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Time variability of prevalent mesozooplankton at Montevideo coast, Río de la Plata, and its relationship with physico-chemical drivers

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Abstract. Rio de la Plata estuary (RPE) is the widest in the world but temporal variability patterns of its mesozooplankton assemblages are unknown. Surveys were conducted at the coast of Montevideo from January to November 2003 in order to study the abundance of most prevalent mesozooplankton populations, and their relationship with the changes in salinity, temperature, chlorophyll-a (Chl-a), and phaeopigments (PHA). Mean temperature and salinity at the study site were 19.25 ± 4.8ºC and 9.25 ± 6.18 respectively. Chl-a ranged from 0.41 to 66.39 mg.m^{-3} (median = 3.63 mg.m^{-3}); the ratio of Chl-a to PHA (RCP) ranged from 0.27 to 8.13 (median = 0.53). Temperature, Chl-a, RCP varied much as seasons changed, but not salinity. Mesozooplankton abundance ranged from 70 to 15381 ind/m^3, median = 4805 ind/m^3. Copepod (e.g. *Acartia tonsa*, *Paracalanus parvus*), larval stages of barnacles *Balanus* sp. and bivalves were among the most prevalent populations of the mesozooplankton assemblage. Species evenness of copepod the community remained unaffected by variability of the physico-chemical variables studied. Copepod *Acartia tonsa*, *Paracalanus parvus* abundances did not follow any clear seasonality. At the onset of spring, abundance of *Acartia tonsa* increased with availability of fresh food in the study site, which could be an indication of grazing, growth and production rates limited by food quality. *Paracalanus parvus* abundance remained unaffected by the fluctuations of the physico-chemical variables analysed. Abundance of *Balanus* sp. rose high as the water temperature increased in RPE, especially in spring-summer. Overall, the results suggest that the most prevalent mesozooplankton of RPE are possibly resilient to environmental fluctuations.

Keywords: Salinity; Temperature; Copepods; Estuaries; Uruguay

Resumen. Variabilidad temporal del mesozooplancton de la costa de Montevideo, Río de la Plata, y su relación con forzantes fisicoquímicos. El estuario del Río de la Plata (RPE) es uno de los más amplios del mundo, pero la variabilidad temporal del mesozooplancton es mayormente desconocida. Se realizaron colectas en la costa de Montevideo desde enero a noviembre de 2003 para estudiar la abundancia de las poblaciones dominantes en el mesozooplancton y su relación con los cambios en salinidad, temperatura, clorofila-a (Chl-a) y feopigmentos (PHA). La temperatura media y la salinidad en el sitio de estudio fueron 19,25 ± 4,8ºC y 9,25 ± 6,18, respectivamente. La Chl - a varió de 0,41 a 66,39 mg.m $^{-3}$ (mediana = 3,63 mg.m – ^{3}). El cociente Chl-a : PHA (RCP) varió entre 0,27 y 8,13 (mediana = 0,53). Temperatura, Chl-a y RCP fluctuaron en función de la estación del año, pero no la salinidad. La abundancia de mesozooplancton osciló entre 70 y 15381 ind / m^3, mediana = 4805 ind / m^3. Los copépodos (por ejemplo, *Acartia tonsa*, *Paracalanus parvus*), estadios larvales de balanos

Balanus sp. y bivalvos se encontraban entre las poblaciones dominantes del mesozooplancton. La uniformidad (*evenness*) de especies de la comunidad de copépodos no se vio afectada por las fluctuaciones de las variables físico-químicas consideradas. La abundancia de los copépodos *Acartia tonsa* y *Paracalanus parvus* no siguió una estacionalidad clara. En el inicio de la primavera, la abundancia de *Acartia tonsa* aumentó con la disponibilidad de alimento fresco en el sitio del estudio. Ello podría ser un indicio de tasas de pastoreo, crecimiento y producción limitadas por la calidad del alimento disponible. La abundancia de *Paracalanus parvus* no se vio afectada por la variabilidad temporal del ambiente físico-químico. La abundancia de *Balanus* sp. aumentó a medida que la temperatura del agua incrementó, especialmente durante primavera-verano. En general, los resultados sugieren que el mesozooplancton dominante en la zona analizada del RPE son resilientes a las fluctuaciones ambientales.

