

Sampling planktonic cnidarians with paired nets: Implications of mesh size on community structure and abundance

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ARTICLE INFO

Keywords:

Hydromedusae
Siphonophores
Mesh selectivity
Zooplankton
Amazon River Plume

ABSTRACT

Mesh selectivity is an important factor to be considered when sampling zooplankton. Selectivity may differ according to the environment and taxon, but has not been evaluated for cnidarians in neritic and/or oceanic ecosystems. In this study, efficiency of plankton nets with two different mesh sizes (120 and 300 μm) were compared to assess the cnidarian assemblage from neritic and oceanic habitats in the equatorial Atlantic Ocean. No statistical differences between meshes were observed in species richness, total and most dominant taxa abundance, or taxonomic and size structure of the community in both oceanic and neritic habitats. Only for the small (0.5–2 mm) neritic *Persa incolorata* did the meshes differ, with higher abundances in the finer one. Despite small particularities, our results show that the 120 and 300 μm meshes produce similar results.

1. Introduction

Marine planktonic animals have a wide variability in size and swimming ability, constituting a quite heterogeneous community which, together with the typical patch distribution, makes it difficult to establish standard sampling protocols in ecological studies and quantitative assessments (Omori and Hamner, 1982; Harris et al., 2000). The accuracy of a particular sampling instrument typically is related to species size, morphology, flexibility, behavior and distribution, and environmental characteristics (Vannucci, 1968; De Bernardi, 1984). Among the many factors that may influence zooplankton sampling, mesh size selectivity is one of the most important (Skjodal et al., 2013). While a small mesh may filter high rates of unwished particles, obstructing water passage and rapidly clogging the mesh, a coarse one may lose smaller species and early life stages (Riccardi, 2010; Vannucci, 1968).

Mesh selectivity and catch efficiency is a controversial subject. For copepods, where mesh size effect is relatively well understood, 200 μm meshes may lose a significant proportion of small species, copepodites and nauplii abundance (up to 90%) and biomass (up to 50%; Hopcroft et al., 1998; Hwang et al., 2007; Di Mauro et al., 2009; Favareto et al., 2009; Riccardi, 2010; Wu et al., 2011). However, the reported underestimation of larger species and individuals by smaller meshes due to the bow wave effect and mesh obstruction (Vannucci, 1968; Hopcroft et al., 2001; Favareto et al., 2009) is not ubiquitous (Antacl et al.,

2010; Miloslavić et al., 2014).

The effects of different mesh sizes on other invertebrate zooplanktonic taxa are poorly known. Few particular information is available considering appendicularians, chaetognaths, mollusks, polychaetes, cnidarians and non-copepod crustaceans (Almeida Prado, 1962; Vidjak, 1998; Di Mauro et al., 2009; Riccardi, 2010; Skjodal et al., 2013; Miloslavić et al., 2014; Nogueira Júnior et al., 2015). Despite most of this literature showing that specific groups and/or sizes are better estimated by particular mesh sizes, the lack of detailed information, such as seasonality and habitat variability, hinders the development of standard sampling protocols. Given the requirement of precise methods to estimate zooplankton abundance, detailed information of taxa-specific mesh size effects considering habitat heterogeneity is a matter of great importance.

Planktonic cnidarians have aroused recent interest in the scientific community due to their high feeding rates and unexpected population blooms (e.g. Purcell et al., 2007). For a complete understanding of these processes, precise quantitative methods need to be established. Although the choice of the mesh size will depend on particular targets in each study, the 500 μm mesh, frequently used in planktonic cnidarian sampling (e.g. Gili et al., 1991; Loman-Ramos et al., 2007; Segura-Puertas et al., 2010), was found to largely underestimate hydrozoan estuarine assemblages (Nogueira Júnior et al., 2015). Since the estuarine cnidarian community usually is dominated by small-sized hydromedusae (Xu and Huang, 1983; Nogueira Júnior et al., 2015), the

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<https://doi.org/10.1016/j.ecss.2019.02.027>

Received 6 November 2018; Received in revised form 15 January 2019; Accepted 9 February 2019

