



Inhibition of coral recruitment by macroalgae and cyanobacteria

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ABSTRACT: Coral recruitment is a key process in the maintenance and recovery of coral reef ecosystems. While intense competition between coral and algae is often assumed on reefs that have undergone phase shifts from coral to algal dominance, data examining the competitive interactions involved, particularly during the larval and immediate post-settlement stage, are scarce. Using a series of field and outdoor seawater table experiments, we tested the hypothesis that common species of macroalgae and cyanobacteria inhibit coral recruitment. We examined the effects of *Lyngbya* spp., *Dictyota* spp., *Lobophora variegata* (J. V. Lamouroux) Womersley, and *Chondrophycus poiteaui* (J. V. Lamouroux) Nam (formerly *Laurencia poiteaui*) on the recruitment success of *Porites astreoides* larvae. All species but *C. poiteaui* caused either recruitment inhibition or avoidance behavior in *P. astreoides* larvae, while *L. confervoides* and *D. menstrualis* significantly increased mortality rates of *P. astreoides* recruits. We also tested the effect of some of these macrophytes on larvae of the gorgonian octocoral *Briareum asbestinum*. Exposure to *Lyngbya majuscula* reduced survival and recruitment in the octocoral larvae. Our results provide evidence that algae and cyanobacteria use tactics beyond space occupation to inhibit coral recruitment. On reefs experiencing phase shifts or temporary algal blooms, the restocking of adult coral populations may be slowed due to recruitment inhibition, thereby perpetuating reduced coral cover and limiting coral community recovery.

KEY WORDS: Allelopathy · Coral–algae interactions · *Dictyota* spp. · *Lyngbya* spp. · Phase shift

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INTRODUCTION

Competition for space is an important factor structuring marine benthic communities, particularly on coral reefs (e.g. Porter 1974, Jackson & Buss 1975). Historically, studies investigating competition for space on coral reefs focused on interactions among invertebrates (Jackson & Buss 1975, Benayahu & Loya 1981, Porter & Targett 1988), as macroalgae were inconspicuous members of most coral reef communities prior to the 1980s (Dahl 1974). Since then, coral reef degradation has occurred on a global scale (Hoegh-Guldberg 1999, Knowlton 2001, Hughes et al. 2003), usually manifested as a marked decrease in live coral cover followed by lasting proliferation of

algae referred to as a phase shift (Hughes 1994, McCook 1999).

A combination of factors can lead to phase shifts, including widespread coral mortality, declines in herbivory (e.g. die-off of the long-spined urchin *Diadema antillarum*, overfishing of herbivorous fishes) and localized nutrient enrichment (Hughes 1994, Hunter & Evans 1995, Szmant 2002). Quantifying the consequences of phase shifts to coral recruitment, growth and competition is critical to the management and restoration of coral reefs. Although the inverse relationship between algal and coral cover on degraded reefs suggests that algae are competitively dominant over adult corals, algal proliferation usually follows coral mortality (reviewed by McCook et al. 2001). This

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may indicate that corals are competitively dominant over algae as adults, but inferior as larvae and recruits (McCook et al. 2001). Experimental evidence for this hypothesized ontogenetic shift in competitive advantage of algae over scleractinians is currently lacking.

Larval recruitment is a key process in modulating coral populations (Hughes & Tanner 2000, Hughes et al. 2003). Coral recruitment is significantly lower in areas that have undergone phase shifts (Edmunds & Carpenter 2001); however, the mechanisms involved remain unknown. Some studies have shown that filamentous algae or 'algal turfs' can negatively affect survival and growth of juvenile corals (Birkeland 1977, Van Moorsel 1985). Recent experimental studies found that the cyanobacterium *Lyngbya majuscula* (Kuffner & Paul 2004) and certain algal turf assemblages (Birrell et al. 2005) can inhibit coral recruitment. Quantifying these effects and identifying the mechanisms involved during these interactions are important steps in investigating the cause and effect relationships responsible for the continuing trend of macroalgal dominance on many reefs today.

In the Florida Keys, coral cover declined from 10.3 to 6.4% cover (a 38% relative decline) between 1996 and 2000 (Porter et al. 2002). During this same period, the brown macroalgae *Dictyota* spp. dominated the benthos at depths between 0 and 25 m on many of these reefs, covering up to 56% of the bottom during the summer months (Lirman & Biber 2000, Beach et al. 2003). *Dictyota* spp. are now a dominant component of the benthos throughout the Caribbean (Rogers et al. 1997, Williams & Polunin 2001, Edmunds 2002). In addition, benthic cyanobacteria are occasionally abundant on reefs, periodically blooming on reefs in Florida (Paul et al. 2005), multiple reefs in the Caribbean (Ritson-Williams et al. 2005), Guam (Nagle & Paul 1998, Thacker & Paul 2001) and Australia (Dennison et al. 1999); in the latter 2 locations they regularly cause considerable health and economic problems. Cyanobacteria have different ecological roles than macroalgae; because they can fix atmospheric nitrogen, their growth may be limited by other nutrients, such as phosphorus or iron (Paerl 1990, Fong et al. 1993, Kuffner & Paul 2001), and their modes of reproduction and dispersal differ from those of macroalgae.

