

## Spatial and temporal variability in algal epiphytes on Patagonian *Dictyota dichotoma* (Dictyotales, Phaeophyceae)

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### ABSTRACT

The present study investigates the epiphytic communities on individuals of a population of the brown macroalga *Dictyota dichotoma*, from Las Grutas beach (40°82' S, 65°09' W), growing on the Patagonian coast, Río Negro Province, Argentina. The composition and spatial and temporal variability were investigated during an annual cycle, to relate the seasonal variations of vegetative and reproductive characteristics of the thallus with the richness, diversity and evenness of the epiphytic community, and their relationship with daylength and seawater temperatures. The epiphytic community consisted of 32 different taxa. The most abundant epiphytic groups were Chlorophyceae, Bacillariophyceae and Cyanophyceae. Only a few epiphytes were present throughout the year. Epiphytic richness and abundance were highest in the middle and basal host regions. The high basal abundance was accounted for by four epiphytic species: *Microchaete aeruginea*, *Pylaiella tenella*, *Ulveella marchantiae* and *Audouinella secundata*. Seasonally, the epiphytic load was highest in winter, and largely accounted for by *Grammatophora marina*, *Cocconeis* sp. and *P. tenella*. The epiphytic load did not show variation among the different host reproductive stages, and no relationship was observed between thallus host length and dry mass.

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### 1. Introduction

A large number of marine algae can grow epiphytically on macroalgae or seagrasses (Rindi and Guiry, 2004), making epiphytism a widespread phenomenon in marine benthic communities. In fact, epiphytes may use the host not only as a substrate for attachment, but also for acquiring enhanced proximity to light and dissolved nutrients (Harlin, 1980). In general, epiphytism generates negative effects on algal hosts, as fitness reduction (Sand-Jensen, 1977) owing to reduced carbon uptake by diminished light penetration (Buschmann and Gómez, 1993), decrease in growth and reproduction rates (Kraberg and Norton, 2007) and increased drag effect with subsequent breakage or detachment of thalli (Arrontes, 1990). By contrast, few direct beneficial effects for hosts are known, e.g., epiphytes may protect hosts against the deleterious effects of both desiccation and excess light at low tides (Richardson, 1980). Epiphytic communities play an essential role in marine ecosystems

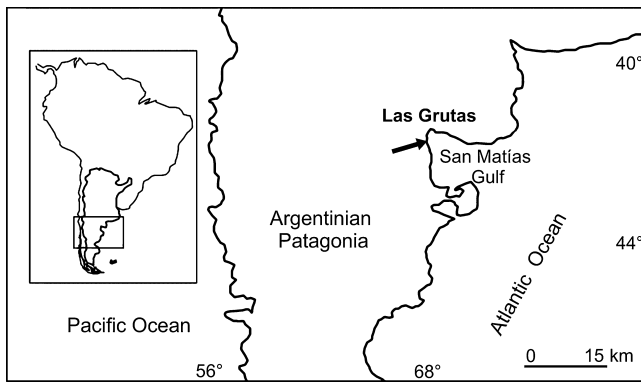
as primary producers, providing food for herbivores (Beleggratis et al., 1999). The removal of the epiphyte cover via herbivory has mostly been regarded as an indirect beneficial effect to hosts by reducing the detrimental impacts of shading and competition for nutrients (Brönmark, 1989).

Little is known about epiphyte dynamics in relation to algal hosts' peculiarities. Evidence suggests that different macroalgae do not support epiphytes in the same way, depending on factors such as life cycles, algal architecture or the possession of chemical defences (Michael et al., 2008). In addition, different epiphytic algal compositions have been attributed to variations in environmental factors, such as water motion (Kendrick and Burt, 1997), light quantity and quality (Hanelt and Roleda, 2009), grazing pressure (Borowitzka, 1981) and nutrient supply (Littler et al., 2010).

Despite the ecological importance and cosmopolitan distribution of Dictyotalean representatives, epiphytism studies on them are scarce. Only a few studies have been made in the order Dictyotales, such as in the population of *Dictyota dichotoma* (Hudson) J.V. Lamouroux (Ballantine, 1979), *Zonaria tournefortii* (J.V. Lamouroux) Montagne (Montañés et al., 2003), *Padina concrescens* Thivy (Ortuño-Aguirre and Riosmena-Rodríguez, 2007) and *Lobophora*

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**Fig. 1.** Location of the sampling site in Las Grutas beach, San Matías Gulf, Río Negro Province, Argentina.

*variegata* (J.V. Lamouroux) Womersley ex E.C. Oliveira (Ballantine, 1979; Fricke et al., 2011, 2013).

*D. dichotoma* is one of the most abundant species along the Northern Patagonian coast of Argentina. At present, it is considered a cryptogenic species; since its biogeographical history is unknown, it cannot be ascribed as native or introduced. Preliminary molecular studies would indicate a recent introduction since the Argentinian population does not diverge from the most common European haplotype (data unpublished, Dr. F. Steen, per. comm.).

There have been no previous studies of temporal variations in epiphytic communities on *D. dichotoma*, although Ballantine (1979) reported an epiphyte list in a *D. dichotoma* population from Puerto Rico. In view of this, this study aims to explore the dynamics of the epiphytic community on this population; in terms of richness, diversity and abundance; in relation to the seasonal variations of vegetative characteristics; and in different host regions under different daylengths and seawater temperatures.

