

Seasonal dynamic of macroalgae in intertidal pools formed by beds of *Crassostrea gigas* (Mollusca, Bivalvia) on the north Patagonian Atlantic coast

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Abstract

Intertidal pools represent restricted habitats for macroalgal assemblages, and their structure is determined by physical and biotic factors. The establishment of the exotic oyster *Crassostrea gigas* on the north Patagonian Atlantic coast led to the formation of intertidal pools; these provide habitats for macroalgal settlement. We aimed to determine the composition, distribution and abundance of the macroalgal assemblage inhabiting these pools, and to establish the structuring variables. Several biotic and abiotic variables were evaluated in 36 pools in winter and summer. Correlations were calculated and permutation analysis of variance and constrained redundancy analysis were conducted to determine the structuring variables. Unlike other Patagonian coasts, richness was low and pools were characterized by a dominance of red algae. Richness was higher in winter, while diversity was lower because of the dominance of *Polysiphonia abscissa*, which had the highest percentage of occurrence. Temperature was a putative factor determining seasonal richness and diversity. Elevation and surface area of the pools were structuring factors for assemblage biomass. We demonstrated distinct assemblage dynamics. *Polysiphonia abscissa* developed large populations in winter, which declined towards the summer, while *Gelidium maggsiae* and *Gelidiella calcicola* formed dense turfs during summer stress.

Keywords: *Crassostrea gigas*; intertidal pools; macroalgae; north Patagonian Atlantic coast; *Polysiphonia abscissa*.

Introduction

Intertidal pools are well-known habitats in coastal environments; they have been the subject of study for many decades (e.g., Johnson and Skutch 1928, Underwood 1976,

Underwood and Jernakoff 1984, Metaxas et al. 1994, Nielsen 2001, Denny and Gaines 2007). However, the amount of information on intertidal pools is much more limited than that for emergent substrata (Metaxas and Scheibling 1993). The main difference between the subtidal habitat and intertidal pools is that the latter are isolated during low tide, and they therefore represent constrained habitats for marine organisms. For example, macroalgae are frequently found in these habitats, where they play a fundamental role as primary producers (Valiela 1991); their life cycles are strongly related to the characteristics of the pools, because changes in the biotic and abiotic conditions of the pools may stimulate the formation and subsequent release of reproductive stages (Dring 1974). As suggested by De Meester et al. (2005), ponds and pools, broadly defined as small and shallow water bodies, are suitable systems for modeling communities because of their diverse responses to ecological gradients. The same concept can be applied to intertidal pools, since they are affected by ecological gradients in the coastal environment.

Oysters are well-known ecosystem engineers (Ruesink et al. 2005, Bouma et al. 2009); they form dense reefs and create new habitats when they occur in soft-sediments (Lang and Buschbaum 2010). The Pacific oyster *Crassostrea gigas* Thunberg, in particular, is among the most important engineer species in marine environments (Klinger et al. 2006, McKindsey et al. 2007). On the north Patagonian Atlantic coast of Argentina, *C. gigas* has developed large populations since it was introduced in Anegada Bay. These populations, commonly known as “beds”, have led to the formation of shallow intertidal pools between oyster individuals, and recent studies have confirmed that these pools are appropriate habitats for the establishment of macroalgal populations (Borges 2006). There is worldwide evidence that introduced exotic marine species can cause disturbances by changing physical and abiotic factors, and consequently, the structure of biological assemblages (Commito et al. 2008, Lang and Buschbaum 2010). For example, the settlement, survival and death of oyster individuals may have a direct influence on the configuration of these pools. In addition, physical factors and biotic interactions combined are significant aspects structuring the intertidal pools assemblages that have been addressed by several authors (Kooistra et al. 1989, Metaxas et al. 1994, van Tamelen 1996, Methratta 2004, Araujo et al. 2006). For example, Zhuang (2006) determined that season and size of tide pools significantly influence richness, biomass and diversity of macroalgal species. Moreover, Dethier (1984) demonstrated that the biota of a pool can be partially predicted by pool variables, such as elevation, size, and season.

As suggested by Sodhi and Ehrlich (2010), given that biodiversity is deteriorating because of the loss of native habitats, it is fundamentally important to understand the composition of the new communities that occupy modified habitats. In Anegada Bay, although the establishment of the oyster has negatively impacted the native seascape of the coast, biological information about this natural ecosystem is scarce and incomplete (Alvarez and Rios 1988, Borges 2005, 2006). Furthermore, to the authors' knowledge, there are no studies on macroalgal communities in intertidal pools on the Patagonian Atlantic coast. Most knowledge of macroalgal communities on the Patagonian Atlantic coast comes from studies carried out in southern Patagonia, and these were focused on commercial species (Casas et al. 2004), while the northern Patagonian Atlantic coasts remain unstudied (Bremec and Roux 1997). In this context, the intertidal pools located among *C. gigas* populations constitute priority habitats for biodiversity studies. The present study attempted to characterize the macroalgal assemblage that inhabits the intertidal pools of Anegada Bay. Specifically, we aimed: (1) to estimate macroalgal diversity, abundance and spatio-temporal distribution in two different seasons; (2) to characterize the pools based on abiotic variables; (3) to ascertain the main abiotic factors structuring the macroalgal assemblage; and (4) to determine the reproductive state of the populations.