Because of their proven chemical deterrence to generalist herbivores (Paul et al. 2001) and their propensity to become spatially dominant on coral reefs around the world, we tested *Dictyota* spp., *Lyngbya* spp., *Lobophora variegata* (J. V. Lamouroux) Womersley and *Chondrophycus poiteaui* (J. V. Lamouroux) Nam (formerly *Laurencia poiteaui*) for their effects on coral recruitment and spat survival using manipulative field and outdoor seawater table experiments. We conducted our study in the Florida Keys because the decline in coral cover and predominance of macroalgae make it

an appropriate location for investigating the potential inhibition of coral recruitment on algae-dominated coral reefs. We hypothesized that exposure of coral larvae to the selected algal and cyanobacterial species herein collectively referred to as 'macrophytes' increases larval mortality, alters larval behavior (e.g. location of recruitment), inhibits coral recruitment and/or reduces recruit survival. Our controlled, manipulative experiments showed that these locally and regionally common species of macroalgae and cyanobacteria inhibited successful recruitment of the scleractinian *Porites astreoides* and the octocoral *Briareum asbestinum*, 2 coral species that are widely distributed throughout the Caribbean.

MATERIALS AND METHODS

The study, based at the Keys Marine Laboratory, Long Key, Florida, was conducted from May 11 to June 9, 2004 and May 7 to 31, 2005. We examined the effect of several species of algae and cyanobacteria on larval recruitment and recruit survival of the common reef coral *Porites astreoides* and on the larval settlement of the gorgonian *Briareum asbestinum*. These coral species were chosen because of their abundance on the reefs in Florida and for the availability of their larvae.

We conducted quantitative abundance and biomass surveys of algae and cyanobacteria at 3 inshore reef areas: '11-Foot Mound' (24° 43.381' N, 80° 51.696' W), 'Coral Gardens' (24° 50.229' N, 80° 43.769' W), and the 'Tennessee Light CARICOMP' site (24° 45.142' N, 80° 45.696' W). To estimate percent cover of algae and benthic invertebrates and the available space that coral larvae could potentially colonize, we placed a 25 × 25 cm grid (a quadrat strung with line to produce 25 boxes) on the substrate in selected locations. Number of fin kicks and compass headings were randomly generated using a random numbers table, and percent cover was visually estimated counting the number of boxes (a quarter of a box = 1% cover) occupied by each species of algae. Suitable settlement substrate (SSS) was defined here as hard substrate with open patches of crustose coralline algae (known settlement cue for some coral larvae; Morse et al. 1988, Heyward & Negri 1999) devoid of macroalgae, dense turf algae, invertebrates or sediments >1 mm thick. Measurement of this benthic category was intended as an index of potential habitat for larval recruitment. Biomass surveys were conducted by haphazardly placing 50 × 50 cm quadrats on the reef, harvesting all macrophytes within the plot, separating the samples by species, and determining wet and dry (60°C for 48 h) weights.

We collected 40 colonies of the brooding coral *Porites astreoides* from on and near the pilings of the

'Seven-Mile Bridge', transported these to the Keys Marine Laboratory in coolers, and maintained them in running seawater. Larvae were collected during the nights of May 14 to 20, 2004 (new moon = May 19) and May 8 to 14, 2005 (new moon = May 8). After enough larvae had been gathered ($\approx 25\,000$) to complete the experiments, the colonies were returned to the sites of collection and re-attached to the reef with Z-Spar® A-788 Splash Zone Compound underwater epoxy. To obtain larvae, each colony was placed in an individual 3 l Rubbermaid® 'Grip 'N Mix'™ bowl constantly supplied with running seawater. The bowls were tilted such that the positively buoyant larvae spilled over the handles of the bowls into plastic beakers fitted with a 180 μm (larvae were ≈ 1 mm diam.) mesh bottom supported 3 cm off the tank floor by attached silicone stoppers. The water level inside the tank was kept at approximately 15 cm so that the larvae remained in the beaker traps until they were collected each morning shortly after sunrise. Larvae were pooled into a common container to be subsampled for the experiments.

Porites astreoides larvae were subjected to control and treatment conditions inside customized larval recruitment chambers (Fig. 1). The chambers were deployed in the field, supplying coral larvae with access to suitable settling habitat (4.5 \times 4.5 \times 1 cm sections of Sunshine Pavers® wire-cut terracotta tiles conditioned at the Tennessee Light CARICOMP site for 5 wk) in a contained environment, while allowing water circulation via the 180 μm mesh sides, and solar irradiance through the clear, extruded acrylic tubing. The 3.2 mm thick, 10.2 cm outer diameter tubing was sliced into 12.7 cm sections to yield 90 cylindrical chambers fitted with nylon bolts for mounting the settlement tiles. Chambers were deployed at least 1 m apart, and attached to Dri-Dek® Tiles (rubber mats) with nylon bolts and nuts in order to anchor the chambers upright and parallel to the prevailing currents (Fig. 1).