## 2. Materials and methods

### 2.1. Study site

This study was performed at Las Grutas located in the NW corner of San Matías Gulf (40°82'S, 65°09'W), Argentina (Fig. 1), a coastline dominated by cliffs about 10 m high, whose intertidal substrate is an abrasive rock platform partially covered by unconsolidated sand and gravel. The shore is characterized by a moderate wave exposure and a large tidal range of up to 9 m. The climate is semi-arid with irregular rainfall distribution; seawater temperature in this region varies from 9 to 12 °C in winter and 14 to 19 °C in summer. Environmental variables, such as seawater temperature and daylength, provide the typical seasonal variation for this region. The temperature was maximal in summer–autumn, and gradually decreasing to winter–spring. The daylength was longer in spring–summer and shorter in autumn–winter.

### 2.2. Sampling design and collection of data

*D. dichotoma* co-dominates the subtidal zone in this area together with *Codium decorticatum* (Woodward) M.A. Howe, *Hymenena* spp., *Rhodomenia pseudopalmata* (J.V. Lamouroux) P.C. Silva and *Sporochnus pedunculatus* (Hudson) C. Agardh.

Isomorphic sporophytes and gametophytes of *D. dichotoma* occur simultaneously throughout the year.

Morphological variables showed temporal variation, correlated with daylength and seawater temperatures. Considering the whole *D. dichotoma* population, without discriminating reproductive stages, a seasonal marked variation in thalli length was

found (PERMANOVA: pseudo- $F=78.846$ ,  $p=0.0001$ ), with highest values in summer and spring, and lowest ones in winter. In addition, the dry mass thalli host seasonality varied (PERMANOVA: pseudo- $F=22.687$ ,  $p=0.0001$ ); the lightest thalli were registered in winter–spring and the heaviest ones in summer–autumn.

Vegetative thalli were less abundant than reproductive thalli, and sporophytes were less abundant than male and female gametophytes. In general, the male/female ratio varied between 1:2 and 1:1. Apical regions were more fertile than basal regions. Significant morphological variation was observed in thalli of both life-cycle generations (Gauna et al., 2013).

Specimens of *D. dichotoma* were collected monthly from the subtidal zone by SCUBA diving at 3 m depth, from April 2009 to March 2010. Surface seawater temperatures and daylength were measured daily in the sampling location. Each month, 60 complete attached individuals were collected randomly. Each selected frond was cut above the holdfast. All specimens were squeezed by hand to remove excess seawater and subsequently transported to the laboratory in closed plastic bags. Samples were stored overnight at 5 °C. The following day, each specimen was washed thoroughly with seawater to remove adhering sand.

In each specimen, algal portions of 12 mm<sup>2</sup> in area were randomly selected. Three different zones for epiphyte position on host thalli were considered: distal, middle and basal. Due to the apical location of growth of *D. Dichotoma* thalli, substratum age was minimal in apical regions and maximal in basal regions. In addition, for each selected individual, the reproductive stage was noted and female and male gametophytes, sporophytes and non-fertile thalli were grouped.

Thallus length and dry weight were measured monthly in 20 individuals. Dry weight was determined after drying the samples for 48 h at 70 °C.

### 2.3. Epiphyte identification and counting

Epiphytes were identified and counted using a Nikon Eclipse TE 300 microscope (Tokyo, Japan) equipped with a Nikon FDX 35 camera. Counting was normally performed using a 40× objective. For filamentous Cyanophyceae, a 100× oil immersion objective was used. Cyanobacterial filaments and non-filamentous colonies were counted as individuals, along with brown algae and single celled green algae.

To record epiphytic incidence, four groups of epiphytes were recognized: (i) very abundant (present on more than 150 thalli), (ii) abundant (75–149 thalli), (iii) rare (15–74 thalli) and (iv) occasional (less than 14 thalli).

Richness, diversity and evenness indices were calculated for each selected host area. Specific richness ( $S$ ) was the total number of epiphytic species for the examined area (sampling unit: 12 mm<sup>2</sup>). The diversity and evenness of epiphytes were calculated using Simpson's indices.

### 2.4. Epiphyte isolation and cultures

Unialgal cultures were necessary to identify Ulvophyceae epiphytes. Small portions of *D. dichotoma* thalli were sectioned and gently rinsed three times with sterile seawater. Crude cultures were initiated by placing these frond portions in Petri dishes containing Provasoli enriched seawater (PES) medium (Provasoli, 1968). Cultures were maintained at 21 ± 1 °C with a light/dark regime of 12:12 h and an irradiance of 15 μmol m<sup>-2</sup> s<sup>-1</sup>. Different germlings were obtained, subsequently transferred into unialgal cultures and maintained under the above conditions with weekly medium changes.

## 2.5. SEM method

Host thallus fragments were fixed in 0.01 M sodium cacodilate (pH 7.2) buffer, containing 2.5% glutaraldehyde at 5 °C for 2 h. They were subsequently mounted on slides covered with 0.5% poly D-lysine and dehydrated in a graded acetone series, following the protocol by Cáceres (1995). The samples were finally critical-point dried for 1 h, coated with gold and observed with a Leo Evo 40 scanning electron microscope (SEM).