Materials and methods

Location and sampling site

The study was carried out in Los Pocitos (40°25' S; 62°25' W), a coastal town located in Anegada Bay on the north Patagonian Atlantic coast of Argentina (Figure 1). The coast is constituted of tidal flats of mobile sediment (mainly lime and clay), and coastal meadows of *Spartina* sp. interrupted by reefs of consolidated sandstone (Spalleti 1980). The tidal regime is semi-diurnal, with maximum amplitude of 2.51 m during the sampling period (SHN 2011). The bay has one freshwater discharge, but the volume is negligible and the currents produce an important mixture of fresh and salt water. Several beds of *Crassostrea gigas* were detected in the 1990s in both subtidal and intertidal habitats; these are the only established beds known in Argentina. Their origin is still unknown (Borges 2005). The most significant features of *C. gigas* organisms are their greater size and more irregular contour compared to the native oysters *Ostrea spreta* d'Orbigny and *O. puelchana* d'Orbigny. Oyster larvae settle on consolidated sandstone and grow adopting the shape of the substratum; new larvae settle and subsequently form a bed. There are no individuals of the native oysters in Los Pocitos beds, but the native mussel *Brachidontes rodriguezii* d'Orbigny is abundant among the oysters and attaches on the top of their valves. The average extension of these beds is 127.98 m², with an average oyster density of 11 m⁻² (Borges 2006). The intertidal pools located in these beds were selected for sampling.

Sampling

Two different sets of 18 pools along the oyster beds were surveyed during the lowest monthly tide, one set in winter (July 2009) and the other in summer (March 2010). The pools were randomly selected along transects located haphazardly and orthogonal to the shoreline on the intertidal gradient between the waterline and the beginning of *Spartina* meadows (approx. 120 m long). In order to characterize the pools, the following abiotic variables were measured in each: surface area, elevation, temperature, salinity and location on the intertidal gradient. Surface area was estimated from photographs taken horizontally above the surfaces of the pools, using the UTHSCSA Image tool version 3.00 software (developed by University of Texas Health Science Center of San Antonio, Texas, ftp://maxrad6.uthscsa.edu, 2010). Elevation was estimated using graduated rules and following the Emery method, which consists of measuring the slope from a fixed point (intertidal pool) relative to the ocean (lining up the rule with the horizon) (Emery 1961). Salinity and temperature were measured with a HORIBA-U10 multisensor (HORIBA, Kyoto, Japan) at the beginning of the period of isolation during ebb tide. The location of each pool on the intertidal gradient was arbitrarily established by measuring the length of the intertidal zone (orthogonal to the shoreline) from the waterline

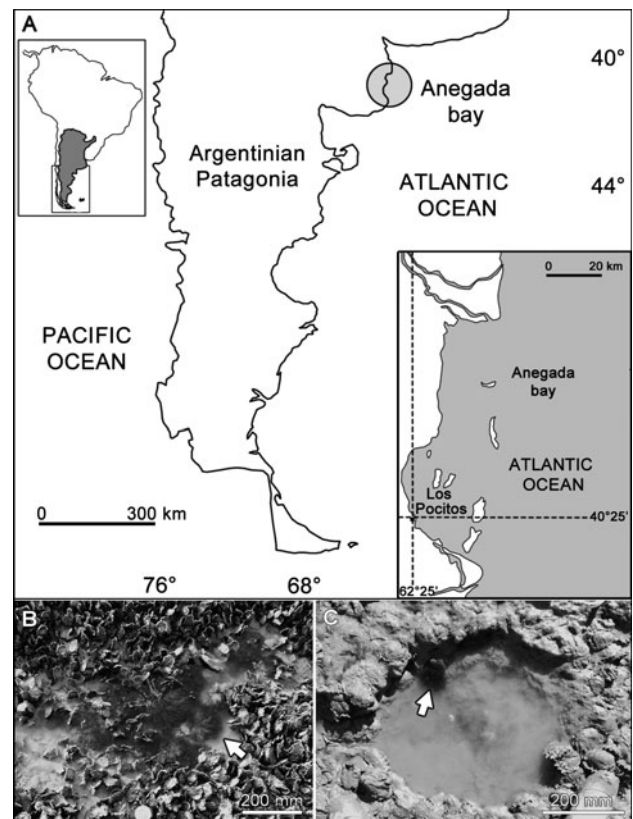


Figure 1 (A) Location of the sampling site on the north Patagonian Atlantic coast (■). (B) and (C) Intertidal pools among *Crassostrea gigas* individuals; white arrows indicate red algae growing inside the pools.

up to the beginning of *Spartina* meadows, and then dividing the length into three sectors of equal length: low, intermediate and high. The location of each pool in each sector was then recorded, resulting in a random stratified design.