The relative level of water motion inside the chambers was measured using Life Savers® (wint-o-green® flavor, individually wrapped variety) candies (Koehl & Alberte 1988). The candies were cable-tied to the nylon posts inside the chambers where the tiles would normally be attached, and to posts that were attached directly to rubber mats without being housed inside a chamber. The mats and mat + chamber assemblies ($n = 5$) were sequentially deployed at the experimental

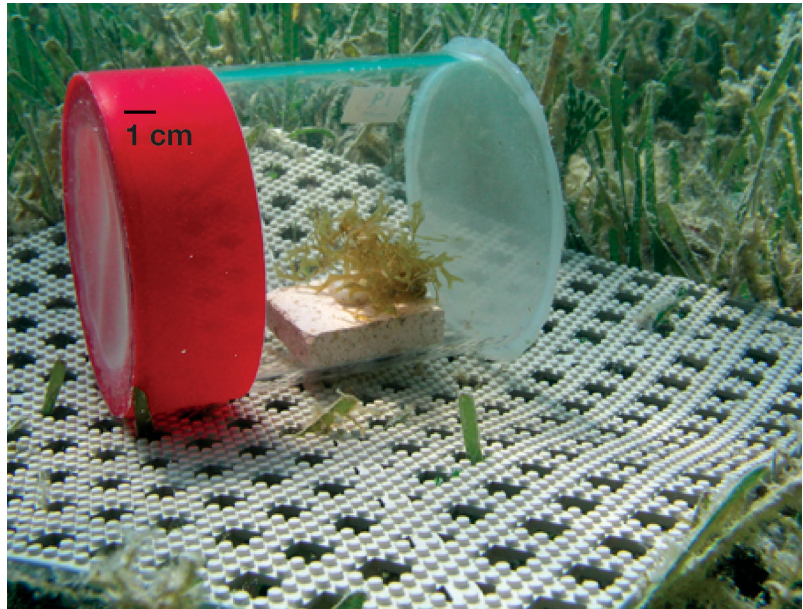


Fig. 1. Larval recruitment chamber (10.2 cm diam. \times 12.7 cm long) with attached stabilizing mat deployed at 1.5 m depth. Mesh size = 180 μm

site in a randomized order and left in the water for the candy to dissolve for exactly 10 min each. A third treatment ($n = 5$) included mats-only assemblies placed in a still bucket filled with seawater for 10 min to measure dissolution rate in the total absence of water motion. The experiment was conducted twice, on May 19 and 20, 2005. The candies were weighed before and after deployment (dried at 60°C for at least 24 h) to calculate mass (g) lost during deployment. In addition to the candy experiments, nontoxic dye was injected into chambers in the field and in a flume to visualize water moving through the mesh on both ends of the chamber.

Hypothesis: macrophytes affect coral recruitment. To test the hypothesis that selected species of macroalgae and benthic cyanobacteria can inhibit coral recruitment, we conducted 3 experiments using algae collected from the 3 sites from which biomass and percent cover data were gathered. From May 19 to 23, 2004 (Expt 1), larvae were subjected to 4 treatments: control (settlement tile with plastic clip), macrophyte mimic (seawater-conditioned plastic aquarium plant), *Dictyota pulchella* Hörnig & Schnetter, or *Lobophora variegata* (decumbent form) attached to the tile by plastic clips (thin cross sections of PVC pipe split to form a 'c' clamp). The plastic aquarium plant was chosen as a mimic because its size, shape and color were comparable to those of the macrophytes tested. From May 21 to 25, 2004 (Expt 2), larvae were subjected to 5 treatments: control, macrophyte mimic, *D. pinnatifida* Kützinger, *Lyngbya polychroa* (Meneghini) Rabenhorst, or *Chondrophycus poiteaui* attached to the tile by cable ties. For both Expts 1 and 2,

there were $n = 10$ replicate chambers per treatment, and each chamber contained 100 larvae. Clumps of algae (roughly 3 cm^3 volume) were attached with the plastic clips such that the thalli covered half of the upward-facing space on the tile. Due to the cylindrical shape of the chamber, the settlement tile did not lie flush with the bottom, allowing larvae easy access to all 6 sides for settlement. Because of foul weather on the reef tract, we placed the chambers in a hard bottom and sparse seagrass community in Florida Bay, directly behind the Keys Marine Laboratory at 1.5 to 2 m depth. After 4 d, we brought the chambers into the laboratory for analysis. Water inside the chambers was carefully sieved to count larvae still swimming, and the tiles and chambers were inspected under a dissecting microscope to record the number and location of recruits. Total survival was defined as swimming larvae plus live recruits. A recruit was defined as a coral larva that underwent complete metamorphosis, including formation of a calcium carbonate calyx.