Palavras-chave: Salinidad; Temperatura; Copepodos; Estuarios; Uruguay

Introduction

Zooplankton couple the energy flow between benthic and pelagic trophic levels of many subtropical estuaries of the Southern Hemisphere (Griffin *et al.,* 2001; Froneman, 2004). Copepods are the world's most abundant metazoan that link protists to higher trophic levels such as mysids, crabs, and fish (Costa, 2009). High temperatures in spring-summer increase phytoplankton production in the sub tropical waters of Americas (Alpine & Cloern, 1992; Pennington & Chavez, 2000). Such environment supports high densities of mesozooplankton especially copepods (Costa, 2009). Some of these copepods (e.g. *Acartia tonsa*) are able to feed on detritus and degraded phytoplankton forms which at times are the dominant class of pigments in certain turbid subtropical estuaries of South America such as Rio de la Plata estuary (RPE) (Calliari *et al.,* 2004; Islam *et al.,* 2006).

RPE is the widest estuaries of the world that runs along the Uruguayan and Argentinean coasts (Figure 1) (Acha *et al.,* 2008). It has a drainage basin of nearly 3,170,000 km^2 spread across the territories of Bolivia, Paraguay, Brazil, Argentina and Uruguay. The RPE is a reproductive nursery and feeding ground for many commercially exploited fish species and macro-invertebrates (Viñas *et al.,* 2002; Marrari *et al.,* 2004). Despite that, attention has been patchy on plankton; to date, literature mostly targeted the spatial distribution of phytoplankton, their abundance and production rates (Gómez *et al.,* 2004; Calliari *et al.,* 2005, 2008, 2009; Carreto *et al.,* 2008). Mesozooplankton studies are few and focused on spatial distribution of community in particular of copepods along salinity gradients of RPE (Marrari *et al.,* 2004; Berasategui *et al.,* 2006). Derisio *et al.,* (2014) analysed the egg production of dominant copepod *Acartia tonsa* in the oligohaline region of the RPE. Calliari *et al.,* (2004) reported community structure, herbivory and egg production rates by *A. tonsa* at the coast of Montevideo during a short period during the summer - autumn transition. Another limitation to current knowledge is that most studies relied on short-term observations (Gomez *et al.,* 2004; Calliari *et al.,* 2005, 2009) or on average responses of pooled results obtained during an extended period (Berasategui *et al.,* 2006). Consequently, time variability and patterns at seasonal and longer time scales are lacking. An exception is the study by Viñas *et al.,* (2005) describing population dynamics and time variability of mysid *Neomysis americana* at Samborombòn Bay (southern RPE, Argentinean coast) during a year cycle. Time variability of most mesozooplankton of RPE and notably that of copepods is unknown despite its importance for understanding food web structure. Thus the aims are: i. to describe the time variability of most prevalent mesozooplankton populations in the brackish zone of RPE (essentially off the Montevideo); and ii. to explore the relationship between the seasonal fluctuations of abundance of most prevalent mesozooplankton and physico-chemical parameters, and iii. species evenness of copepod community and if that was affected by the physico-chemical variability of the study site.

Materials and methods

Sampling Design: Sampling was intended to explore the temporal variability of the mesozooplankton assemblages' of a coastal site within RPE during 2003 (Figure 1 and Table I). Except December, samples were collected monthly during the whole period, and weekly every third month. For resource limitation, in winter samples were obtained every second week. Samples were collected at a near shore