Available online 21 February 2019

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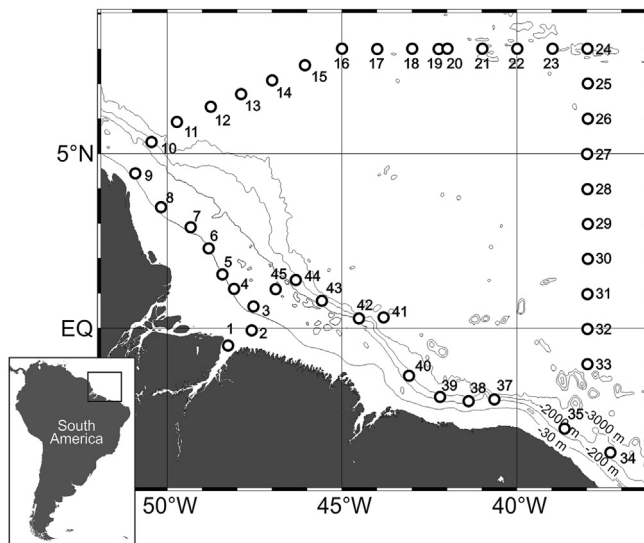


Fig. 1. Map of the North Brazilian continental shelf and adjacent equatorial Atlantic Ocean, showing the sampled stations.

absence of studies testing the effect of mesh sizes in neritic and oceanic waters precludes the possibility to expand the results to other taxonomic and size composition ecosystems.

In aiming to improve the understanding of this subject, in this study we evaluated the efficiency of plankton nets with two different mesh sizes (120 and 300 μm) to assess the cnidarian assemblage from neritic and oceanic habitats in the equatorial Atlantic. This is based on the hypothesis that the meshes will provide different results regarding diversity, abundance and community structure.

2. Material and methods

2.1. Study area and sampling

Samples were obtained between October 9th and November 1st, 2012 during the oceanographic cruise Camadas Finas III, aboard the research vessel NHO. Cruzeiro do Sul - H38 (DHN/Brazilian Navy) along the North Brazilian continental shelf and adjacent equatorial Atlantic Ocean (Fig. 1). The area is a heterogenic environment, affected by the large freshwater discharge of the Amazon River, strong currents, eddies, wind fields and high tidal variation (Molleri et al., 2010; Nitttrouer and DeMaster, 1996). Zooplankton samples were obtained at 44 stations in oblique hauls, using nets with 120 and 300 μm mesh and 0.3 and 0.6 m opening respectively (except for station 39, where only the 300 μm mesh sample was available). The 300 μm net was hauled immediately after the 120 μm one. Stations were sampled from the near bottom to the surface on the continental shelf, and from 200 m to the surface in the open ocean. These nets were towed at approximately 2 knots, at various times of day and night. Due to differences in depth, duration of hauls was variable, ranging from 6 to 35 min. Both nets were fitted with a flowmeter (Hydro-Bios) to estimate the volume filtered during each trawl. Volume sampled averaged (\pm SD) 65.1 ± 42.7 and $196.2 \pm 172.9 \text{ m}^3$ in the 120 and 300 μm mesh respectively. Samples were fixed with 4% formaldehyde buffered with sodium tetraborate (0.5 g l^{-1}).

2.2. Analysis of samples

Whole samples of both meshes ($N = 87$) were analyzed under a stereomicroscope and specimens were identified (Boltovskoy, 1999) and counted. Abundances were standardized as number of individuals 100 m^{-3} for medusae and number of colonies 100 m^{-3} for

siphonophores. For calyphorans, the number of anterior nectophores was used for estimating the polygastric stage abundance, and eudoxid bracts for the eudoxid stage abundance (e.g., Hosia and Båmstedt, 2007). Physonects and the calyphoran *Hippopodius hippopus* were not considered in the analysis due to the well-known difficulties in accurate quantifying them from net samples. In each sample, up to 30 individuals per species were measured, considering the medusae bell diameter and siphonophore largest width.