To characterize the macroalgal assemblages, the presence of each macroalgal species was recorded in every pool, and all biomass was collected into plastic bags and transferred to the laboratory in cold boxes. Macroalgae were rinsed with filtered seawater to remove sediment. All specimens were examined under a binocular microscope to find reproductive individuals. Macroalgae were drained on blotting paper, then weighed and placed in aluminum cups to be dried in an oven at 60°C to constant weight. Macroalgal wet weight (WW) and dried weight (DW) were measured using an analytical scale.

Data analyses

In order to determine seasonality and spatial distribution of macroalgal assemblages, macroalgal species richness was determined for each pool and season, and the seasonal percentage of occurrence for each species in the pools was also calculated. Seasonal diversity was estimated with the Shannon index (H') using DW data, and the theoretical maximum value (H'_{\max}) for each season was calculated as:

$$H' = -\sum_{i=1}^S (p_i \cdot \log_2 p_i)$$

$$H'_{\max} = \log S$$

where S is the total number of species and p_i is the proportional abundance of species i (Shannon and Weaver 1949).

Specific relative biomass was estimated for every pool using weight and surface area data. The total relative biomass corresponding to red, brown and green algal groups was also calculated for each pool. Weight data were first transformed to $\ln(x+1)$. In order to test the effects of season on the abundance of the dominant group of macroalgae, two analyses were performed using DW data: Kruskal-Wallis test was used for red algal abundance because of non-normality of DW data (tested with Shapiro-Wilk test, Royston 1982), and analysis of variance (ANOVA) was used for the dominant species abundance, after testing normality and homoscedasticity of data.

ANOVA was used to compare elevation of pools located in the three intertidal levels previously defined. The results showed no significant differences between pools located in the high and intermediate levels ($F=2.07$; d.f.=22; $p=0.8$), but both were significantly different from pools located in the lower intertidal ($F=2.14$; d.f.=12; $p<<0.01$). This allowed the re-classification of the pools within two levels: low intertidal pools (pools located between 0 m and 38 m from the waterline) and high intertidal pools (pools located higher than 38 m from the waterline up to the *Spartina* meadow limit). We used this classification of pools as a categorical variable to compare macroalgal distributions along the intertidal gradient.

In order to determine the main abiotic factors structuring the macroalgal assemblage, several analyses were conducted. Seasonal salinity and temperature values were analyzed with

ANOVA. Correlations between biotic and abiotic variables were calculated, using the Spearman's rank coefficient and tested for significance (Zuur et al. 2007). Because of non-normality of richness and biomass data (tested with Shapiro-Wilk test, Royston 1982), two non-metric analyses were conducted in order to evaluate richness, total DW and red algal DW in relation to the abiotic variables. Permutation analysis of variance (PERMANOVA) was conducted for the categorical variables: season and location on the intertidal gradient (Anderson 2001). A constrained distanced based redundancy analysis (db-CCA) was used for the continuous variables temperature, salinity, and surface area (Legendre and Legendre 1998). Pools were classified based on Jaccard dissimilarities in macroalgal species occurrences, and a non-metric multidimensional scaling (NMDS) ordination was conducted. The result of the ordination was plotted and the value of "stress" was obtained, which indicated minimum distortion incurred by compressing data into two dimensions. All statistical analyses were done using R software (R Development Core Team 2011).

Results

Assemblage composition and abiotic factors

In the 36 pools analyzed, Rhodophyceae (four species), Phaeophyceae (three species) and Chlorophyceae (three species) were evenly represented. A total of 10 macroalgal species were identified, of which five were found only in winter: *Punctaria latifolia* Greville, *Scytosiphon lomentaria* (Lyngbye) Endlicher, *Ulva* sp., *Ectocarpus siliculosus* (Dillwyn) Lyngbye, and *Cladophora albida* (Hudson) Kützing; four were found only in summer: *Ceramium strictum* Greville ex Harvey, *Gelidium maggsiae* Rico et Guiry, *Gelidiella calcicola* Maggs et Guiry and *Bryopsis corticulans* Setchell; one was found in both seasons: *Polysiphonia abscissa* Hooker et Harvey. *P. abscissa* had the highest percentage occurrence; whereas *E. siliculosus* had the lowest, appearing in only one of the 36 pools surveyed (Table 1). *G. maggsiae* and *G. calcicola* were always found in the form of dense turfs that were difficult to separate; they were thus pooled as "red algal turfs" in the analyses.