Expt 3 was conducted in an outdoor sea table from May 22 to 25, 2004 using the plastic beakers ($n = 6$ per treatment) that were used as traps during larval collection. *Porites astreoides* larvae were subjected to 4 treatments: control, macrophyte mimic, *Dictyota pinnatifida* and *Lyngbya confervoides* C. Agardh. Conditioned settlement tiles were placed on the mesh in each beaker as a substrate for recruitment. Despite lying directly on the mesh, larvae could still easily access the bottom of the tile. The experiment was shaded with neutral density mesh to approximate irradiance levels at 1 to 3 m depth. Instead of counting the larvae upon placement into the beakers, 50 ml aliquots of water containing the larvae were prepared while constantly stirring the mixture and delivered to each beaker. Because we did not know the actual number of larvae that were placed in each beaker, % survival or % recruitment could not be known, so we assumed a normal distribution in number of starting larvae and analyzed counts instead of percents. Beakers were placed in a running seawater table with the water level adjusted to approximately 10 cm deep. Positions of the beakers within the flume were shifted each day to minimize positional effects.

Recruitment data for all 3 *Porites astreoides* recruitment experiments were analyzed using 1-way ANOVA with treatment as the fixed factor, and 2-way ANOVA with treatment and position of recruits as the 2 fixed factors. We used *a priori* contrasts (Student's *t*-tests) of the treatments and the appropriate controls to test explicit hypotheses. When the raw data failed to meet parametric assumptions, they were transformed as specified in 'Results'.

Hypothesis: macrophytes affect recruit survival. Settlement tiles containing live coral recruits from the

field experiments were used to test the effects of algal and cyanobacterial contact on recruit survival from May 28 to June 2, 2004 (Expt 4) and May 23 to 28, 2005 (Expt 5). Tiles were haphazardly selected and prepared by removing all but 1 recruit from the top of the tile, and attaching by cable tie either a live algal thallus, a macrophyte mimic (plastic flagging tape), a cable tie only (cable tie control), or nothing (control). In experiment 4, the live macrophytes tested were *Dictyota menstrualis* (Hoyt) Schmitter, *D.* and *Lyngbya confervoides*. In experiment 5, we tested *Chondrophyucus poiteaui* and *Lobophora variegata*. The algae were attached so that the thallus was in direct but loose contact with the recruit. Tiles ($n = 12$ for experiment 4 and $n = 14$ for experiment 5) were placed in a running seawater table approximately 50 cm deep and haphazardly arranged. Shade cloth was placed over the tank to reduce light intensity to levels at 1 to 3 m depth. Recruit survival data were analyzed using contingency analyses and Fisher exact tests.

Hypothesis: macrophytes affect gorgonian settlement. *Briareum asbestinum* larvae were obtained by suctioning the surface-brooded larvae off reproductive colonies in the field with a wide-mouth syringe. Expt 6 was conducted from May 31 to June 9, 2004 using the same field chambers as in Expts 1 and 2 ($n = 10$ per treatment except where noted) and deployed at the same location. Branches of dead gorgonian skeleton were presented as substrate material in lieu of terracotta settlement tiles, which were cable-tied to the nylon bolt; 100 larvae were placed in each chamber. Larvae were subjected to 7 treatments, 6 of which included a piece of gorgonian skeleton (required for metamorphosis cue, M. A. Coffroth pers. comm.): negative control (no skeleton), positive control (gorgonian skeleton only), macrophyte mimic (plastic plant), *Dictyota pinnatifida*, *D. pulchella*, *Chondrophyucus poiteaui* and *Lyngbya majuscula* (Dillwyn) Harvey. Because we did not observe complete metamorphosis of the larvae into polyps with bipinnate tentacles, we considered firm attachment to the substrate as settlement rather than recruitment. Larval survival and settlement data were analyzed using 1-way ANOVA and *a priori* contrasts.

RESULTS

Field surveys showed that *Dictyota* spp. were the most abundant taxa in 2 of the reef areas, covering 39 and 48% of the benthos at 11-Foot Mound and Tennessee Light, respectively (Fig. 2). Cyanobacteria were generally scarce (<3% cover), and suitable settlement substrate was available at all sites but was only abundant at Coral Gardens (31%, Fig. 2). For

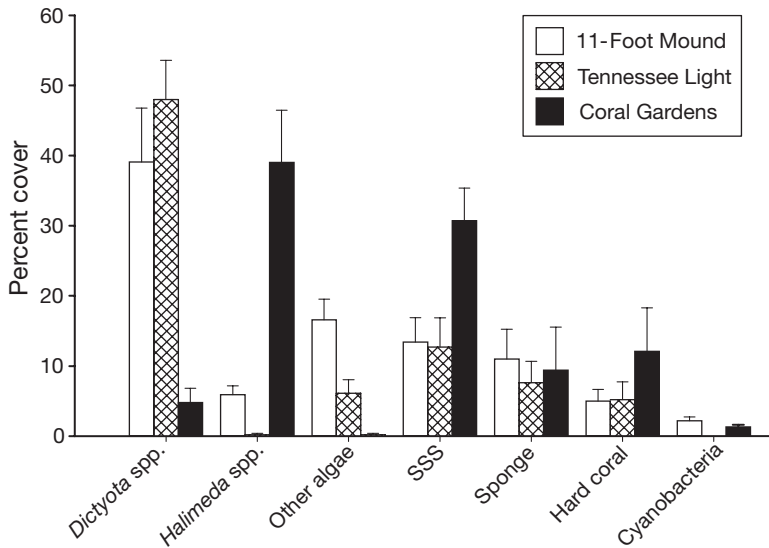


Fig. 2. Mean (+1 SE) percent cover of benthic groups at 3 inshore reef sites off Long Key, Florida Keys. SSS: suitable settlement substrate, defined as hard substrate with patches of crustose coralline algae, devoid of macroalgae, dense turf algae, invertebrates, or sediments >1 mm thick

fleshy algae, the biomass plots were dominated by *D. menstrualis* at Coral Gardens (70 ± 25 g m⁻² dry wt.), and *D. pulchella* at 11-Foot Mound and Tennessee Light (27 ± 11 and 74 ± 25 g m⁻² dry wt., respectively, Table 1).

