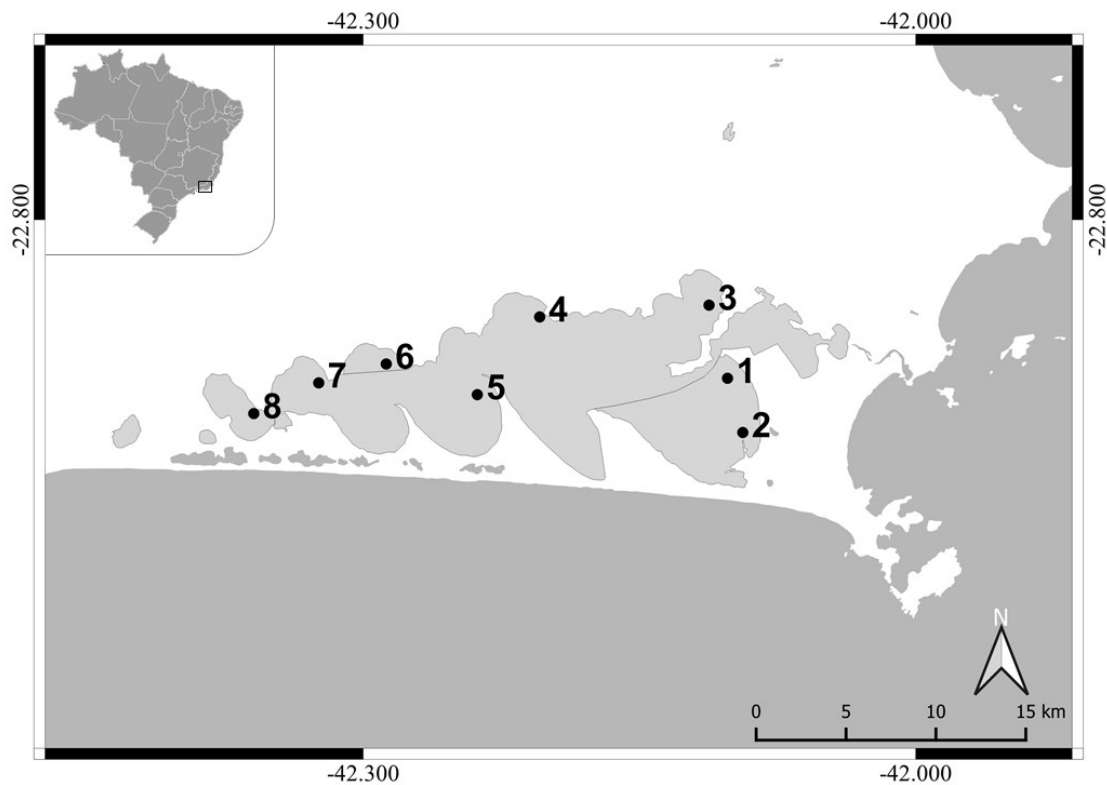


Figure 1. Map of the coast of the state of Rio de Janeiro pointing out the 8 sampling stations of the Araruama lagoon.

(2.68 ml). Taxonomy and species ecology were based on Boltovskoy (1981; 1999). To evaluate the potential effect of continental runoff upon plankton abundance, major environmental parameters, namely salinity, temperature, dissolved oxygen (mg.L^{-1}) and pH, were measured. For analysis of abiotic data the water samples were measured in the field and with a single value for each parameter, except pH, which was measured in the laboratory. The salinity was measured by a refractometer, temperature and oxygen dissolved by a portable oximeter (ITT datalogger 71440) and pH was measured by a portable pH meter (SKU: SP3630-45). These parameters were considered related to the continental flow, mainly discharge of sewage in estuaries and bays (Pereira, 2007).

Data analysis

The community structure was described in terms of the Shannon-Weaver diversity index (H') and the Pielou's uniformity index (J'). To evaluate the effects of the abiotic data on the zooplankton community, the relative abundance of the organisms with the highest frequency of occurrence were correlated (linear regression) with temperature, salinity, and pH.