2.3. Data analysis

Stations were grouped into neritic and oceanic habitats based on their location, considering the shelf break at the isobath of 100 m. Species richness, total abundance, and abundance of individual dominant taxa estimated with both meshes in neritic and oceanic habitats were compared using Bi-Factorial ANOVA (Zar, 2010). PERMANOVA was used to test whether the taxonomic and size structure of the cnidarian community changed according to mesh size, type of habitat (neritic or oceanic) and possible interactions between these factors. PERMANOVA were based on a Bray–Curtis similarity matrix after data transformation ($\log(x+1)$; Anderson, 2001). P values < 0.05 were considered significant in both analyses. The collection efficiency of the 300 μm mesh (CE_{300} , %) in relation to the 120 μm mesh was calculated as follows: $\text{CE}_{300} = (\text{Ab}_{300} * 100) / \text{Ab}_{120}$; where Ab_{300} and Ab_{120} are abundance estimated by 300 and 120 μm meshes respectively. Bi-Factorial ANOVA was performed using Statistica 10. PERMANOVA analysis were performed using Primer v.6 + PERMANOVA.

3. Results and discussion

A total of 80 taxa were sampled (37 siphonophores, 41 hydromedusae and 2 scyphomedusae). The 300 μm mesh sampled more taxa in the neritic stations (33 vs 29) and the 120 μm sampled more in the oceanic ones (61 vs 47), but differences were not significant (see below). While 19 taxa were found exclusively in the 120 μm mesh, 12 were exclusive in the 300 one. All these taxa were rare, with very few individuals. Their absence in one of the meshes probably was random, as a consequence of increasing the sampling effort. However, some of the exclusive species found in the 120 μm mesh are small ($\sim 1 \text{ mm}$) hydromedusae such as *Cytaeis* sp. and *Corynidae* sp. which may have been missed by the 300 μm mesh. Damaged unidentifiable specimens represented 1.3% and 2% of 120 and 300 μm mesh respectively.

Significant differences ($p < 0.05$) were not observed in species richness and total abundance obtained with 120 and 300 μm meshes (Fig. 2; Table 1), the same pattern was obtained when considering the size and taxonomic structure in the PERMANOVA or the interaction in the neritic and oceanic habitats in all cases (Fig. 2; Table 1). In all analysis differences were observed between the habitats (Table 1). Although average CE_{300} was slightly lower in the neritic stations, the index presented a high variability, ranging from 9.9 to 619.6% in the oceanic stations and from 24.3 to 289.4% over the continental shelf (Fig. 2e). No clear pattern in the CE_{300} was observed when considering its distribution by size classes, with average values always around 100% and high variability (Fig. 2f).

Dominant taxa were similar in both meshes, only differing in rank order and proportion (Table 1). The neritic habitat was dominated by *Liriope tetraphylla*, *Persa incolorata*, *Enneagonum hyalinum*, *Muggiaea kochii* and *Diphyes dispar*. Oceanic stations were dominated by *Diphyes bojani*, *Aglaura hemistoma*, *Chelophyes appendiculata*, *Eudoxoides mitra* and *Bassia bassensis* (Table 1). This condition was shown both in the size and taxonomic structure, which differ significantly (PERMANOVA; $p < 0.05$) between the neritic and oceanic habitats, but not between the meshes or the interaction of the factors (Table 1).

Mesh pore aperture is often one of the main factors to be considered in the selection of the sampling procedure, and studies comparing paired nets such as the presented here provide substantial support to