The results of the water flow experiments showed that the dissolution of Life Savers® candies was significantly greater in both field treatments (inside and outside of chambers) compared to the still bucket treatment, and there was no significant difference in dissolution inside vs. outside the chambers (2-way ANOVA significant interaction between run and treatment $p = 0.004$, Tukey HSD post-hoc multiple comparison of treatments $\alpha = 0.05$). Evidence of the significant interaction term was seen in the difference between the field treatments and the still bucket treatment being greater during the first run when conditions were windier, but the difference between the 2 field treatments was similar in both runs. The dye injection experiments revealed that water could easily move through the mesh, and oscillatory flow generated by wavelets was observed as the dye was sucked back and forth through the chamber.

Hypothesis: macrophytes affect coral recruitment

Algae did not affect larval survival in Expt 1 (Fig. 3A; 1-way ANOVA, $p = 0.390$). Recruitment, however, was significantly less in the presence of both algal species compared to the macrophyte mimic (1-way ANOVA on square-transformed data, $p = 0.013$, *a priori* contrasts in Fig. 3B), and there was no significant difference between the macrophyte mimic and control ($p < 0.584$). More larvae recruited to the tile compared to the chamber sides in the control and in the treatment with *Dictyota pulchella* (position \times treatment interaction: $p = 0.0124$, *a priori* contrasts in Fig. 3C). Larvae that recruited to the tile recruited proportionally more to the bottom in the control treatment (2-way ANOVA on rank-transformed data, position \times treatment interaction: $p = 0.007$, *a priori*

Table 1. Taxonomic identity and biomass (g m⁻² dry wt, mean \pm SE) of macrophytes at 3 inshore reef sites off Long Key, Florida Keys. For 11-Foot Mound $n = 12$, Tennessee Light $n = 6$, Coral Gardens $n = 6$

Species	11-Foot Mound	Tennessee Light	Coral Gardens
<i>Amphiroa rigida</i>	0.47 \pm 0.28	0.01 \pm 0.01	
<i>Avrainvillea</i> sp.	0.03 \pm 0.03		
<i>Botryocladia</i> sp.	0.03 \pm 0.03		
<i>Chondrophycus poiteaui</i>	3.02 \pm 0.89		
<i>Cladophoropsis membranacea</i>	58.33 \pm 26.39		
Crustose coralline algae	0.20 \pm 0.20		
<i>Dasycladus</i> sp.	0.10 \pm 0.10		
<i>Dictyota caribaea</i>	1.33 \pm 1.20		
<i>Dictyota menstrualis</i>	0.17 \pm 0.17	0.22 \pm 0.13	70.27 \pm 10.81
<i>Dictyota pinnatifida</i>	3.83 \pm 3.21		
<i>Dictyota pulchella</i>	26.80 \pm 11.15	74.13 \pm 25.18	0.67 \pm 0.67
<i>Galaxura rugosa</i>	13.35 \pm 3.86	0.31 \pm 0.19	
<i>Gelidiella</i> sp.	1.07 \pm 0.79		
<i>Halimeda goreau</i>	3.57 \pm 2.52	54.24 \pm 45.23	
<i>Halimeda opuntia</i>	129.93 \pm 67.23		811.93 \pm 270.73
<i>Halimeda tuna</i>	52.63 \pm 16.94		545.00 \pm 55.49
<i>Hypnea</i> sp.	2.90 \pm 1.37	0.65 \pm 0.17	0.33 \pm 0.33
<i>Jania</i> sp.	3.37 \pm 2.30	1.73 \pm 0.76	
<i>Liagora</i> sp.	0.40 \pm 0.40		
<i>Lobophora variegata</i>	0.25 \pm 0.20	1.03 \pm 0.44	
<i>Neomersia annulata</i>	0.02 \pm 0.01	0.01 \pm 0.01	
<i>Penicillus capitatus</i>	0.40 \pm 0.29		
<i>Penicillus pyriformis</i>	4.33 \pm 3.98		
<i>Rhipocephalus</i> sp.	0.20 \pm 0.20		
<i>Sargassum</i> sp.	3.49 \pm 1.49	0.02 \pm 0.02	
<i>Udotea cyathiformis</i>	4.27 \pm 1.89	1.07 \pm 0.92	
<i>Udotea flabellum</i>	0.07 \pm 0.07		
<i>Valonia</i> sp.			0.93 \pm 0.86
Cyanobacteria	0.10 \pm 0.10		