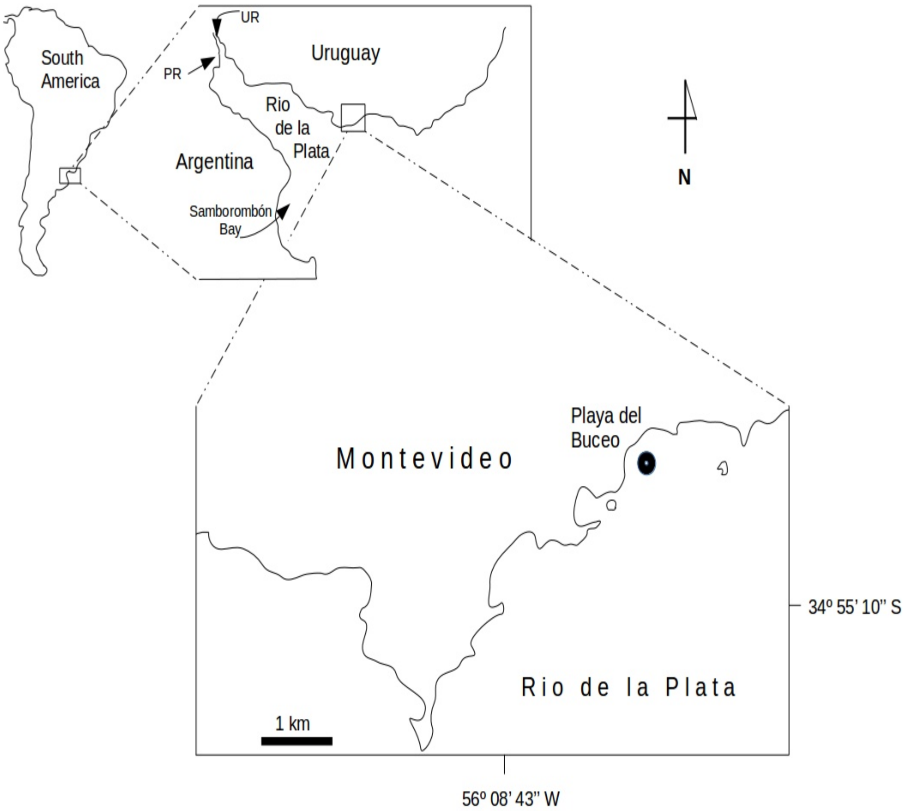

Figure 1: Map of the study area. Black circle at the small scale map indicates the location of the sampled station. Arrows indicate the outflow of Uruguay (UR) and Paraná (PR) rivers into the RPE

Table I Mesozooplankton assemblages (individuals / m^3) and their relative abundance (% of the total catch) off Montevideo (Rio de la Plata estuary of Uruguay) in 2003.

Species/Groups	Summer	% Sum	Autumn	% Aut	Winter	% Win	Spring	% Spr
Acartia tonsa	20263	35	25606	75	7097	95	20778	84
Euterpina acutifrons	14	0	0	0	0	0	0	0
Coryceus amazonicus	31	0	0	0	0	0	0	0
Oithona nana	0	0	62	0	3	0	0	0
Oithona sp.	0	0	23	0	0	0	6	0
Oncea sp.	54	0	3	0	6	0	16	0
Paracalanus parvus	637	1	1377	4	79	1	155	1
Calanoid 1	105	0	57	0	0	0	28	0
Calanoid 2	371	1	2609	8	8	0	323	1
Cyclopoid	4062	7	0	0	0	0	0	0
Cladocerans	6615	11	0	0	3	0	40	0
Balanus sp.	16720	29	3946	12	228	3	2563	10
Neomysis americana	45	0	42	0	0	0	289	1
Oikopleura dioica	17	0	0	0	0	0	0	0
Ostracoda	122	0	6	0	7	0	114	0
Bivalvia	5459	9	255	1	4	0	448	2
Polychaete	6	0	0	0	1	0	0	0
Hydromedusa	3000	5	20	0	0	0	0	0
Gastropoda	82	0	0	0	3	0	11	0
Chaetognatha	0	0	26	0	0	0	1	0
Amphipoda	0	0	0	0	0	0	15	0

site off Montevideo (Playa del Buceo, 1.5 – 2.0 m depth) just after sunset (Fig. 1). Briefly, sub-surface (~1.5 – 2.0 m depth) tows were made using a 35 cm mouth plankton net fitted with 180µm pore size mesh. Volume of water sampled was about 19 m^3 on each tow. Samples were preserved in a formalin solution 4% (final concentration). Mesozooplankton were identified and then counted under a microscope to the lowest possible taxonomic level according to the regional keys. Abundance of each species was expressed as individuals per cubic meter (ind/m^3).