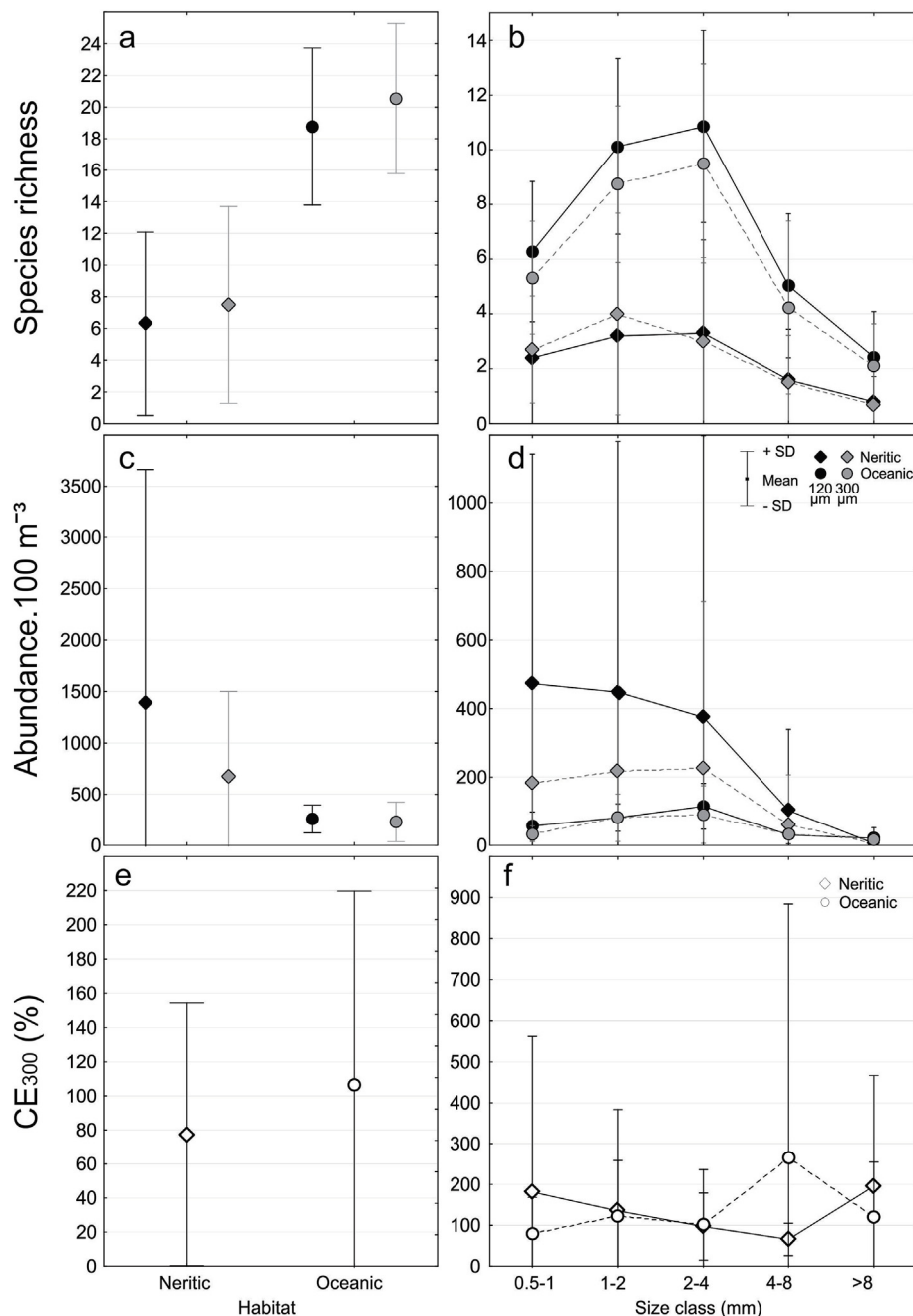


Fig. 2. Average species richness (a; b), abundance (c; d) and collection efficiency of 300 µm mesh (e; f) of total (a; c; e) and size-based (b; d; f) planktonic cnidarian community in neritic and oceanic habitats in the equatorial Atlantic Ocean.

help scientists choose the right equipment (Antacili et al., 2010). Overall, the results of this study show that the two mesh sizes tested produced similar results both at neritic and oceanic habitats. Yet, the use of multiple net sizes in zooplankton sampling may enhance the understanding of the community structure and the prediction power of ecological models by demonstrating its whole size spectrum was correctly considered, particularly for ecosystems where such variability has not been tested (Antacili et al., 2010; Miloslavić et al., 2014; Nogueira Júnior et al., 2015), emphasizing the importance of methodological tests such as the present study.