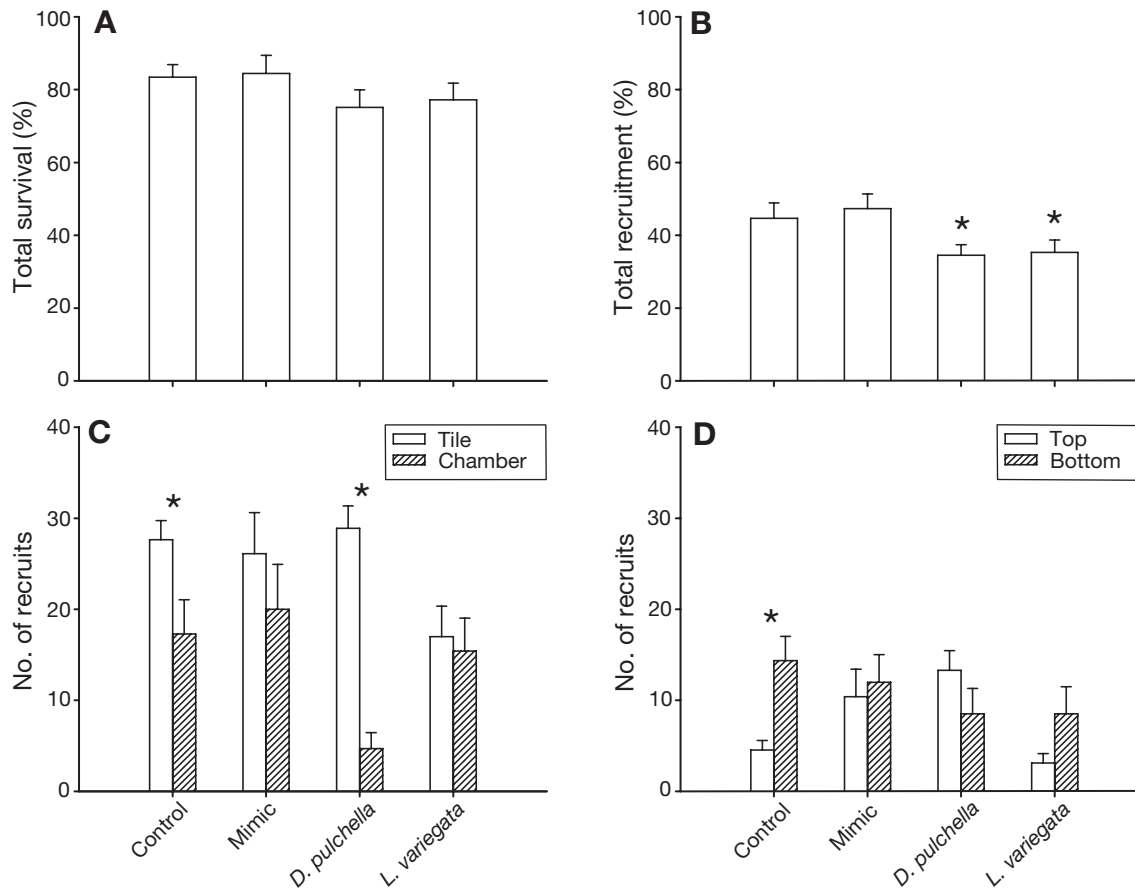


Fig. 3. *Porites astreoides*. Expt 1. Test of hypothesis that macrophytes affect recruitment, showing mean (+1 SE) percent of larvae in chambers ($n = 10$) that (A) survived (swimmers + recruits), (B) recruited (*: significant difference [$p < 0.05$] between macrophyte mimic and *Dictyota pulchella* or *Lobophora variegata* treatment), (C) recruited to tile vs. chamber sides (*: significant difference [control $p = 0.032$, *D. pulchella* $p < 0.0001$] between tile and chamber sides), and (D) recruited on top vs. bottom of tile (*: significant difference [$p = 0.004$] between top and bottom)

contrast in Fig. 3D) and equally to top and bottom of the tiles in the other 3 treatments.

The presence of algae or cyanobacteria did not affect larval survival or total recruitment in Expt 2 (Fig. 4A,B; 1-way ANOVA $p = 0.460$, $p = 0.059$, respectively). However, we found that significantly more larvae recruited onto the tile than onto the chamber in the control treatment, while the opposite was true in the *Lyngbya polychroa* treatment (2-way ANOVA on rank-transformed data, position \times treatment interaction: $p < 0.0001$, *a priori* contrast in Fig. 4C). When only recruits on the tile were considered, there was proportionally more recruitment to the bottom of the tiles, overall (2-way ANOVA on rank-transformed data, position: $p < 0.0001$, treatment: $p = 0.004$, treatment \times position: $p = 0.223$). The proportion of larvae recruiting onto the top of the tile out of total recruitment onto the tile was significantly lower in the presence of *L. polychroa* compared to the macrophyte mimic (1-way ANOVA on arcsine-transformed proportions: $p = 0.006$, *a priori* contrast: $p = 0.001$; Fig. 4D).