Water temperature and salinity were measured at ~ 0.5 m depth using a multi-probe sensor (Horiba U 10, Japan). Subsurface (0.5 m depth) water samples were collected for Chl-a, and PHA analyses. 50 mL subsamples were filtered through GF/F filters, and pigments extracted in ethanol 96% in the dark at 0°C for 24 hours. The fluorescence of the resulting extract was read before and after acidification (HCl 1.2 N) in a Turner 111 Fluorometer calibrated against standard Chl-a extract. Chl-a, and PHA levels were expressed as $mg.m^{-3}$. Chl-a is a routinely used indicator of phytoplankton biomass thereby a proxy of food availability for herbivore grazers (Kiorboe, 1993). PHA is the degradation products of Chl-a, and is closely associated to turbidity in turbid estuaries (Irigoien *et al.*, 1995; Islam *et al.*, 2006). The ratio of Chl-a, and PHA (RCP= Chl-a / PHA). provides a rough indication of the relative amounts of living cells to detritus material, and the freshness of the phytoplankton food (Calliari *et al.*, 2004); dominance of fresh material (on a molar basis) is indicated by RCP > 1.

Data analysis: Fifteen taxonomic groups were found along the 11-months of sampling period (Table 1). Only few taxa were consistently present throughout 2003 (Table 1), these were the copepods *Acartia tonsa* and *Paracalanus parvus*, and barnacle *Balanus* sp. larval stages (nauplii and cyprid larvae together). Dataset consists of abundances of above mentioned species along with salinity, temperature, Chl-a, and RCP (i.e. Chl-a / PHA). Salinity, temperature, Chl-a, and RCP data were checked for normality by Shapiro tests. Temperature (W = 0.96, P-value = 0.44) salinity (W = 0.94, p-value = 0.1649) data were normally distributed but Chl-a (W = 0.49, P-value < 0.001) RCP (W = 0.58, p-value < 0.001) were not. Annual means (for normally distributed data) or medians (for not normally distributed data) were reported with standard deviations and ranges, respectively. Population data were not normally distributed but followed a quasi-Poisson distribution (overly dispersed). Such distribution represents better abundance data with occasional high peaks. The aims were to evaluate the changes in abundance of specific species in response to the changes of seasons, salinity, temperature, Chl-a, and RCP. A random variable (i.e. 1| sampling date) was included at the intercepts of all the generalised linear models (glms) that were built for aforesaid purpose. 'GlmmPQL' function of the 'MASS' package of CRAN-R3.3.2 (R Core Development Team, 2017) was used. Confidence intervals were set at 95%. If $\alpha \leq 0.05$ respective W, *t*, p values along with *DF* are reported.

A separate community analysis was performed only on the copepods because they were found as the most prevalent taxonomic group out of 15 others (see Table 1). Number of different copepod species was low (only ten), among them few had high abundances. Species evenness index was calculated (see Table-2) as the ratio of Shannon-Wiener index (H) over the total sample size for sampling site (when a sample was collected). Evenness index (E) was calculated using Community Analysis Package (3.0) for Windows (Seaby and Henderson, 2004). Index (E) was then regressed against above mentioned physico-chemical variables. Response variable E had a normal distribution (W = 0.91, p-value = 0.06) thereby glms were built by considering Gaussian family. A random variable (i.e. 1| sampling date) was included at the intercepts of all the glms. 'GlmmPQL' function of the 'MASS' package of CRAN-R3.2.3 (R Core Development Team, 2016) was used for the purpose. *t*, and p values along with *DF* reported if results were found to be statistically significant.

Results

Mean water temperature was 19.25 ± 4.8ºC; ranged between 10.4 (August) and 26.8 (January) and its variability followed the seasonal pattern (Figure 2). Mean salinity was 9.25 ± 6.18 which typically represents mesohaline conditions; ranged between 1.2 and 24.7. Salinity in autumn and winter were relatively lower than in spring-summer (early) (Figure 2). Chl-a ranged between 0.41 $mg.m^{-3}$ (August) and 66.39 $mg.m^{-3}$(February), had a median of 3.63 $mg.m^{-3}$, and was higher in summer than other seasons (Figure 2). RCP ranged between 0.27 and 8.13 (median 0.53) (Fig. 2). Summer was characterized by low values of RCP (characterised by high values of Chl-a, PHA resulting in a small