Pool species richness varied between one and five. Most pools had one species; one pool located in the high intertidal attained maximum richness in winter (Figure 2). Richness, diversity and abiotic variables are summarized in Table 2. The seasonal averages were similar for all variables, except for temperature, diversity and H'_{\max} . Diversity was higher in summer; however in both seasons it was lower than H'_{\max} . Highly significant differences were found between seasons in pool temperature ($F=1229.15$; d.f.=1; $p<<0.01$) and pool salinity ($F=9.79$; d.f.=1; $p<<0.01$). Significant correlations were found in all biotic and abiotic factors (Table 3). Richness was positively correlated with green and brown algal DW, but negatively correlated with pool area. Area and elevation were negatively correlated with green and red algal DW, respectively. Brown algal DW was positively correlated with green algal DW, but negatively correlated with temperature. Finally, salinity was positively correlated with temperature.

Table 1 Presence-absence and percentage occurrence of macroalgae in intertidal pools, in winter (A) and summer (B).

(A)	Winter																		Occurrence (%)
	Low intertidal						High intertidal												
Pool number	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	
<i>Punctaria latifolia</i> Greville							X	X	X	X		X							27.8
<i>Cladophora albida</i> (Hudson) Kützing	X						X		X	X				X					27.8
<i>Scytosiphon lomentaria</i> (Lyngbye) Endlicher								X	X	X		X							22.2
<i>Ulva</i> sp.							X			X		X		X	X				27.8
<i>Polysiphonia abscissa</i> Hooker et Harvey	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	83.3
<i>Ceramium strictum</i> Greville ex Harvey																			
<i>Ectocarpus siliculosus</i> (Dillwyn) Lyngbye																X			5.6
<i>Bryopsis corticulans</i> Setchell																			
Red algal turfs ¹																			
Pool richness	2	1	1	1	1	1	4	3	4	5	1	4	1	3	3	0	0	0	
(B)	Summer																		Occurrence (%)
	Low intertidal						High intertidal												
Pool number	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	
<i>Punctaria latifolia</i> Greville																			
<i>Cladophora albida</i> (Hudson) Kützing																			
<i>Scytosiphon lomentaria</i> (Lyngbye) Endlicher																			
<i>Ulva</i> sp.																			
<i>Polysiphonia abscissa</i> Hooker et Harvey	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	94.4
<i>Ceramium strictum</i> Greville ex Harvey				X	X	X													16.7
<i>Ectocarpus siliculosus</i> (Dillwyn) Lyngbye																			
<i>Bryopsis corticulans</i> Setchell								X		X				X					16.7
Red algal turfs ¹	X	X	X	X	X					X			X	X	X	X	X	X	66.7
Pool richness	2	2	3	3	3	1	1	2	1	3	1	1	2	3	1	2	2	2	

¹*Gelidium maggsiae* and *Gelidiella calcicola* in mixed stands.

Seasonal abundance and distribution

Total biomass was 5598.88 g WW (849.26 g DW) in winter and 1158.45 g WW (240.37 g DW) in summer. Relative biomass was 354.78 g m⁻² WW (53.81 g m⁻² DW) in winter

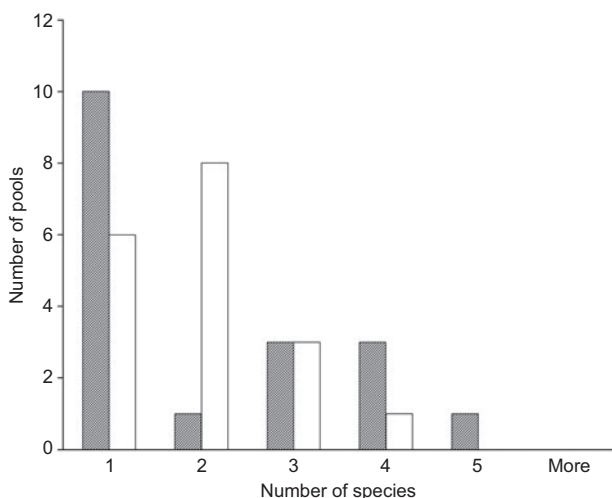


Figure 2 Frequency distribution of numbers of macroalgal species in intertidal pools.

Hatched and white bars represent pools in winter and summer, respectively.

and 69.28 g m⁻² WW (14.37 g m⁻² DW) in summer. Figure 3 shows mean relative biomass of each macroalgal species in both seasons and both locations. *Polysiphonia abscissa* was the most abundant taxon in both seasons, with 342.2 g m⁻² WW (51.77 g m⁻² DW) in winter and 61.3 g m⁻² WW (12.26 g m⁻² DW) in summer. There were no significant differences in red algal DW between seasons (Kruskal-Wallis $\chi^2=2.22$; d.f.=1; p=0.13), but *P. abscissa* DW was significantly higher in winter than in summer ($F=5.12$; d.f.=1; p<0.03).