## 2.6. Data analysis

To describe the epiphyte distribution patterns on *D. dichotoma*, we used the abundance, expressed as the number of individuals, of each epiphytic taxa registered, in relation to host regions and time of the year (months and seasons). The distribution and composition of the epiphyte assemblages were tested using multivariate data analysis, PRIMER (Plymouth Routines in Multivariate Ecological Research) statistical package (Clarke and Warwick, 2001). Differences between epiphytic algal classes, specific richness, diversity and abundance were tested by one-way analysis of similarities test (ANOSIM). In addition, epiphytic load on differential *D. dichotoma* reproductive stage was tested by one-way ANOSIM. Comparisons among epiphytes classes, between seasons and inside each host region were analyzed by a two-way nested ANOSIM test, with 999 permutations. These tests were made by Bray-Curtis

similarity matrix applying the square-root transformation of the data. In each ANOSIM test, the null hypothesis that there were no significant differences among groups was rejected if the significance level ( $p$ ) was  $<0.05$  (groups for host regions: distal, middle, basal; groups for season comparison: autumn, winter, spring and summer). When significant differences were detected among *a priori* groups, the  $R$ -statistic was used to determine the extent of those differences. Non-metric multidimensional scaling (nMDS) was used to produce two-dimensional ordinations. Similarity percentages (SIMPER) were used to explain which epiphytic taxa characterized each group and distinguished among each pair of groups. The significance differences in length and dry mass in *D. dichotoma* individuals among seasons were tested using one-way analysis of variance based on 9999 permutations (PEMANOVA). These tests were made in resemblance matrix using Euclidean distance with normalized data.

Biological data were related to environmental factors by means of a canonical correspondence analysis (CCA), and Pearson correlation coefficients were also estimated. The biological variables considered in the multivariate analysis were the following: Hl, host length (cm); Hdm, host dry mass (g); and Chl, Bacill, Phaeo, Rhodo, Cyano: Chlorophyceae, Bacillariophyceae, Phaeophyceae, Rhodophyceae and Cyanophyceae abundances (number of individuals). The abiotic factors considered in the CCA were:  $T$ , seawater temperature (°C) and  $D$ , length of daily light period.

**Table 1**  
Algal epiphytes from April 2009 to March 2010 at Las Grutas, Argentina.

Epiphytic Taxa	Distal		Middle		Basal	
	No. of epiphytized thalli	Incidence rate	No. of epiphytized thalli	Incidence rate	No. of epiphytized thalli	Incidence rate
<i>Calothrix fusca</i>	23	Rare	52	Rare	64	Rare
<i>Lyngbya nordgaardii</i>	18	Rare	37	Rare	24	Rare
<i>M. aeruginosa</i>	96	Frequent	165	Very frequent	137	Frequent
<i>Oscillatoria corallinae</i>	5	Occasional	2	Occasional	25	–
<i>Oscillatoria pulchra</i>	1	Occasional	22	Rare	3	Rare
<i>Pseudanabaena balatonica</i>	2	Occasional	5	Occasional	1	Occasional
<i>Spirulina</i>			1	Occasional		Occasional
<i>Peridinium</i> sp.	2	Occasional	14	Occasional	5	Occasional
<i>Proocentrum lima</i>	4	Occasional	13	Occasional	10	Occasional
“Central diatoms” group	12	Occasional	48	Rare	52	Rare
<i>Cocconeis</i> sp.	105	Frequent	113	Frequent	86	Frequent
<i>Fragilaria</i> sp.	12	Occasional	15	Rare	14	Occasional
<i>G. marina</i>	109	Frequent	132	Frequent	97	Frequent
<i>Licmophora</i> sp.	5	Occasional	19	Rare	6	Occasional
<i>Navicula</i> spp.	23	Rare	46	Rare	44	Rare
<i>Nitzschia</i> sp.	11	Occasional	24	Rare	21	Rare
<i>Pleurosigma</i> sp.	2	Occasional	1	Occasional	3	Occasional
<i>Synedra</i> sp.	5	Occasional	6	Occasional	2	Occasional
<i>A. secundata</i>	7	Occasional	64	Rare	82	Frequent
<i>Ceramium</i> sp.	–	–	4	Occasional	1	Occasional
<i>Polysiphonia</i> spp.	18	Rare	20	Rare	22	Rare
<i>Stylonema alsidii</i>	56	Rare	132	Frequent	137	Frequent
<i>U. marchantiae</i>	11	Occasional	124	Frequent	130	Frequent
<i>Acrochaete polymorpha</i>	4	Occasional	29	Rare	47	Rare
<i>Bolbocoleon piliferum</i>	10	Occasional	29	Rare	19	Rare
<i>Chaetomorpha</i> sp.	–	–	–	–	2	Occasional
<i>Pringsheimiella scutata</i>	–	–	3	Occasional	1	Occasional
<i>Ulothrix</i> sp.	–	–	2	Occasional	2	Occasional
<i>Hecatonema maculans</i>	3	Occasional	11	Occasional	22	Occasional
<i>Microspongium globosum</i>	3	Occasional	18	Rare	13	Occasional
<i>Myrionema</i> sp.	–	–	5	Occasional	5	Occasional
<i>P. tenella</i>	5	Occasional	46	Rare	73	Rare
<i>Sphacelaria cirrosa</i>	–	–	5	Occasional	7	Occasional

The number of epiphytized *D. dichotoma* fronds and incidence rate are reported.

**Table 2**

Results of the one-way ANOSIM test (host regions) showing the significance level (*p*), global *R* and *R*-statistics for the Bacillariophyceae, Cyanophyceae, Phaeophyceae and Rhodophyceae abundances, for each pair of regions.