A multiple regression was performed in order to understand which parameter(s) (temperature, salinity, and pH) most influenced the abundance of Cirripedia and *Acartia tonsa*. We also used a correlation to investigate the temporal variation in the relative abundance of Cirripedia with that of the copepod *Acartia tonsa*. From the linear regression results, the month of May 2010 was chosen to make the multiple regression, since this month the larvae correlated well with the three parameters (temperature, salinity, and pH).

RESULTS

Salinity ranged seasonally from 42‰ in autumn (May 2010) to 53‰ in summer (February 2010), with an annual average of 45‰. The lowest temperature was recorded in October 2010 (23.3 °C), and the highest, in January 2010 (32.1 °C). Mean annual temperature was 27.9 °C. Dissolved oxygen ranged from 3.08 to 10 mg.L^{-1} with an overall average of 6.36 mg.L^{-1} , while pH was lower in February 2011 (6.2) and higher in January 2010 (9.3). The overall average pH was 8.15 (Fig. 2).

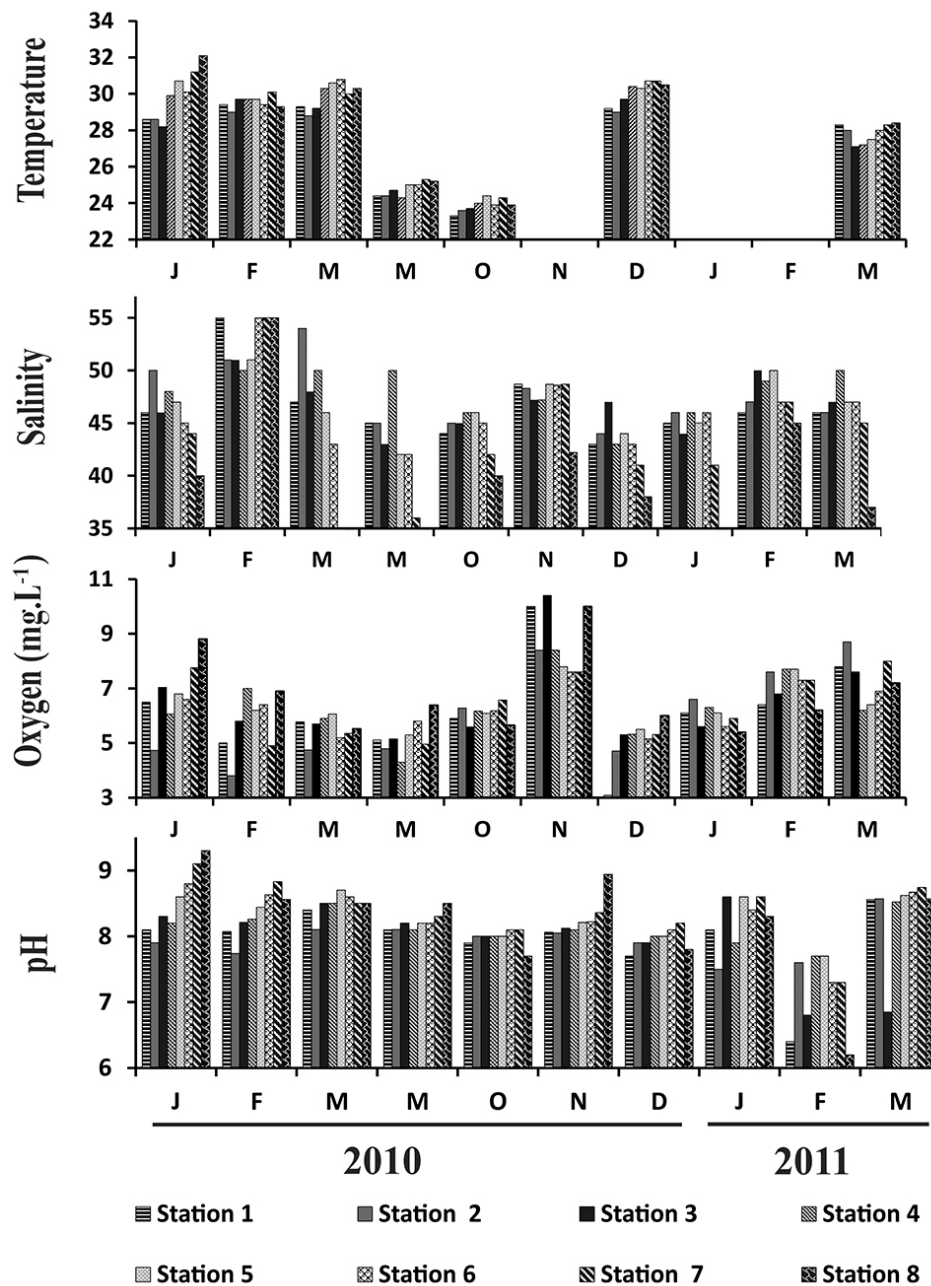


Figure 2. Variation of multiple factors (temperature, salinity, oxygen (mg.L^{-1}), and pH) in each collection station over the sampled months of 2010 and 2011.

The temperature, salinity, oxygen, and pH influenced the spatial variation of the larvae of Cirripedia in Araruama lagoon: at lower temperatures and pH and higher salinity the density of the Cirripedia larvae was higher. There was a negative correlation between temperature and pH and Cirripedia larvae abundance in terms of spatial variation and a positive correlation between salinity and Cirripedia larvae abundance (Tab. 1 and Fig. 3). The multiple regression analysis indicated that the three variables (temperature, salinity, and pH)

had the greatest influence on the spatial variation of Cirripedia larvae ($R^2 \text{ adj} = -0.67$, $p = 0.00$). In the spatial correlation between *A. tonsa* and abiotic data no significant results were found. (Tab. 2). However, *A. tonsa* had a negative correlation with temperature ($R^2 = -0.68$; $p = 0.02$), but for the other parameters no correlations were found. For the Cirripedia larvae, no significant results were found for temporal variation (Tab. 3). The density of *A. tonsa* was correlated with that of Cirripedia larvae ($R^2 = 0.52$; $p = 0.02$) (Fig. 4).