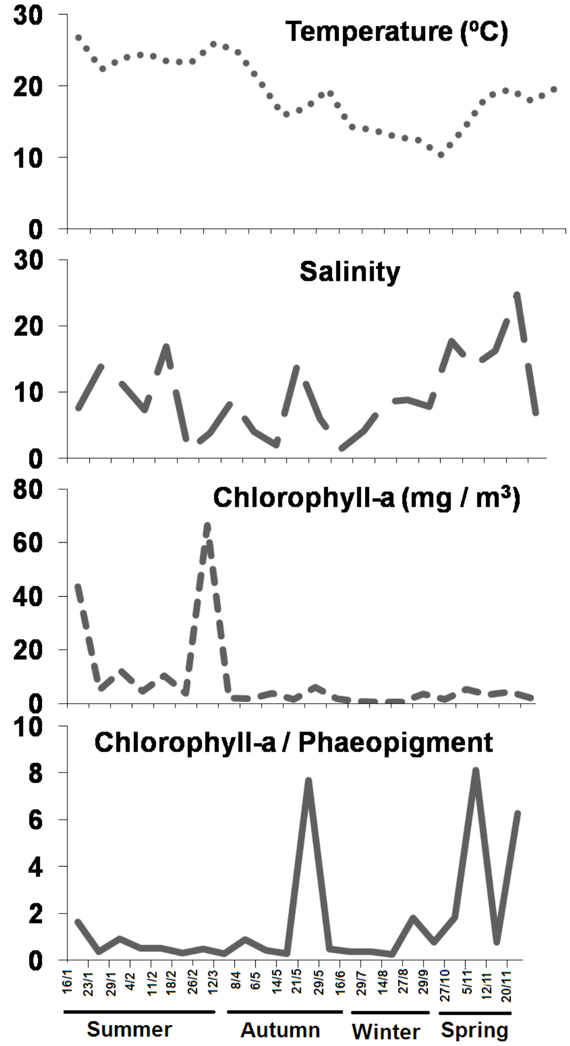

Figure 2: Temporal variability of different physico-chemical drivers' off Montevideo (Rio de la Plata Estuary, Uruguay) in 2003.

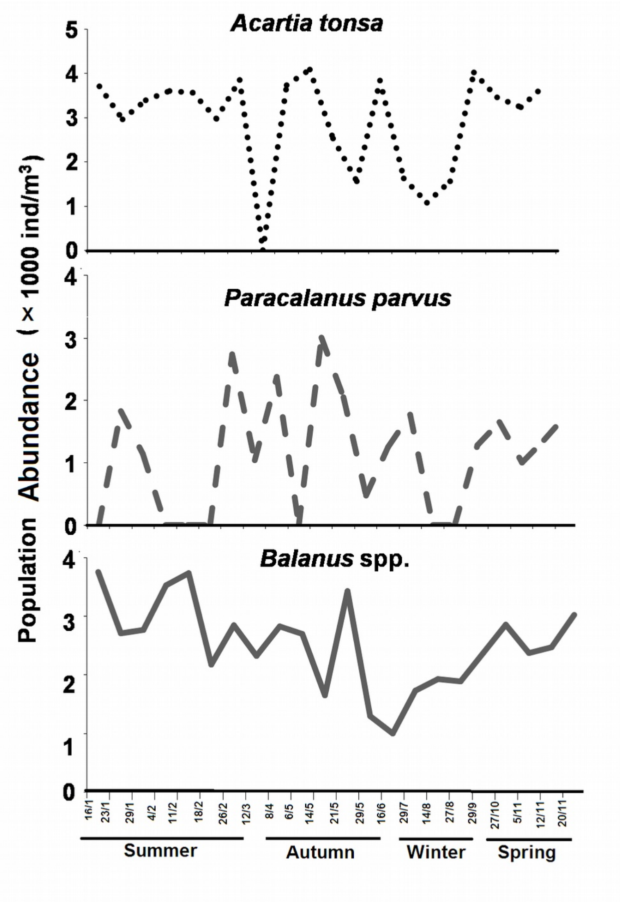

Figure 3: Abundance (Ind/m^3) of some prevalent mesozooplankton sampled off Montevideo (Rio de la Plata Estuary, Uruguay) in 2003.

value of their ratio). The ratio (i.e. RCP) was once high in autumn then declined considerably in winter but in spring it was consistently high (characterised by higher Chl-a andlow PHA values) (Fig. 2).