Host regions	Bacillariophyceae		Cyanophyceae		Chlorophyceae		Phaeophyceae		Rhodophyceae	
	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>
Global	0.775	−0.031	0.002	0.181	0.001	0.341	0.001	0.231	0.001	0.277
Distal vs Middle	0.615	−0.029	0.002	0.339	0.001	0.421	0.001	0.339	0.001	0.333
Distal vs Basal	0.733	−0.039	0.002	0.219	0.001	0.453	0.002	0.320	0.001	0.413
Middle vs Basal	0.615	−0.024	0.596	−0.020	0.035	0.119	0.100	0.055	0.108	0.064

### 3. Results

#### 3.1. Epiphytic algae identified on *D. dichotoma* individuals

During the study, 32 taxa of epiphytic algae (Table 1) were found on *D. dichotoma* individuals: of these, 7 were Cyanophyceae, 9 Bacillariophyceae (including centric diatoms), 2 Dinophyceae, 4 Rhodophyceae, 6 Chlorophyceae and 4 Phaeophyceae. Based on the scale used (see Section 2), only *Microchaete aeruginea* Batters was recorded as a very abundant species (Table 1, Fig. 2A). Five taxa were abundant: *Cocconeis* sp., *Grammatophora marina* (Lyngbye) Kützing, *Stylonema alsidii* (Zanardini) K.M. Drew, *Ulvela marchantiae* (Setchell & N.L. Gardner) Nielsen, O'Kelly & Wysox and *Audouinella secundata* (Lyngbye) Dixon (Table 1, Fig. 2B–F). The remaining recorded epiphytes were either rare (Table 1, Fig. 2G–N), or occasional (Table 1).

Epiphytes belonging to the classes Chlorophyceae, Bacillariophyceae and Cyanophyceae were significantly more abundant (ANOSIM: global *R* = 0.228, *p* = 0.001) (Fig. 3A). Epiphyte representatives of all classes did not show seasonality abundance, as the ANOSIM two-way nested results [seasons (months)] indicated *R* global close to 0.

A non-regular epiphyte spatial distribution was observed. Representatives of classes Cyanophyceae, Chlorophyceae, Phaeophyceae and Rhodophyceae showed differential distribution along the host (Table 2). The main differences were observed among the distal and middle-basal regions (Fig. 3B, Table 2). The distal regions had less epiphytes than both the middle-basal ones (Fig. 3D). By contrast, Bacillariophyceae epiphytes did show differential, i.e., uniform distribution (Table 2).

Twenty-six taxa were reported in distal regions, with only three being abundant epiphytes: *M. aeruginea*, *Cocconeis* sp. and *G. marina* (Table 1). The middle and basal regions showed the highest numbers of epiphytic taxa since 32 taxa were observed in both (Table 1). The very abundant epiphyte *M. aeruginea* appeared in the middle sector together with four abundant epiphytes, *Cocconeis* sp., *G. marina*, *S. alsidii* and *U. marchantiae* (Table 1). Very abundant epiphytes were not reported in basal regions, but only those in the abundant category. Moreover, in basal regions, the number of rare species was lower than in middle and distal regions (Table 1).

#### 3.2. Specific richness, diversity and abundance of epiphytes along host

The ANOSIM results show significant differences in epiphyte-specific richness and abundance between the three host sectors (Table 3). By contrast, the epiphytic diversity among them is equivalent (Table 3).

In middle and basal regions, the values of specific richness and epiphyte abundances are higher than those found in distal regions (Table 3, Fig. 3C). The highest specific richness is registered in the middle section, with 12 epiphytic taxa as a maximum value. Nevertheless, the highest abundance is observed in basal sections

(average abundance:  $187 \pm 141.5$  individuals). In middle regions, in turn, the average abundance is  $131.7 \pm 117.7$  individuals in middle regions (Fig. 3C).

The epiphytic abundances estimated in the three regions are scattered throughout the nMDS plot, a fact that shows a clear separation among the host regions (Fig. 4). Furthermore, the SIMPER analyses show high abundances of *U. marchantiae*, *M. aeruginea*, *Pylaiella tenella* Setchell & N.L. Gardner and *A. secundata*, which characterize the basal region and clearly differentiated it from other regions (Table 4a). Similarly, lower *Cocconeis* sp. and *G. marina* abundances characterize the distal region compared with the middle and basal sections.

#### 3.3. Seasonal specific richness, diversity and abundance of epiphytes

Epiphyte-specific richness, diversity and abundance show different peaks in different seasons without discrimination between frond regions (Table 3). Specific richness is different among winter and spring. In spring, fronds support more epiphytic taxa (Fig. 3D). Epiphytic diversity has also different peaks, maximal in autumn and minimal in winter. Conversely, epiphyte abundance was higher in winter compared with all other seasons (Fig. 3D), explained by the high abundances of *G. marina*, *Cocconeis* sp. and *P. tenella* (Table 4b). The SIMPER analyses determined an important difference in autumn, accounted for by the high abundance of *S. alsidii* (Table 4b).