The presence of macrophytes did not significantly affect the total number of live larvae (Fig. 5A, 1-way ANOVA, $p = 0.078$) or total recruitment (Fig. 5B, 1-way ANOVA, $p = 0.110$) in Expt 3. We found no larvae recruited to the beaker sides or mesh. Despite an overall pattern of more recruitment to tile bottoms (Fig. 5C), a greater proportion of larvae recruited to the top of the tile in the macrophyte mimic compared to treatments with *Dictyota pinnatifida*, *Lyngbya confervoides* and in the control (1-way ANOVA: $p = 0.001$, *a priori* contrasts in Fig. 5D).

Hypothesis: macrophytes affect recruit survival

Recruit survival was significantly affected by the presence of the macrophytes tested in Expt 4 (contingency analysis Pearson chi square $p < 0.0001$). All 12 replicate recruits survived in the control and cable tie treatments, and 2 died in the macrophyte mimic treatment. Only 2 recruits survived with *Dictyota*

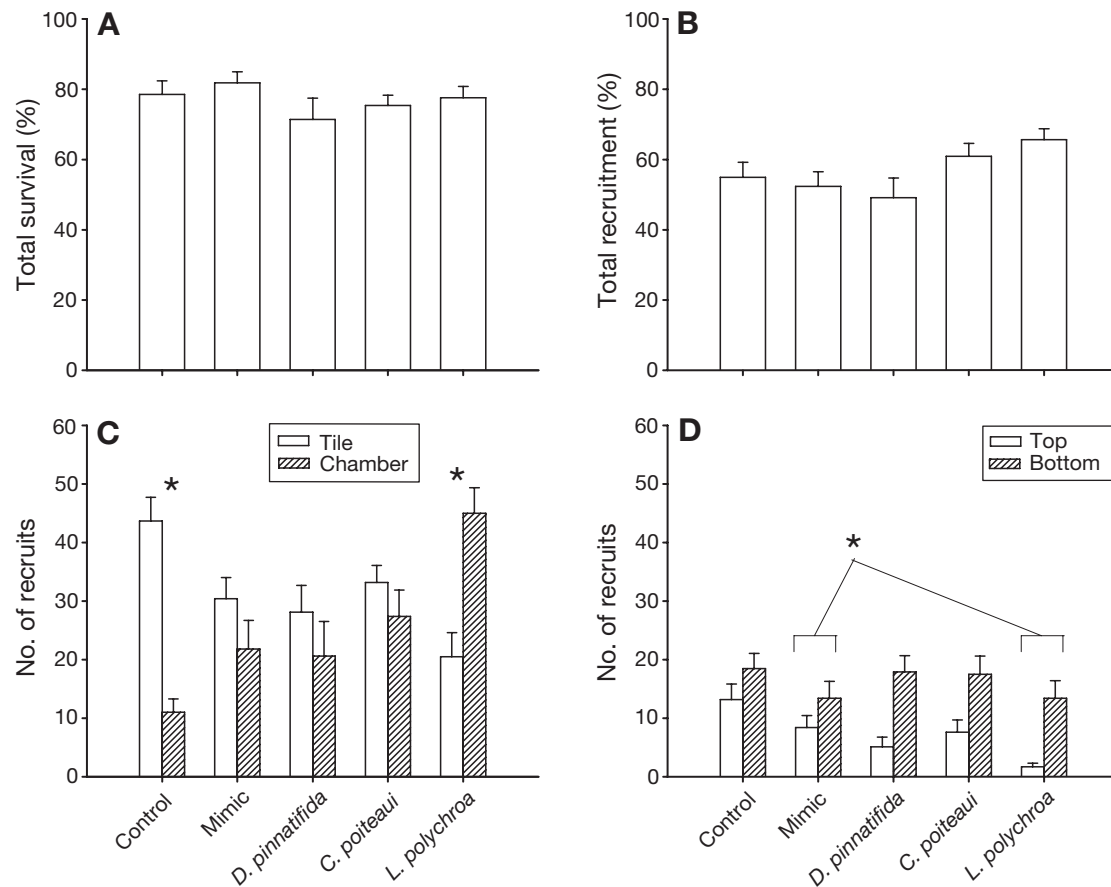


Fig. 4. *Porites astreoides*. Expt 2. Test of hypothesis that macrophytes affect recruitment, showing mean (+1 SE) percent of larvae in chambers (n = 10) that (A) survived (swimmers + recruits), (B) recruited, (C) recruited to tile vs. chamber sides (*: significant difference [$p < 0.05$] between tile and chamber sides), and (D) recruited to top vs. bottom of tile (*: significant difference [$p = 0.001$] in proportion of larvae recruiting on top of tile). *L. polychroa*: *Lyngbya polychroa*; *C. poiteaui*: *Chondrophycus poiteaui*; *D. pinnatifida*: *Dictyota pinnatifida*

menstrualis (significantly lower than macrophyte mimic, Fisher exact test, $p = 0.003$) and none survived in the *Lyngbya confervoides* treatment ($p < 0.0001$). Survival was also low (4 survivors) with *D. pulchella* (compared to the macrophyte mimic, Fisher exact test, $p = 0.036$). All comparisons except *D. pulchella* satisfy a Bonferroni adjustment for multiple tests. In Expt 5, recruit survival was not significantly affected by treatment (contingency analysis Pearson chi square $p = 0.86$); 1 recruit out of 14 died in the cable-tie control and macrophyte mimic treatments (plain tile control was eliminated in this experiment), and 2 out of 14 died in each of the *Chondrophycus poiteaui* or *Lobophora variegata* treatments.