Even though general ecological results were similar, anomalies were observed in specific taxa such as the small trachymedusa *Persa incollarata* (0.5–2 mm), in which abundance differed between the two meshes, habitats and the interaction of the factors (Table 1). It occurred almost exclusively in a few neritic stations, where average abundance

estimated by the 120 µm mesh was nearly six times that estimated by the 300 µm mesh (Fig. 3b). Most of the sampled individuals were juveniles, smaller than 1 mm bell diameter, and probably lost by 300 µm mesh. Similar effects probably occurred with less-abundant small-species such as *Obelia* spp. (< 1 mm). In general, a particular mesh would efficiently sample organisms at least 1/3 larger than its pore apertures (Bernhard et al., 1973; Nichols and Thompson, 1991). Thus, the 120 and 300 µm meshes would work well for specimens > 160 and > 400 µm respectively and filtering capacities of 300 µm mesh would be sufficient even for the smaller hydromedusae found (approximately 0.5 mm). However, the absence of rigid structures such as appendices, antennae and spines and gelatinous fragile body of small hydromedusae may facilitate deformation of mesoglea and the passage of larger specimens through the mesh pores.

Although the other dominant species and total abundance in neritic

Table 1

Dominant planktonic cnidarian taxa relative abundance and size (mean and range) in neritic and oceanic stations sampled with two mesh sizes. Summary of Bi-factorial ANOVA testing differences in individual taxa abundance, species richness and total abundance, and PERMANOVA (Pseudo F) testing differences in size-based and taxonomic structure of planktonic cnidarian community considering mesh size and habitat as factors. E = Calicophoran eudoxid stage. P = Calicophoran poligastric stage. Bold = significant ($p < 0.05$) differences.

Habitat/Taxa/Indicator	Relative abundance		Mean species size and interval (mm)		Mesh		Habitat		Mes x Hab	
	120 μm	300 μm	120 μm	300 μm	F	p	F	p	F	p
Neritic										
<i>Liriope tetraphylla</i>	31.07%	32.53%	2.6 (0.5–10)	2.2 (0.5–9)	2.46	0.12	20.0	< 0.01	2.09	0.15
<i>Persa incolorata</i>	27.84%	9.31%	1.17 (0.5–2)	1.17 (0.8–2)	4.42	0.03	8.51	< 0.01	4.40	0.03
<i>Enneagonum hyalinum</i>	17.67%	25.52%	E = 2.8 (0.8–9) P = 3.5 (2–10)	E = 3.2 (2–8) P = 2.9 (2–10)	0.25	0.61	8.34	< 0.01	0.25	0.61
<i>Muggiaea kochii</i>	9.59%	8.88%	E = 0.8 (0.6–1) P = 2.5 (0.8–4)	E = 0.8 (0.8–1.2) P = 2.5 (1–4)	1.11	0.29	7.79	< 0.01	1.12	0.29
<i>Diphyes dispar</i>	6.63%	7.9%	E = 4 (1.5–5) P = 4.8 (2–15)	E = 3.4 (1.5–6) P = 4.9 (2–17)	0.68	0.41	9.55	< 0.01	0.73	0.39
Others (n = 31 spp.)	7.2%	15.86%	2.3 (0.5–9)	2 (0.5–8.8)						
Oceanic										
<i>Diphyes bojani</i>	17.58%	24.31%	E = 2.4 (0.8–5) P = 5.4 (1–12)	E = 2.1 (0.7–5) P = 5 (0.9–13)	0.42	0.51	2.39	0.12	0.01	0.90
<i>Aglaura hemistoma</i>	16.02%	10.92%	1.4 (0.5–3)	1.4 (0.5–3)	0.40	0.52	6.19	0.01	1.27	0.26
<i>Chelophyes appendiculata</i>	14.01%	17.08%	E = 1.2 (0.8–2) P = 7 (2–12)	E = 1.1 (0.7–2) P = 6.9 (1–14)	0.73	0.39	10.4	< 0.01	0.28	0.59
<i>Eudoxoides mitra</i>	7.64%	13.92%	E = 2.8 (0.8–5) P = 5.8 (1–13)	E = 2.8 (2–5.5) P = 6.5 (1–13)	0.04	0.84	13.2	< 0.01	0.06	0.79
<i>Bassia bassensis</i>	5.25%	8.61%	E = 2.5 (0.5–5) P = 2.4 (0.8–5)	E = 2 (0.5–5) P = 2 (0.5–4)	0.03	0.85	23.6	< 0.01	0.04	0.83
Others (n = 64 spp.)	39.5%	25.16%	2.7 (0.5–13)	2.7 (0.6–15)						
Species richness					1.29	0.26	95.9	< 0.01	0.05	0.83
Total abundance					3.24	0.08	14.6	< 0.01	2.76	0.10
Size structure					2.41	0.07	11	< 0.01	0.79	0.5
Taxonomic structure					0.8	0.66	42.4	< 0.01	0.81	0.64