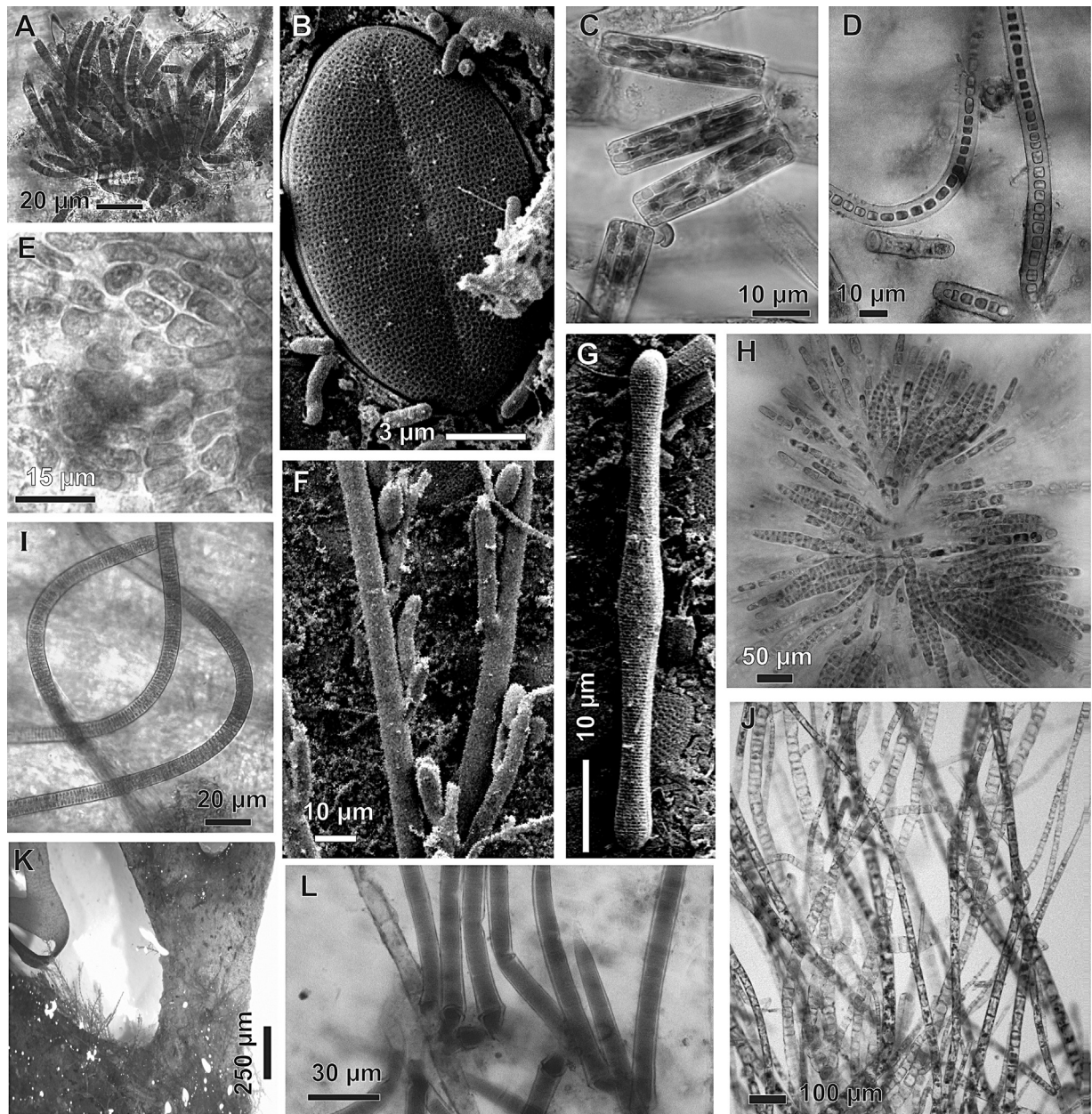
The results of the ANOSIM seasonality analyses associated with host regions were not statistically significant. Differences between seasons were not observed considering the factors of host region, specific richness, epiphyte diversity and abundance. The 2D MDS plot clearly did not distinguish epiphyte seasonality when abundances were analyzed as a function of different host regions (Fig. 4).

**Table 3**

Results of the one-way ANOSIM test (host regions and seasons) showing the significance level (*p*), global *R* and *R*-statistics for the specific richness, Shannon–Weaver diversity and abundance epiphytes for each pair of regions and seasons.

Host regions	Richness		Diversity		Abundance	
	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>	<i>P</i>	<i>R</i>
Global	0.001	0.122	0.757	−0.004	0.001	0.229
Distal vs. Middle	0.001	0.228	0.973	−0.020	0.001	0.314
Distal vs. Basal	0.001	0.151	0.677	−0.007	0.001	0.381
Middle vs. Basal	0.160	0.009	0.104	0.004	0.001	0.035
Seasons						
Global	0.002	0.014	0.031	0.007	0.001	0.01
Au vs. W	0.371	0	0.004	0.023	0.035	0.008
Au vs. Sp	0.05	0.008	0.058	0.01	0.196	0.003
Au vs. Sm	0.310	0.011	0.180	0.003	0.153	0.004
W vs. Sp	0.003	0.020	0.337	0.001	0.040	0.009
W vs. Sm	0.350	0	0.105	0.006	0.001	0.03
Sp vs. Sm	0.001	0.043	0.672	−0.003	0.037	0.008

Seasons: autumn (Au), winter (W), spring (Sp) and summer (Su).



**Fig. 2.** Light and SEM micrographs of some epiphytic species on *D. dichotoma*. (A) Very abundant species: *M. aeruginea*. (B–F) Abundant species: (B) *Cocconeis* sp. (C) *G. marina*. (D) *Stylonema alsidii*. (E) *U. marchantiae*. (F) *A. secundata*. (G–L) Rare species: (G) *Fragilaria* sp. (H) *Microspongium globosum*. (I) *Oscillatoria pulchra*. (J) *P. tenella*. (K) *Polysiphonia* sp. (L) *Calothrix fusca*.