Irrespective of the season copepod *Acartia tonsa* was the most abundant species (ranged from 7097 to 25606) (Table I) but occasionally *P. parvus* and summed up larval stages of *Balanus* sp. reached high numbers (Table I). Abundances of *A. tonsa* and *P. parvus* were lower in winter but pronounced seasonality was absent (Fig. 3). Abundances of *Balanus* sp. reached the highest values in summer and declined in winter to rise again in spring (Figure 3). Abundances of *A. tonsa* and *P. parvus* were largely uncorrelated with salinity, temperature, and Chl-a. *Acartia tonsa* abundance increased significantly (DF= 20, t = 2.25, P = 0.035) with the high RCP but RCP had no such effects on the abundance of *Paracalanus parvus*. Warmer temperatures increased abundance of *Balanus* sp. (DF= 20, t = 3.95, P = 0.008), which was otherwise unaffected by salinity, Chl-a, and RCP variability. Copepod species evenness (mean = 0.45, SD= ± 0.18, range = 0 to 0.67) (Table 2) remained unaffected despite seasonal cycles (evenness did decrease (t = - 0.54, DF = 18) in winter a little but not significant). Physico-chemical changes such as time variability of salinity, temperature, Chl-a, RCP hardly effected species evenness of copepod community.

Discussion

Results of salinity, temperature, Chl-a, and RCP variability patterns are consistent with the seasonal forcing in the region. In 2003, low salinity (mostly < 10) was persistent from late March to end of August possibly for higher freshwater discharge from the Uruguay River which receives higher

Table II Time variability of copepod species evenness off Montevideo (Rio de la Plata Estuary, Uruguay) in 2003.

Sampling Date	Season	Copepod Evenness
16.01.2003	Summer	0.30
23.01.2003	Summer	0.59
29.01.2003	Summer	0.60
04.02.2003	Summer	0.31
11.02.2003	Summer	0.31
18.02.2003	Summer	0.63
26.02.2003	Summer	0.49
12.03.2003	Summer	0.43
08.04.2003	Autumn	0.30
06.05.2003	Autumn	0.00
14.05.2003	Autumn	0.67
21.05.2003	Autumn	0.66
29.05.2003	Autumn	0.50
16.06.2003	Autumn	0.55
29.07.2003	Winter	0.57
14.08.2003	Winter	0.12
27.08.2003	Winter	0.43
29.09.2003	Spring	0.49
27.10.2003	Spring	0.45
05.11.2003	Spring	0.60
12.11.2003	Spring	0.65

precipitation at that time of the year (Berbery and Barros, 2002). Even such physico-chemical variability of the habitat did not always affect the mesozooplankton there. Seasonal changes had little role in limiting the copepod abundance. Abundance of *Balanus* sp. increased in spring and summer with the rise of temperature in the RPE. Availability of fresh food (RCP) in the habitat possibly triggers the rise of *A. tonsa* numbers. Species evenness of copepod community did neither vary with the seasons nor with the physico-chemical variables studied.

RPE is amongst the widest (38,000 km^2) in the world but it is rather shallow (maximum depth ~ 25 m) (Acha *et al.*, 2008). There are usually neither strong horizontal nor vertical thermal gradients, and temperature tends to oscillate seasonally but within a moderate range (Guerrero *et al.*, 1997), as recorded during the present study. Instead, synoptic hydrography of the RPE is characterized by a horizontal salinity gradient, and a persistent and quasi-permanent salt wedge which generates salinity and turbidity fronts in surface and bottom layers (Guerrero *et al.*, 1997; Mianzan *et al.*, 2001). Gradients and the horizontal position of salinity and turbidity fronts within the RPE generally change at different temporal scales driven by freshwater runoff, winds, and bottom topography (Guerrero *et al.*, 1997). In the current study, the study site Playa del Buceo Bay showed oligohaline to polihaline conditions without a clear seasonal pattern. Chl-a showed a seasonal trend, with highest values in summer and lowest in winter matching the typical pattern of Patos Lagoon in Southern Brazil (Muxagata *et al.*, 2012) and subtropical estuaries of eastern South Africa, and San Francisco bay in the USA (Cole & Cloern, 1984; Froneman, 2004; Murrell & Lores, 2004). In spring, RCP reached its maximum, suggesting active growth conditions of the phytoplankton and a higher fraction of fresh phytoplankton available as food during that season.