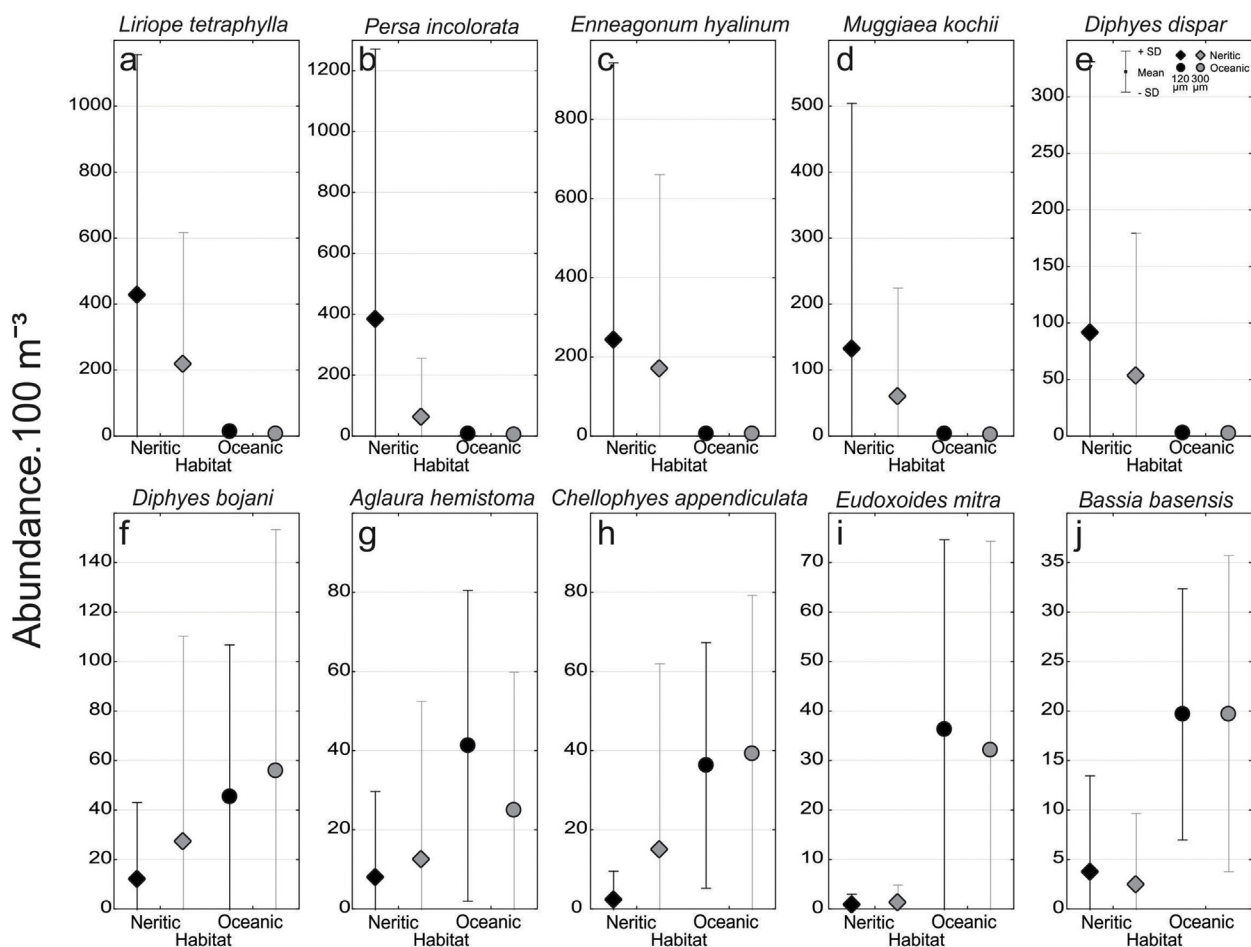


Fig. 3. Average abundance (ind. 100 m⁻³) of dominant planktonic cnidarians sampled with 120 and 300 μm meshes in neritic and oceanic habitats in the equatorial Atlantic Ocean.

stations did not significantly differ between the two meshes, mean values were quite higher at the 120 µm one (Figs. 2c–d, 3a–e). The absence of significant results may in part be a consequence of the high variance in the data, a common feature of biological communities in general and of marine zooplankton in particular in part due to the patchy distribution common of zooplanktonic organisms in many spatial scales (Steele, 1978; Omori and Hamner, 1982; Messié and Chavez, 2017). This may be particularly true in the present study, with a high variability of habitats over the large area sampled, with many oceanographic processes occurring simultaneously (Lentz and Limeburner, 1995; Nittrouer and DeMaster, 1996; Moller et al., 2010), where even adjacent stations presented high variability as can be seen by the high standard deviations.

Despite small anomalies (mostly *P. incolorata*), our results show that the 120 and 300 µm meshes produce similar results sampling planktonic cnidarians in neritic and oceanic habitats. Studies use meshes usually ranging from 150 to 500 µm for oceanic and neritic environments (Pagès and Gili, 1992; Lo and Biggs, 1996; Thibault-Botha et al., 2004; Nogueira Júnior et al., 2014), although coarser and finer meshes also may be used (e.g. Pagès and Kurbjeweit, 1994; Morita et al., 2017). Since these studies did not evaluated mesh efficiency, this divergence in the sampling procedures hinders the possibility of direct comparisons considering abundance, species richness and assemblage structure. Thus, future ecological studies with the group in oceanic habitats should also consider mesh selectivity aiming to produce better estimates, more comparable results, and for eventual calibrations due to possible regional particularities.