### 3.4. Relationships of both host reproductive stages and vegetative characteristics with epiphytic load, in relation to environmental factors

Female and male gametophytes, sporophytes and non-fertile *D. dichotoma* thalli did not present statistical differences in epiphytic load (ANOSIM: global  $R = -0.001$ ,  $p = 0.706$ ).

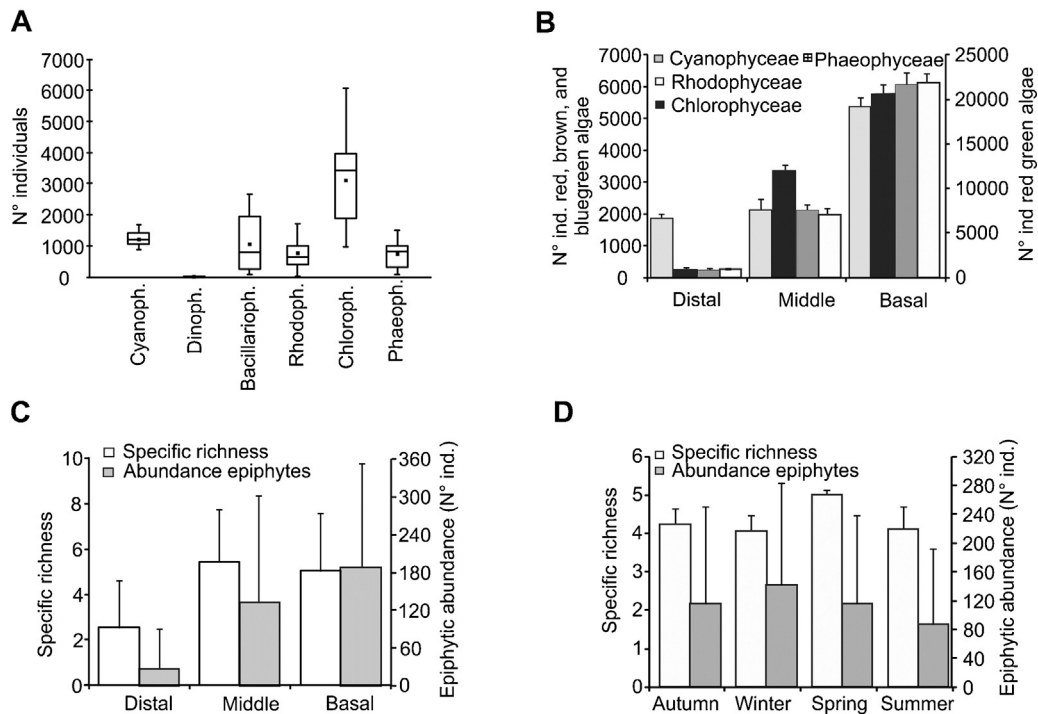
In the CCA, the first tree axes explained 83.7% of the joint variation of environmental factors and biological variables. Axis I was mainly related to the distribution of host differences between distal vs. middle and basal regions (Fig. 5). The middle and the basal sectors were located on the positive semi-plane of axis I, characterized by high abundances of Bacillariophyceae, Chlorophyceae, Phaeophyceae, Cyanophyceae and Rhodophyceae representatives.

The correlations between epiphyte load and both host vegetative characteristics and environmental parameters were not

statistically significant. Similarly, environmental variables were not correlated with epiphyte classes. High peaks in length and dry mass did not coincide with the highest epiphyte abundance registered in winter. In addition, strong correlations were observed between epiphyte abundances of four classes, i.e. Phaeophyceae epiphytes demonstrate high correlations with Cyanophyceae ( $\rho = 0.629$ ) and Chlorophyceae ( $\rho = 0.588$ ) epiphytes, and Rhodophyceae epiphytes showed strong correlations with Chlorophyceae ( $\rho = 0.821$ ) and Phaeophyceae ( $\rho = 0.681$ ) classes.

## 4. Discussion

Our study showed that thalli of *D. dichotoma* in the Northern Atlantic Patagonian coast of Argentina served as hosts for 32 algal epiphytic taxa. We found that the species *M. aeruginea*, *Cocconeis* sp., *G. marina*, *S. alsidii*, *A. secundata* and *U. marchantiae* were not



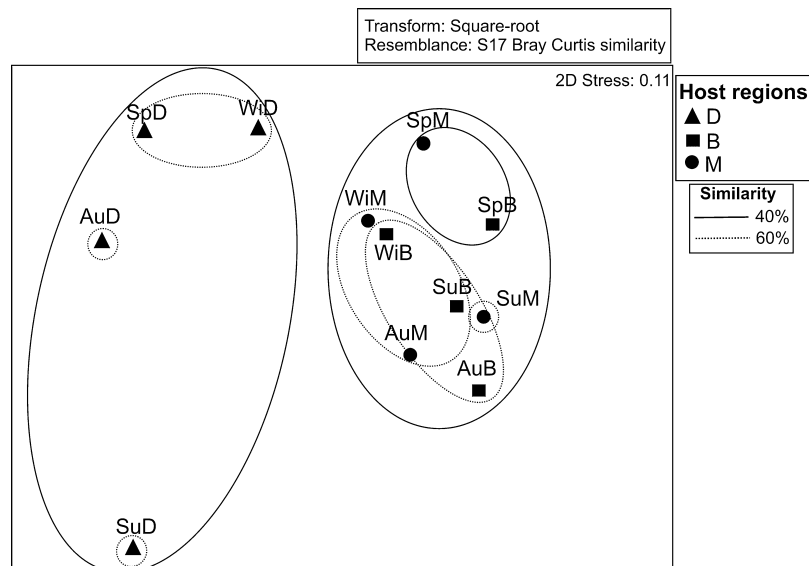
**Fig. 3.** Epiphytism of *D. dichotoma* between April 2009 and March 2010. (A) Epiphyte abundance (number of individual) of different algal groups (means  $\pm$  SE,  $n = 240$ ). (B) Cyanophyceae, Chlorophyceae, Phaeophyceae and Bacillariophyceae abundances (means  $\pm$  SE;  $n = 240$ ) in distal, middle and basal host regions. (C) Richness and epiphytic load (means  $\pm$  SE;  $n = 240$ ) on different host regions. (D) Richness and epiphytic load seasonality of *D. dichotoma*.