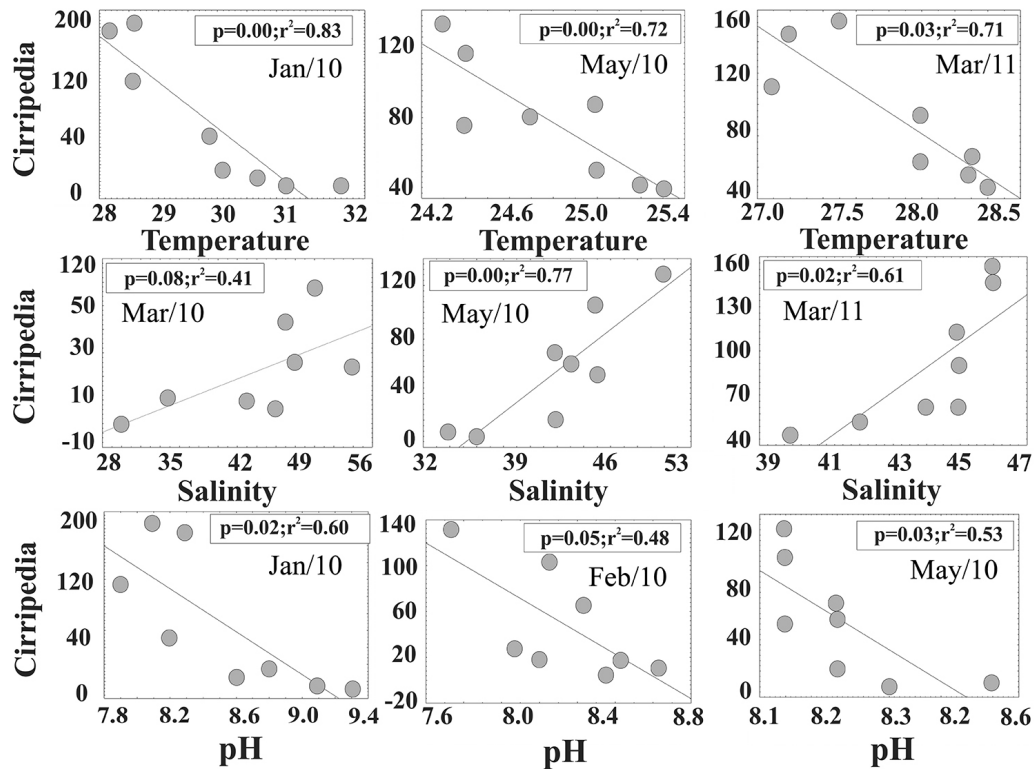


Figure 3. Relationship between temperature, salinity, and pH and their effect on the abundance of Cirripedia larvae over the months.

Table 1. Spatial variation correlations between abiotic data (Temperature, Salinity, and pH) and Cirripedia larvae over the sampled months.

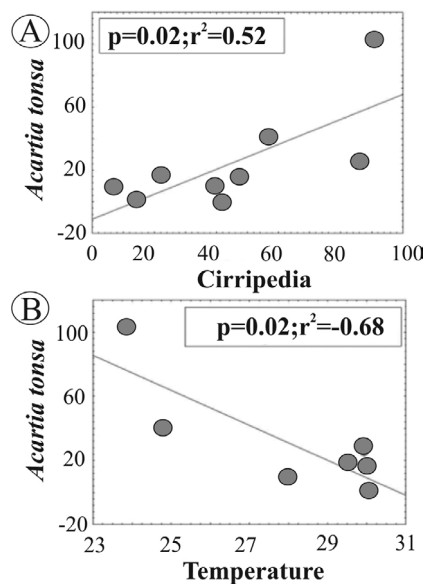
Months	Temperature	Salinity	pH
Jan/2010	$R^2 = -0.83$; $p = 0.00$	$R^2 = 0.19$; $p = 0.26$	$R^2 = -0.60$; $p = 0.02$
Feb/2010	$R^2 = -0.14$; $p = 0.36$	$R^2 = -0.55$; $p = 0.03$	$R^2 = -0.48$; $p = 0.05$
Mar/2010	$R^2 = -0.12$; $p = 0.39$	$R^2 = 0.41$; $p = 0.08$	$R^2 = -0.08$; $p = 0.48$
May/2010	$R^2 = -0.72$; $p = 0.00$	$R^2 = 0.77$; $p = 0.03$	$R^2 = 0.53$; $p = 0.03$
Oct/2010	$R^2 = 0.10$; $p = 0.42$	$R^2 = 0.61$; $p = 0.02$	$R^2 = 0.07$; $p = 0.51$
Nov/2010	-	$R^2 = 0.01$; $p = 0.76$	$R^2 = 0.07$; $p = 0.49$
Dec/2010	$R^2 = 0.10$; $p = 0.42$	$R^2 = 0.00$; $p = 0.82$	$R^2 = 0.02$; $p = 0.42$
Jan/2011	-	$R^2 = 0.16$; $p = 0.31$	$R^2 = 0.05$; $p = 0.57$
Feb/2011	-	$R^2 = 0.02$; $p = 0.70$	$R^2 = 0.03$; $p = 0.66$
Mar/2011	$R^2 = -0.71$; $p = 0.03$	$R^2 = 0.11$; $p = 0.40$	$R^2 = 0.01$; $p = 0.78$

Table 2. Spatial variation correlations between abiotic data (Temperature, Salinity, and pH) and *Acartia tonsa* over the sampled months.