There were significant differences in total DW between seasons ($F=3.97$; d.f.=1; p=0.03), and among locations ($F=4.76$; d.f.=1; p=0.01). The interaction between season and location

Table 2 Seasonal averages of abiotic variables, species richness and diversity.

	Winter	Summer
Area (m ²)	0.88 (± 0.57) ¹	0.93 (± 0.86)
Elevation (cm)	52.72 (± 30.96)	63.22 (± 30.93)
Temperature (°C)	9.17 (± 1.77)	21.51 (± 0.53)
Salinity	31.1 (± 0.7)	32.3 (± 1.5)
Richness	6	5
H ²	0.26	0.69
H ³ _{max}	2.58	2

¹Values in parenthesis are SDs. ²H¹ Shannon diversity. ³H¹_{max} theoretical H¹ maximum value.

Table 3 Significant correlation coefficients between biotic and abiotic variables.

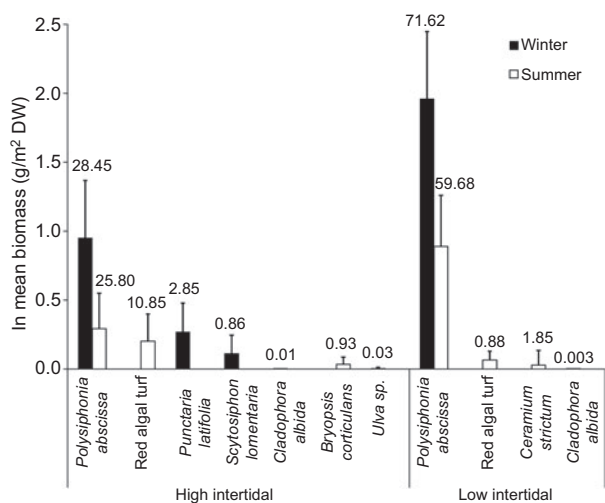
	Correlation
Area vs. richness	-0.86**
Area vs. green algae DW	-0.93**
Elevation vs. red algae DW	-0.77*
Temperature vs. brown algae DW	-0.80*
Salinity vs. temperature	0.85**
Richness vs. brown algae DW	0.78*
Richness vs. green algae DW	0.88**
Brown algae DW vs. green algae DW	0.74*

*Significance, $p < 0.05$. **Significance, $p < 0.01$. DW, dry weight.

was not significant ($F=1.25$; d.f.=1; $p=0.27$). The db-CCA for species richness was not significant ($F=0.34$; d.f.=4; $p=0.87$), but that for total DW was highly significant ($F=4.46$; d.f.=4; $p=0.005$), and the variables that explained most of the variance in DW were pool area ($F=8.05$; d.f.=1; $p=0.02$) and pool elevation ($F=6.09$; d.f.=1; $p=0.03$). The constrained distance-based redundancy analysis for red algal DW was also highly significant ($F=5.40$; d.f.=4; $p=0.005$), and the significant explanatory variables were again pool area ($F=10.05$; d.f.=1; $p < 0.01$) and pool elevation ($F=7.48$; d.f.=1; $p < 0.01$).

The NMDS ordination plot revealed a separation between winter and summer pools. Except for pools number 11 and 13, the rest of the winter pools located in different intertidal elevations were separated in two distinct groups: low intertidal pools, pools 1–6 and 19–24; high intertidal pools, pools 7–18 and 25–36; summer pools did not segregate by intertidal elevation (Figure 4).

Species ordination in the plot also denotes the seasonal and spatial segregation of species. Except for *Cladophora albida*,

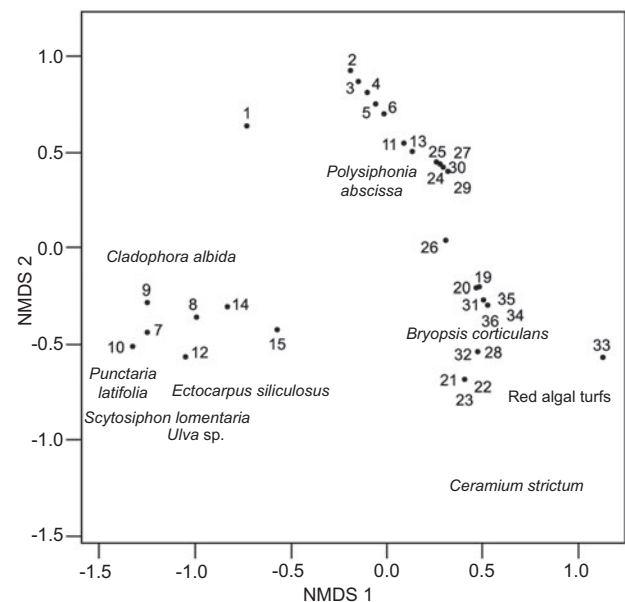
**Figure 3** Abundances of macroalgal species. Numbers above bars indicate percentage of total dry weight.