Sampled zooplankton assemblages are typical of the estuarine species of the region (Montu *et al*. 1997, Berasategui *et al.* 2006, Cowan *et al.*, 2013). Brackish waters and variable salinity typically make zooplankton assemblages at this sector of the RPE to be taxonomically-poor (Berasategui *et al.* 2006), as found here. High densities of *A. tonsa* and *P. parvus*, and *Balanus* sp. were expected according to previous studies in RPE and nearby waters (Marrari *et al.*, 2004; Calliari *et al.*, 2004, Derisio et al. 2014). Copepods were the most abundant taxa that persisted in all seasons; abundance of *A. tonsa* was high. Species evenness did not change between seasons. The variability of *A. tonsa* or *P. Parvus* did not show seasonality, and neither did species evenness of copepod community. In the subtropical estuaries of USA, *Acartia tonsa* abundance peaks in summer, especially towards the more saline zones (Lawrence *et al.*, 2004). Within the same geographic region, in Patos Lagoon estuary (aprox. 32ª S, North of RPE), Southern Brazil, and in Bahia Blanca estuary (aprox. 39ª S, South of RPE) of Argentina, seasonality in the structure of the mesozooplankton assemblage and the dominance of the copepods are pronounced (Hoffmeyer, 2004; Berasategui *et al.*, 2009, 2013; Montú*et al.*, 1997; Muxagata *et al.*, 2011). In the temperate Bahia Blanca estuary, the seasonal temperature change triggers species succession with *A. tonsa* and *Eurytemora americana* alternating dominance between seasons (Hoffmeyer, 2004). In the subtropical Patos Lagoon of Brazil, mesozooplankton assemblage changes with local hydrography, primarily driven by winds and freshwater runoff (Montú, 1980; Montú *et al.*, 1997). In the Patos Lagoon low salinities prevail during the warmer months and the mesozooplankton assemblage is generally dominated by freshwater copepods (*Notodiaptomus incopositus*, cyclopoids) and cladocerans (*Eubosmina tubicen* and *Diaphanosoma sarsi*). During the cold season higher salinities favor dominance of species of marine origin, with *A. tonsa* and *Balanus* larvae among them (Montú, 1980; Montú *et al.*, 1997). In the present study seasonality in terms of copepod abundance was absent which is uncommon but not unprecedented. For instance, seasonality of mesozooplankton abundance was not pronounced for the sub tropical estuaries of Australia (Robertson *et al.*, 1988) and for most species in the Araike Bay estuary, Japan (Islam *et al.*, 2004, 2006).

Population densities of copepods *A. tonsa* and *P. parvus* remained unaffected despite fluctuations in Chl-a. Abundance of *A. tonsa* did rise in spring with the increase of RCP. Results may be taken as an indication that abundance was not limited by the total amount of algal food, but rather by its quality (freshness). Copepods such as *A. tonsa* are omnivorous, able to feed on a wide spectrum of food including detritus (Roman, 1984). In estuaries which bear high loads of suspended particulate matter (SPM) and accumulate large volume of detritus (e.g., Gironde estuary, France), *A. tonsa* may not be able to obtain its daily ration, and its specific growth rates (P: B ratio) are modulated by the ratio of Chl-a to suspended particulate matter (Irigoien *et al.*, 1995). A similar mechanism may explain the positive relationship between *A. tonsa* abundance and RCP, and the lack of seasonality of *A. tonsa* populations in RPE. Earlier findings on ingestion and egg production rates at 'Playa del Buceo' Bay suggested that *A. tonsa* actually feeds on detritus but attains rather overall low production (Calliari *et al.*, 2004). Exceptionally high egg production rates were attained when abundant high quality food, and particularly microzooplankton, was available. Results on stable isotopic composition and egg production rates from open waters of RPE also support the interpretation of detrital ingestion and production limitation by food quality for *A. tonsa* (Derisio *et al.*, 2006).

Copepods hardly responded to environmental physico-chemical variability which indicates their resilience to environmental fluctuations in the RPE. Such results follow the observations on *A. longipatella* densities in the Gamtoos estuary of South Africa (Paul *et al.,* 2016). In both cases time variability in the population abundances of the most abundant mesozooplankton species were largely uncorrelated to the variability of the physico-chemical drivers studied. This could be an indication of their proper eco-physiological adaptation to the immediate habitat conditions (Paul *et al.,* 2016). The coasts of Brazil and Argentina are experiencing higher variability of salinity and temperature (Reguero *et al.*, 2013; Barros *et al.*, 2015). Such may negatively affect the survival of crustacean mysids along Uruguayan coast (Paul and Calliari, 2017). It would be also of interest to assess the potential responses of prevalent mesozooplankton of RPE to altered environmental scenarios.

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