Months	Temperature	Salinity	pH
Jan/2010	$R^2 = -0.10$; $p = 0.43$	$R^2 = -0.02$; $p = 0.69$	$R^2 = -0.05$; $p = 0.58$
Feb/2010	$R^2 = 0.00$; $p = 0.90$	$R^2 = 0.06$; $p = 0.54$	$R^2 = 0.00$; $p = 0.86$
Mar/2010	$R^2 = 0.06$; $p = 0.54$	$R^2 = 0.04$; $p = 0.59$	$R^2 = 0.25$; $p = 0.19$
May/2010	$R^2 = -0.26$; $p = 0.19$	$R^2 = 0.21$; $p = 0.25$	$R^2 = -0.21$; $p = 0.24$
Oct/2010	$R^2 = 0.34$; $p = 0.12$	$R^2 = 0.20$; $p = 0.25$	$R^2 = 0.05$; $p = 0.58$
Nov/2010	-	$R^2 = 0.00$; $p = 0.89$	$R^2 = 0.00$; $p = 0.77$
Dec/2010	$R^2 = 0.01$; $p = 0.80$	$R^2 = 0.15$; $p = 0.34$	$R^2 = 0.00$; $p = 0.82$
Jan/2011	-	$R^2 = 0.12$; $p = 0.39$	$R^2 = -0.01$; $p = 0.74$
Feb/2011	-	$R^2 = 0.21$; $p = 0.24$	$R^2 = 0.06$; $p = 0.53$
Mar/2011	$R^2 = 0.39$; $p = .009$	$R^2 = 0.13$; $p = 0.36$	$R^2 = 0.06$; $p = 0.54$

Table 3. Correlations of temporal variations between abiotic data, Cirripedia and *Acartia tonsa*.

Abiotic data	Cirripedia	<i>Acartia tonsa</i>
Temperature	$R^2 = -0.22$; $p = 0.28$	$R^2 = -0.68$; $p = 0.02$
Salinity	$R^2 = 0.04$; $p = 0.54$	$R^2 = -0.04$; $p = 0.56$
pH	$R^2 = -0.08$; $p = 0.40$	$R^2 = -0.08$; $p = 0.40$

**Figure 4.** Temporal correlations between larvae of Cirripedia and *Acartia tonsa* (A) and between *Acartia tonsa* and temperature (B).

The Araruama lagoon had a low species richness: the meroplankton was represented by six groups and the holoplankton by 16 taxa. The zooplankton community showed a high relative abundance and frequency of both Cirripedia larvae (48.49% relative abundance and 100% frequency of occurrence) and *Acartia tonsa* (18.78% relative abundance or 89% frequency of occurrence). The meroplankton was represented by Ascidiacea, Cirripedia, Decapoda, Bivalvia, and Polychaete larvae. The holoplankton consisted of *Creseis acicula*, *Limacina inflata*, Subclass Ostracoda, *Paracalanus quasimodo*, *Parvocalanus crassirostris*, *Acrocalanus monachus*, *A. tonsa*, *A. lilljeborgii*, *Temora turbinata*, *T. stylifera*, *Oithona hebes*, *O. simplex*, *Oncaea media*, *On. venusta*, and a single specimen of Monstrilloidea (all Copepoda) (Tab. 4).

Parvocalanus crassirostris was found in seasons 1 to 7 with minimum salinity of 35‰ and maximum of 55‰, while *T. turbinata* was found only in March 2010 with salinity of 54 ‰. *Monstrilla bahiana* was found in station 4 in January 2011 at salinity of 46‰. The density of zooplankton varied from 3 ind. m⁻³ (station 8, March 2010) to 912 ind. m⁻³ (station 4, October 2010) with an average of 121 ± 148 ind. m⁻³ (Fig. 5).

Table 4. Relative abundance, frequency of occurrence, mean and standard deviation of all groups identified during the study period.