As a result of the low biomass of the single specimen found, *Ectocarpus siliculosus* was not included in the graphic. "Red algal turf" comprised *Gelidium maggsiae* and *Gelidiella calcicola* in mixed stands. Values are means+SD.

the winter species, *Punctaria latifolia*, *Ectocarpus siliculosus*, *Scytosiphon lomentaria*, and *Ulva sp.*, were associated with high intertidal pools. On the other hand, summer species were restricted to different levels of the intertidal. *Ceramium strictum* was limited to low intertidal, *Bryopsis corticulans* was associated with the high intertidal, and red algal turfs were distributed through the intertidal gradient. As seen in the plot, *Polysiphonia abscissa* occurred in unialgal pools located through the intertidal and during both seasons.

Phenology

Some specimens of brown and red algae were fertile in some of the surveyed pools. Fertile red macroalgae were *Polysiphonia abscissa*, *Gelidium maggsiae* and *Ceramium strictum*. Carposporophytes and tetrasporophytes of *P. abscissa* occurred in both seasons. Female gametophytes carrying fertile branches and developed carposporangia were also found on specimens of *G. maggsiae*, but only in summer. Female gametophytes of *C. strictum* with mature carposporangia were also found in summer. The only fertile brown macroalga was *Ectocarpus siliculosus* with plurilocular sporangia in winter. There were no fertile specimens of green macroalgae. No fertile male gametophyte was found for any of the species. It is important to notice that the winter individuals of *P. abscissa* were well developed, whereas in summer they were smaller, bleached and heavily covered with diatoms and protozoans.

**Figure 4** Two-dimensional non-metric multidimensional scaling ordination plot of intertidal pools based on Jaccard dissimilarities in macroalgal species occurrence (stress=0.06).

Winter pools: numbers 1–15; summer pools: numbers 19–36; low intertidal pools: numbers 1–6 and 19–24; high intertidal pools: numbers 7–18 and 25–36. Pools 16, 17 and 18 were not included in the analysis since no macroalgae were recorded in them. Species were plotted according to their maximum correlation with the configuration of pools in the ordination space.

Discussion

Richness, diversity and seasonality

The three major groups of marine macroalgae were present in the intertidal pools of Los Pocitos oyster beds, with almost the same number of species in each group, although there was a marked seasonality in their occurrences. In winter the three groups were well represented, while in summer green algae were represented by only one species, and brown algae were absent. Red algae were found during both seasons. Similarly, Connor (1980) observed that some species of red algae are present throughout the year in intertidal pools associated with oyster reefs, including two species of *Polysiphonia*, while other genera such as *Ceramium*, *Chondria* and *Griffithsia* are occasional. Zhuang (2006) also found that red algae are the principal group inhabiting intertidal pools throughout the year. Unlike other Patagonian coasts, where brown algae form a dominant canopy (Quartino and Boraso de Zaiuso 2008), intertidal pools from Los Pocitos were characterized by a dominance of red algal turfs.

There was also a marked seasonality in the composition of macroalgal assemblages. Winter pools were more divergent because half of the pools were uni-algal (i.e., only one species was present) while the rest had different assemblages of from two to five species of macroalgae. Summer pools were very similar in composition and typically bi-algal (i.e., only two species were present), since the majority of the pools were inhabited by the assemblage comprising *Polysiphonia abscissa* and red algal turfs. As in the present study, higher richness in winter was also observed by Araujo et al. (2006) in intertidal pools from Portugal, and by Connor (1980) among oyster reefs in Chesapeake Bay.

The total number of species in our collections was low compared to southern Patagonian coasts (Casas et al. 2004). For example, calcified macroalgae, which are typical of intertidal pools, were not recorded even though they are common along the Patagonian coast (Mendoza 1974, Bertness et al. 2006). Borges et al. (2005) studied algal recruitment on subtidal plates in Los Pocitos oyster beds and identified the species *Ectocarpus siliculosus*, *Hinckesia mitchelliae* (Harvey) P.C. Silva, *Chaetomorpha aerea* (Dillwyn) Kützinger, *Cladophora albida* (as *C. pellucida* (Hudson) Kützinger), *Ulva rigida* C. Agardh, *Polysiphonia abscissa* (as *P. argentinica* W.R. Taylor) and unidentified species of *Ulothrix* and *Cutleria*. In our survey of intertidal pools, *E. siliculosus*, *C. albida*, *P. abscissa* and *Ulva* sp. were found.