only the most common, but were also permanent representatives of the studied epiphytic community.

Considering the classes to which the reported epiphytes belong, they have also been found on other algal hosts, as Chlorophyceae (Nielsen, 1988), Rhodophyceae (Womersley, 1994; Aguilar-Rosas et al., 2006), Bacillariophyceae (Mendoza-González and Mateo-Cid, 1996), Phaeophyceae (Taşkin et al., 2006), Cyanophyceae (Crispino and Sant’anna, 2006) and Dinophyceae (Levasser et al., 2003), indicating they include typical members of epiphytic communities in marine environments worldwide.

The composition of the epiphytic assemblages and the patterns of distribution of the most common species showed considerable temporal variations. There were remarkable differences in abundance between winter and summer. In winter, the high abundances were mainly accounted for by *P. tenella*. In summer, the epiphytic community was dramatically reduced quantitatively.

A comparison of epiphytic diversity on *D. dichotoma* from Argentina and Puerto Rico (Ballantine, 1979) shows important differences that are most likely explained by their different geographical locations. Ballantine (1979) observed a high proportion



**Fig. 4.** nMDS plot of epiphytic load (number of individual) associated with *D. dichotoma* from different host regions and seasons. (D) Distal, (M) middle and (B) basal regions. Superimposed cluster analysis at similarity levels of 40 and 60%.

**Table 4**  
Algae epiphytic taxa characterizing assemblages in host regions (a) and seasons (b).

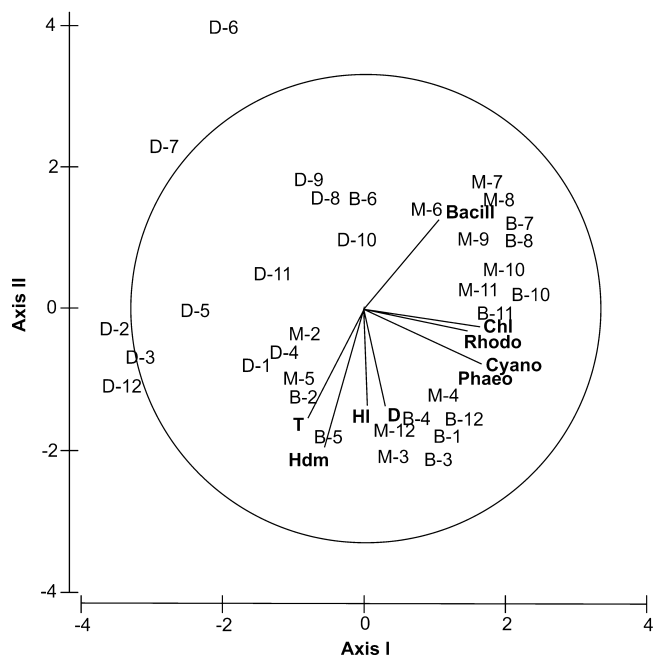
(a)				
	Distal	Middle	Basal	
Distal	<i>M. aeruginea</i> <i>G. marina</i> <i>Cocconeis</i> sp. (63.3%)			
Middle	<i>A. secundata</i> <i>U. marchantiae</i> <i>M. globosum</i>  (38.86%)	<i>U. marchantiae</i> <i>M. aeruginea</i> <i>A. polymorpha</i> <i>A. secundata</i> (72.49%)		
Basal	<i>U. marchantiae</i> <i>A. secundata</i> <i>P. tenella</i> <i>M. globosum</i> (41.20%)	<i>B. piliferum</i> <i>Polysiphonia</i> spp. <i>L. nordgaardii</i> (24.56%)	<i>U. marchantiae</i> <i>M. aeruginea</i> <i>P. tenella</i> <i>A. secundata</i> (76.44%)	
(b)				
	Autumn	Winter	Spring	Summer
Autumn	<i>U. marchantiae</i> <i>M. aeruginea</i> <i>S. alsidii</i> (69.39%)			
Winter	<i>L. nordgaardii</i> <i>H. maculans</i>  (32.38%)	<i>G. marina</i> <i>A. polymorpha</i> <i>Cocconeis</i> sp. <i>M. aeruginea</i> <i>U. marchantiae</i> <i>P. tenella</i> (76.49%)		
Spring	<i>B. piliferum</i> <i>O. pulchra</i>  (36.46%)	<i>B. piliferum</i> <i>H. maculans</i> (29.05%)	<i>M. aeruginea</i> <i>Cocconeis</i> sp. <i>B. piliferum</i> <i>G. marina</i> (70.30%)	
Summer	<i>O. pulchra</i> <i>U. marchantiae</i> <i>B. piliferum</i> (34.61%)	<i>L. nordgaardii</i> <i>U. marchantiae</i> (37.21%)	<i>L. nordgaardii</i> <i>U. marchantiae</i> (35.85%)	<i>M. aeruginea</i> <i>C. fusca</i> <i>L. nordgaardii</i> (67.02%)

Assemblages of epiphytes on *D. dichotoma* responsible for significant differences tests (SIMPER) between host regions (a) (distal, middle and basal) and (b) seasons (autumn, winter, spring and summer) are indicated between brackets.

of Rhodophycean species, mainly Ceramiales, representing 20% of total epiphytism, whereas in the present study only a few Ceramialian epiphytes were recorded.