Individuals	Abundance	Frequency	Mean	Deviation
Hydromedusa	0.10	6%	1.80	0.60
Ascidian larvae	1.79	19%	11.80	10.32
Cirripedia larvae	48.49	100%	64.49	71.52
Decapoda larvae	0.16	16%	1.31	0.52
Bivalvia larvae	0.01	1%	1.00	0.11
Mytilidae larvae	0.02	3%	1.00	0.16
Polychaeta larvae	0.04	4%	1.33	0.27
<i>Creseis acicula</i>	0.01	1%	1.00	0.11
<i>Limacina inflata</i>	0.02	3%	1.00	0.16
Ostracoda	0.02	3%	1.00	0.16
<i>Paracalanus quasimodo</i>	0.01	1%	1.00	0.11
<i>Parvocalanus crassirostris</i>	1.90	44%	5.17	5.94
<i>Acrocalanus monachus</i>	0.05	3%	2.50	0.46
<i>Acartia tonsa</i>	18.78	89%	27.27	67.86
<i>Acartia lilljeborgii</i>	0.01	1%	1.00	0.11
<i>Temora turbinata</i>	0.01	1%	1.00	0.11
<i>Temora stylifera</i>	0.01	1%	1.00	0.11
<i>Oithona oswaldocruzi</i>	0.26	15%	2.17	1.09
<i>Oithona simplex</i>	0.02	3%	1.00	0.16
<i>Oncaea media</i>	0.02	3%	1.00	0.16
<i>Oncaea venusta</i>	0.01	1%	1.00	0.11
<i>Monstrilla bahiana</i>	0.01	1%	1.00	0.11
Copepodite	26.58	93%	40.32	66.23
Nauplius	0.16	11%	1.67	0.75

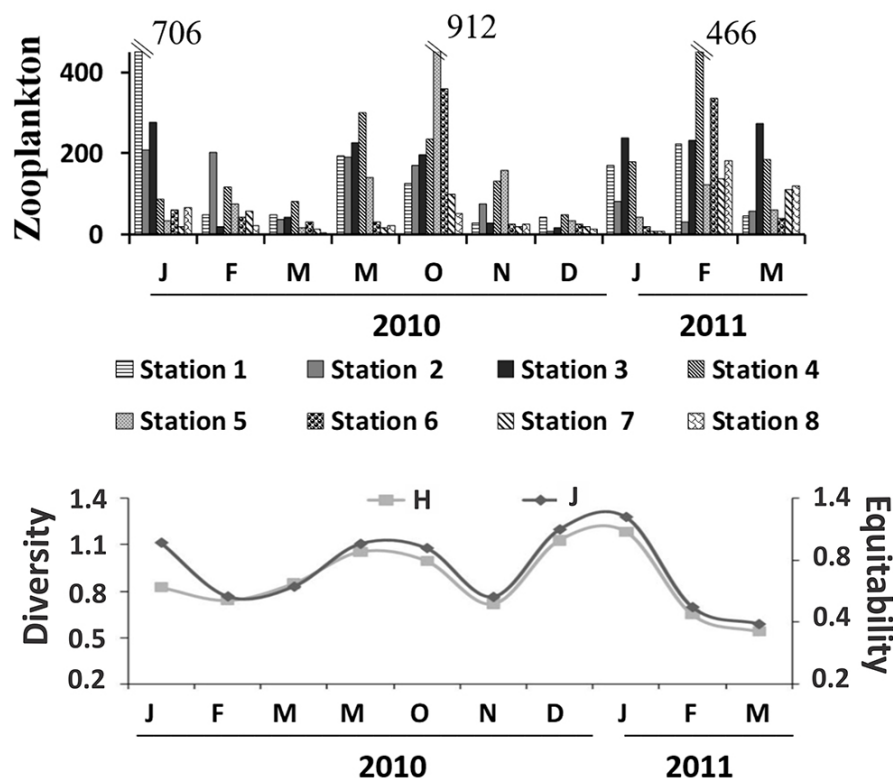


Figure 5. Variation of zooplankton density in each collection station, variations in the index of Shannon-Weaver which measures the Diversity (H) and the Pielou's uniformity which measures the Equitability (J) over the sampled months.