Low richness has also been observed in other soft bottom systems and oyster reefs, which has been attributed to great variability of salinity, temperature, turbidity and nutrient concentrations (Connor 1980, Thomsen et al. 2006). In Los Pocitos, salinity was within the usual range for this region (Alvarez and Rios 1988). Although there were significant differences in salinity between seasons, correlation between salinity and richness was not significant, neither was the correlation between salinity and biomass. This suggests that salinity is not related to low richness in the short term. However, water temperature was a putative factor explaining seasonal

changes in diversity and biomass (up to 10°C higher in summer than in winter). There is considerable evidence that sediment deposition is negatively related to species richness and diversity (e.g., Moore 1972, Neushul et al. 1976, Wittenberg and Hunte 1992, Airoidi and Virgilio 1998). Although turbidity and sedimentation rates were not measured in the present study, these parameters may be related to low richness, since there is high mobility of the fine sediment in this habitat (Borges 2006).

Species in this habitat can be classified as perennial (*Polysiphonia abscissa*), frequent in summer (*Gelidium maggsiae* and *Gelidiella calcicola*), ephemeral (*Punctaria latifolia*, *Scytosiphon lomentaria*, *Cladophora albida*, *Ulva* sp., *Bryopsis corticulans* and *Ceramium strictum*) and rare (*Ectocarpus siliculosus*).

Spatial distribution and structuring factors

Elevation was a structuring factor in the spatial distribution of macroalgal biomass in the intertidal pools. Several authors have pointed out that biomass in tidal pools decreases with increasing elevation and that low intertidal pools, are usually more diverse and similar to subtidal communities (Gustavsson 1972, Femino and Mathieson 1980, Hugget and Griffiths 1986, Metaxas and Scheibling 1993). In the present study, the same relationship was found for biomass, but not for richness. Higher pools had more species in winter, while in summer the number of species was higher in pools located in the low intertidal zone, also agreeing with Araujo et al. (2006). This pattern may be related to more stressful conditions in higher pools during the summer, particularly increased desiccation. This postulate was examined by Therriault and Kolasa (2001), who found that richness, diversity and abundance of organisms decreased with increased frequency of pool desiccation in rock pools. Our graphical ordination of pools emphasized the seasonal and spatial differences in pool assemblages. Algal assemblages in high intertidal pools were different than those located in low intertidal pools, but this gradient of distribution was more evident in winter. In winter, lower pools were dominated by *Polysiphonia abscissa*, and higher pools contained the rest of winter species (*Punctaria latifolia*, *Scytosiphon lomentaria*, *Ectocarpus siliculosus*, and *Ulva* sp.). *Cladophora albida* was found at both intertidal levels, suggesting that this species may endure desiccation stress to some point; similar results were found by Kooistra et al. (1989). In summer, this difference between levels was less marked because there were fewer species and the three dominant species (*P. abscissa*, *Gelidium maggsiae* and *Gelidiella calcicola*) were found through the intertidal gradient.

Surface area of the pool was another structuring factor, and was negatively correlated with green algal DW, which suggests that green algae are more abundant in small pools. To the contrary, Zhuang (2006) found that larger pools (0.5–1.2 m²) contained higher macroalgal biomass than smaller pools (<0.2 m²). However, since red algae were more abundant in lower pools, our results suggest that macroalgae living in lower and/or smaller pools have an advantage, because larger as well as higher pools may be more environmentally stressful,

probably as a result of a higher evaporation rate (Hugget and Griffiths 1986).

The results of the multivariate analyses suggest that pool richness was not structured by the variables considered in this study. It is more likely that richness is not an appropriate parameter for a multivariate approach to characterize the macroalgal assemblages from Los Pocitos pools, because low richness led to insignificant differences in richness between pools, despite the differences in composition of the assemblages.

Importantly, macroalgae were always living inside the pools and were never found on exposed substratum. A possible explanation is that the wide tidal amplitude and the large span of the intertidal zone (orthogonal to the waterline) limit the establishment of macroalgae. The presence of macroalgae limited to intertidal pools may suggest that *Crassostrea gigas* populations have extended their range of distribution; nevertheless experiments are needed to test this hypothesis.

Macroalgal abundance

Unlike other studies in temperate regions, which have found maximum macroalgal biomass in summer (Connor 1980, Wolfe and Harlin 1988, Airolidi and Virgilio 1998, Thomsen et al. 2006, Zhuang 2006), total biomass in Los Pocitos was three times higher in winter than in summer, with red algae forming the largest percentage of the total DW, thus explaining lower diversity during this season. Batzli (1969) found similar results in the intertidal pools from the American Pacific coast.

The dynamics of the macroalgal assemblage were dominated by red algae, as demonstrated by the redundancy model for total DW and red algal DW. Dominance of red algae in the intertidal zone was also reported on other coasts (Shepherd 1981, Núñez-López and Casas Valdez 1998, Araujo et al. 2006). However, seasonal changes in macroalgal biomass and composition revealed interesting dynamics for red algae. *Gelidium maggsiae*, *Gelidiella calcicola* and *Polysiphonia abscissa* were dominant during the warm season, whereas in the cold season, brown and green algae became prominent and *P. abscissa* was the only dominant red alga. As a consequence of this dominance in both seasons, diversity never reached the maximum theoretical value estimated for each season.