Similarly, comparison of the epiphytic diversity on *D. dichotoma* from Argentina with those on other Dictyotales species shows clear since in *Z. tournefortii*, Ceramialian epiphytes represented 43% of total epiphytic load (Montañés et al., 2003) and in *Cystoseira tamariscifolia* (Hudson) Papenfuss, 36 Ceramialian epiphytic species were recognized, both macroalgae growing from Spain (Morales-Ayala and Viera-Rodríguez, 1989).

It is known that morphological and physiological characteristics of seaweeds hosts may be important for the settlement and recruitment of epiphytes (Rindi and Guiry, 2004). Morphologically, the fact that *D. dichotoma* has localized apical growth may explain the direct relationship between epiphytic load and frond sector age, namely that newer sectors were colonized by fewer epiphytes. In this study, we observed a clear zonation pattern on the host thalli, indicating that the abundance and the species number were decreasing towards apical parts. This has also been observed in *D. dichotoma* from Puerto Rico and another Dictyotalian genus, e.g. *Sargassum* (Ballantine, 1979; Bjaerke and Fredriksen, 2003). We consider that the epiphyte absences in younger thallus parts may well be due to a time lag in colonization. The lesser colonized meristematic regions (host distal regions) have not been exposed to the environment as long as the older portions and have had less time to develop an



**Fig. 5.** CCA biplot: association between biological and environmental variables. HI: length of host thalli; Hdm: dry mass of host thalli; Chl: Chlorophyceae abundance; Bacill: Bacillariophyceae abundance; Phaeo: Phaeophyceae abundance; Rhodo: Rhodophyceae abundance; Cyano: Cyanophyceae abundance; T: seawater temperature and D: daylength.

epiphytic community. Additionally, the unequal epiphyte distribution may also be due to others factors as variable environmental conditions (e.g. light and hydrodynamics) in different host parts and/or differences in surface tension and roughness.

Considering physiological aspects, it is well known that *Dictyota* species produce a high variety of secondary chemical compounds, which may act as antifouling agents (Schmitt et al., 1998; Barbosa et al., 2007). Recently, two secondary metabolites were determined on *D. dichotoma* from Las Grutas (Argentina) (Siles, 2012). Possibly these compounds were responsible for the overall low epiphytism observed on this host.

The present study showed that Bacillariophyceae representatives were a very abundant epiphyte group with uniform distribution along host thalli. Host morphological attributes may favor the homogeneous composition and structure of epiphytic diatom communities (Comte and Cazaubon, 2002). But this could be explained mainly because diatoms are normally the most abundant components in epiphytic communities of marine macroalgae (Hernández-Almeida and Siqueiros-Beltrones, 2008). Adhesive strength, which is a significant ecological characteristic of diatoms (Tanaka, 1986), may be an important factor. In addition, the high *Cocconeis* sp. abundances could be due to the fact that *Cocconeis* cells are broadly oval-shaped, giving them a high tolerance to wave action predominantly near the surface. Chains of *G. marina* were also especially abundant, putatively as a result of a beneficial nutrient and light competition (Sato et al., 2009).

Studies in brown seaweeds show that epiphytic load level is higher when the host presents both major surface area and biomass because larger host thalli would expedite epiphyte attachment (Arrontes, 1990). This relationship was found in *Sargassum* species, in which biomass was directly related with *Hypnea* epiphytism (Menezes de Széchy and Faria de Sá, 2008). By contrast, our study did not observe a significant relationship between host thalli length and dry mass with epiphytic load. Complementary studies evaluating host growth rate simultaneously with epiphytic assemblage developments will be necessary to better understanding this

paradoxical result. In this way, we also could determine variations in epiphytic assemblages and the distribution patterns of the most common species in a space and time gradient.

In conclusion, the epiphytic community of *D. dichotoma* from Las Grutas (Argentina) consisted of a limited number of species, which changed abundances through the sampling period. A few epiphytes were found throughout all seasons, but most appeared temporally distributed. Bacillariophyceae, Chlorophyceae and Cyanophyceae were the most abundant groups. A clear zonation of colonization was observed on *D. dichotoma*. The younger parts presenting meristematic growth were less epiphytized than older regions. Chlorophycean, Phaeophycean, Cyanophycean and Rhodophycean epiphytes were more abundant in middle and basal host sections, but the Bacillariophycean did not show differential pattern distribution. No different epiphytic classes or epiphytic taxa showed seasonality. Instead, the total epiphytic load showed seasonality; in winter the epiphytes were more abundant. However, a direct relationship between epiphytic load and vegetative host features was not demonstrated. Additional studies incorporating other populations of *D. dichotoma* from other areas are necessary to verify whether the patterns observed can be generalized to the whole Patagonian coast.

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