There is clear spatial variation (because the density varies considerably across the collection stations) in the density of zooplankton in the Araruama lagoon. In relation to the indices of community structure, richness was low and there was a high taxonomic similarity due to the dominance of Cirripedia and *A. tonsa* and the low contribution of other species. Equitability ranged from $E_H = 0.39$ in March 2011 to $E_H = 0.73$ in January 2011, with an overall average of $E_H = 0.57 \pm 0.11$. Shannon diversity varied from $0.54 \text{ bits.ind}^{-1}$ in March 2011 to $1.18 \text{ bits.ind}^{-1}$, with an overall mean of $0.86 \pm 0.21 \text{ bits.ind}^{-1}$ (Fig. 5).

DISCUSSION

A low number of taxa was found during the study (Copepod, Pteropods [Order Thecosomata], Cirripedia larvae, Decapoda, and Polychaeta), when compared to past data from regions near the Araruama lagoon (Arraial do Cabo) (Rosa *et al.*, 2016b). A previous study also found only three copepod genera (Coutinho *et al.*, 1999). This may be associated with the high salinity of Araruama Lagoon (Rosa *et al.*, 2016a).

Indeed, Wooldridge and Deyzel (2009) demonstrated that salinity and temperature are two of the main factors determining the spatio-temporal distribution of zooplankton. On a global scale, Buskey *et al.* (1998) claimed that hypersaline estuaries with salinity between 41‰ and 50‰ are usually characterized by low richness and low average abundance of zooplankton.

Over the study period, peaks in zooplankton abundance occurred during January 2010, October 2010, and February 2011, which were the months with the lowest salinities. These peaks may be related to peaks in phytoplankton abundance because, as mentioned by Souza *et al.* (2003), phytoplankton density is highest in the Araruama lagoon when the salinity is at its lowest. Studies with zooplankton have shown that when phytoplankton peaks are accentuated, the greater food availability consequently leads to an increase in the density of zooplankton (Fernandes *et al.*, 2012; Highfield and Eloire, 2010; Rosa *et al.*, 2016b).

The copepod assemblage is usually more abundant than the meroplanktonic larvae, but in the present study the Cirripedia larvae were almost always more abundant. Fernandes *et al.* (2012) showed that the

Cabo Frio region has a high percentage of benthic invertebrates that have larval stages. However, the dominance of Cirripedia larvae in almost every study period was unexpected since the copepod assemblage is usually dominant along the coast of Brazil (Dias and Bonecker, 2008). The high relative abundance and frequency of occurrence of the copepod *A. tonsa* is a strong indicator of eutrophication (Coelho-Botelho *et al.*, 1999). The constant temporal occurrence of this species in the lagoon suggests that *A. tonsa* is adapted to its conditions and can tolerate the high salinity and eutrophication. This species was also found in the Mar Menor in the Mediterranean by Gilabert (2001) and in the Madre Lagoon in South Texas, where high salinity adversely affected its populations (Buskey *et al.*, 1998).

The species *P. crassirostris* and *O. hebes* are characteristic of Brazilian estuaries (Björnberg, 1981). *Parvocalanus crassirostris* is a species that has a great tolerance to high salinity and temperatures (Mageed, 2006) and is found in several parts of the world (Montú, 1980; Björnberg, 1981; Mageed, 2006; Alajmi *et al.*, 2015) but is limited to the coastal waters of tropical and subtropical regions. It is one of the most common copepods in the coastal and estuarine waters of Brazil (Montú, 1980).