The fact that *Polysiphonia abscissa* and red algal turf were dominant throughout the year could be associated with sediment dynamics, which could interfere with the recruitment of algae that do not form turfs (Sousa et al. 1981, D'Antonio 1986). There are also studies that demonstrate that prevalence of algal turfs is related to their ability to accumulate sediment (Sousa et al. 1981, Seapy and Litter 1982, Stewart 1989). Although *P. abscissa* was the only species present in both seasons, given the high mobility of the sediment in this coast (Cuadrado and Gomez 2010), it can be expected that the basal portions of red algal turfs, remained buried by sediment and sprout in summer, as suggested by Airolidi et al. (1996). Airolidi and Virgilio (1998) found a negative relationship between *P. setacea* biomass and the amount of trapped sediment. In this context, scouring by sediment could be regarded

as responsible for the summer decrease of *P. abscissa* (Airolidi et al. 1995).

Zhuang (2006) related seasonal differences in biomass to the temperature for growth. In Los Pocitos, high temperature seemed to be crucial for *Polysiphonia abscissa* populations, significantly reducing their abundance in summer. *P. abscissa* has also been recorded for colder latitudes on southern Patagonian Atlantic coasts (Pujals 1963), suggesting that this species could be classified as a cold-water species, which also explains its higher abundance during the winter season.

Mei and Schiel (2007) demonstrated that the prostrate basal axes play an important role in establishing and maintaining *Polysiphonia* turfs in rocky coast of New Zealand by vegetative propagation. The results from the present study led us to consider that *P. abscissa* has a perennial habit because of the prostrate basal system of the thalli which may resist temperature changes and sediment deposition, and from which the whole plant regenerates when conditions become favorable again.

There is evidence that oyster beds can modify habitat hydrodynamics, surface roughness and consequently alter the dynamics of nutrients, sediments and temperature of the environment (Dame 1996, Folkarda and Gascoigne 2009, Padilla 2010). In this context, the seasonal influence of temperature on the macroalgal assemblage located on *Crassostrea gigas* beds may be increased compared to other unaltered coasts in Anegada Bay, because of the presence of the exotic oyster.

The establishment of *Crassostrea gigas* in Anegada Bay is relatively recent, so little is known about its interaction with the benthic native organisms. One study demonstrated higher densities of infaunal and epifaunal organisms inside oyster beds, possibly related to increasing habitat structure and refuge (Escapa et al. 2004). Moreover, there is no information about oyster-macroalgal interactions in this habitat. It is generally recognized that oysters and macroalgal associations can be used positively in aquaculture (Qian et al. 1996, Shpigel and Neori 1996, Jones et al. 2001). In Anegada Bay, *P. abscissa* may benefit from nutrients loading by *C. gigas*, thus leading to an important increase in biomass during the winter season.

Phenology

There was evidence that complete triphasic life cycles occurred in all red algal species found. The presence of carposporophytes indicates that sexual reproduction indeed occurred, thus the absence of male plants suggests that the fertilization phase had already passed. Fertile specimens of *Ulva* sp. were not found. It is hardly probable that this species reproduces sexually in this habitat, because of its low occurrence and low biomass in the intertidal pools, and DeWreede and Klinger (1988) suggest that the probability of successful sexual reproduction may be determined in part by some critical gamete density.

Polysiphonia abscissa has delicate filaments and branches, which become overgrown by diatoms and bleached when

habitat conditions become adverse. As in the present study, Borges et al. (2005) observed that *P. abscissa* (as *P. argentinica*) was highly epiphytized on the recruitment plates. On the contrary, *Gelidium maggsiae* and *Gelidiella calcicola* were less epiphytized, possibly as a result of some mechanical, physical or chemical defense of the thalli (Wahl 1989).

Conclusion

This is the first study on the macroalgal assemblages of intertidal pools formed by beds of *Crassostrea gigas* in Anegada Bay, and provides original and up-to-date information on the benthic communities of the north Patagonian Atlantic coasts. The macroalgal assemblage was species poor, had low diversity and was dominated by three species of red algae with different life strategies: *Gelidium maggsiae* and *Gelidiella calcicola* withstood summer abiotic stress, and presumably sediment dynamics, by growing in dense turfs, whereas the perennial individuals of *Polysiphonia abscissa* took advantage of the low temperature and developed large populations from prostrate axes. Surface area of the pool, pool elevation and season were the main factors structuring biomass dynamics, especially during the warm season. It remains to be determined which advantages *P. abscissa* and red algal turfs have over green and brown algal populations. Focus on sedimentation rates and macroalgal-oyster interactions should be considered in further studies.

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