Although the frequency of ecological studies involving planktonic cnidarians has increased significantly in the last decades, the discussion of the most accurate way to sample them is still in the early stages. Much is to be done to define the best way of sampling planktonic cnidarian with net hauls. While the actual evaluations indicate 200 µm mesh is adequate for estuarine environments (Nogueira Júnior et al., 2015) and both 120 and 300 µm are adequate to neritic and oceanic habitats (present study), these and coarser meshes need be tested elsewhere in the world to endorse our results. Other aspects such as tow duration, speed and shape, net opening and size, and bow wave effect also were never evaluated and may influence the samplings. In addition to nets, new technologies such as video recorders (Luo et al., 2014) and active acoustics (Båmstedt et al., 2003) are promising zooplankton sampling techniques for the future and combination of these approaches and others to come are likely to broaden our perspectives on the functioning of ecological systems.

Funding

This work was supported by the Brazilian National Institute of Science and Technology for Tropical Marine Environments–INCT AmbTropic (CNPq/FAPESB grants 565054/2010-4 and 8936/2011), Brazilian Research Network on Global Climate Change – Rede CLIMA (FINEP grant 01.13.0353-00) and European Integrated CARBOCHANGE (FP7 264879). EGT was supported by CNPq (grant 140897/2017-8).

Acknowledgements

We thank the support of Brazilian National Institute of Science and Technology for Tropical Marine Environments, Brazilian Research Network on Global Climate Change, European Integrated CARBOCHANGE and Conselho Nacional de Desenvolvimento Científico e Tecnológico. And also to all the boarded scientific team of Camadas Finas III and Amadeus II research project.

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