An individual of copepod from the order Monstrilloida (*M. bahiana*) was found in the lagoon in summer at a salinity of 46‰. Although representatives of Monstrilloida are widely distributed, they are poorly known compared to other planktonic copepods. This is mainly due to the rarity of individuals caught in trawls as they are only planktonic as adults (Dias and Bonecker, 2007). Their life cycle is different from other copepods because the larvae parasitize a considerable variety of benthic macroinvertebrates, such as polychaetes, mollusks, and other invertebrates (Grygier and Ohtsuka, 2008). Adults are planktonic and incapable of feeding. They live for a short period and only to reproduce (Suárez-Morales and Palomares-García, 1995). This is the first record of this order in the region of Cabo Frio (Valentin *et al.*, 1987; Monteiro-Ribas and Mureb, 1991; Rosa *et al.*, 2016b; Rosa and Monteiro-Ribas, 2012; Rosa *et al.*, 2019).

The copepod *T. turbinata* was recorded in low densities in the Araruama lagoon. This species has been cited as one of the most dominant species in estuarine environments and has an extensive distribution along

the Brazilian coast (Santos *et al.*, 2009; Rosa *et al.*, 2016b). *Temora turbinata* is considered as an invader off the Brazilian coast and was first mentioned in the 1980s in the northeast part of the country, and later recorded in the south. Consequently, this species seems to have adapted well to the environmental conditions of the Atlantic and has expanded its distribution along the Brazilian coast (Villac *et al.*, 2009). The holoplankton at Araruama lagoon was represented only by Copepoda, Pteropoda and Hydromedusa groups. This demonstrates that the holoplankton diversity of the lagoon is poorer than that of meroplankton, with the notable absence of groups commonly reported in other estuaries as well as in the Cabo Frio region, such as Cladocera, Ostracoda, Appendicularia and Chaetognatha (Santos *et al.*, 2009; Rosa *et al.*, 2016b; Araujo *et al.*, 2017). Species of Appendicularia and Ctenophora were previously found at high salinities in the Mar Menor, region of the Mediterranean Sea (Gilabert, 2001), revealing that they can survive in hypersaline environments. However, the species present in the Cabo Frio region could not withstand the conditions of the lagoon.

When exposed to the atypically higher temperatures of the Araruama lagoon the results demonstrate a lower release of the Cirripedia larvae, which may be related to the preference to release larvae at lower temperatures. According to Fernandes *et al.* (2012), meroplanktonic larvae may also have peaks in cold water periods and associated with phytoplankton peaks. In addition, the larvae also prefer an acidic pH which corroborates the work of Pansch *et al.* (2013), who found that the survival rate of Cirripedia larvae was greater at lower pH levels. In this study, six groups of larvae were found: ascidians, cirripedians, decapods, bivalves, Mytilidae, and polychaetes. Comparatively, on the island of Cabo Frio, which is located near the lagoon, nine groups of larvae were found including Ostreidae, Isognomonidae, and Bryozoa, which were absent in Araruama lagoon (Oliveira *et al.*, 2015). Cirripedia larvae were the most representative group in terms of abundance in the present work, but in Fernandes *et al.* (2012) and Oliveira *et al.* (2015), the larvae of Mytilidae were found to be just as abundant as Cirripedia. In contrast, we found Mytilidae larvae to be almost absent, especially at the innermost stations.

CONCLUSION

After analyzing the data, it is evident that the spatial variation of the Cirripedia larvae is directly influenced by abiotic factors. The Cirripedia have higher salinity, lower temperature and pH as the main factors that influence their preference for environments and an inversely proportional relationship with the opposite conditions (low salinity, high temperature and pH). The distribution of cirripedes in Araruama lagoon seems to follow the same pattern regarding the gradient of these abiotic factors along the lagoon. The low quantity of taxa presented in the lagoon when compared to geographically close environments, such as Cabo Frio, seems to be related to the ability of the species to acclimate to the extreme conditions. Few species can adapt to the conditions found in the lagoon, particularly those of salinity and temperature. New studies are needed to precisely define if the abiotic factors work in isolation in the zooplankton assembly or if they are correlated factors, as well as their specific impact on the biomass and trophic dynamics of the environment.

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