Hypothesis: macrophytes affect gorgonian settlement

Overall survival and settlement were much lower for *Briareum asbestinum* than for *Porites astreoides*, with an overall survival rate of $25.3 \pm 1.2\%$ and $2.0 \pm 0.3\%$

successfully settling. However, survival was significantly higher in control chambers than in chambers with gorgonian skeleton only, and significantly lower in *Lyngbya majuscula* chambers compared to the macrophyte mimic (1-way ANOVA: $p < 0.001$, *a priori* contrasts in Fig. 6A). Settlement was significantly higher when gorgonian skeleton was present than in the negative control, and significantly lower in the presence of *L. majuscula* than in the macrophyte mimic (1-way ANOVA on rank-transformed data: $p = 0.024$, *a priori* contrasts in Fig. 6B).

DISCUSSION

We have presented evidence that locally common species of algae and cyanobacteria deter coral larvae from recruiting nearby despite the availability of suitable settlement substrate; most of the species we tested affected the location of coral recruitment. Our macrophyte mimics had positive effects on recruitment (probably due

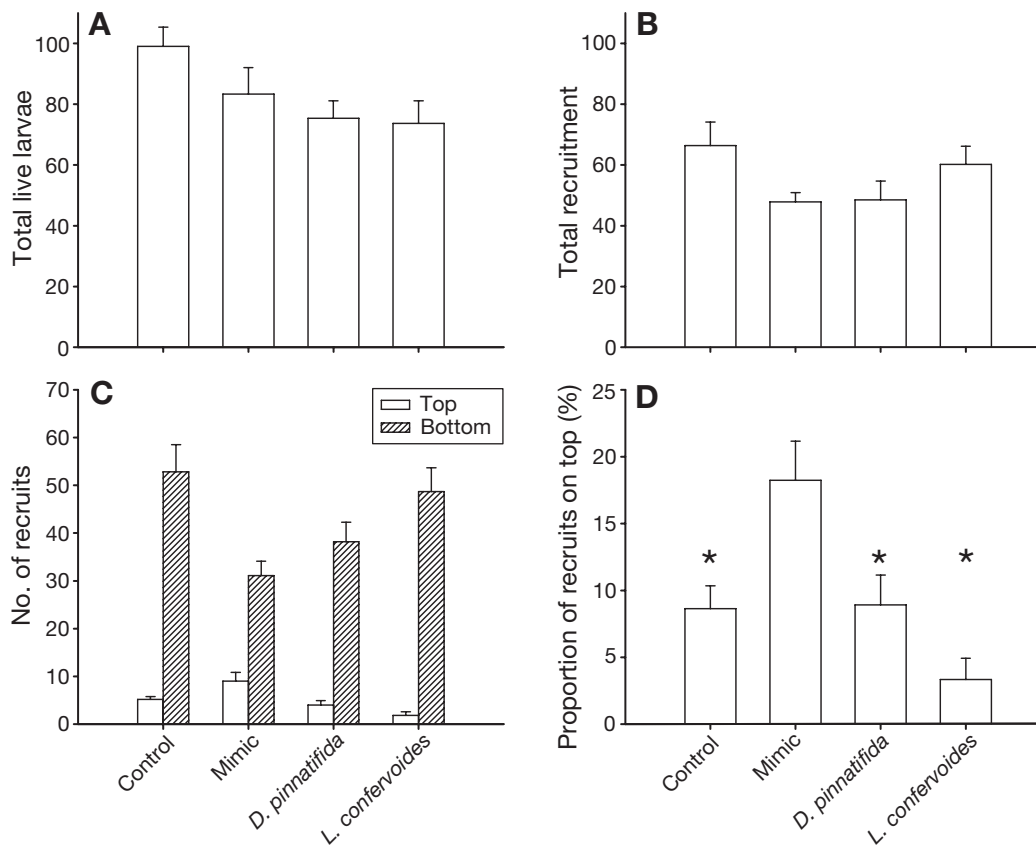


Fig. 5. *Porites astreoides*. Expt 3. Test of hypothesis that macrophytes affect recruitment, showing mean (+1 SE) number of larvae in mesh-bottomed beakers ($n = 6$) that (A) survived (swimmers + recruits), (B) recruited, and (C) recruited to top vs. bottom of tile. (D) Percent of total recruits found on top of tile (*: significant difference between macrophyte mimic and the control [$p = 0.006$], *Dictyota pinnatifida* [$p = 0.007$], or *Lyngbya confervoides* [$p = 0.0001$] treatment)

to shading and creation of cryptic habitat), and selected species of live macrophytes had negative effects that counteracted the positive effects created by an algal canopy. Furthermore, some of these macrophytes caused increased mortality rates when in contact with coral recruits. Our results show that these macroalgae and cyanobacteria can inhibit coral recruitment beyond actual space occupation, presenting a mechanism by which abundant macrophytes can perpetuate phase shifts by reducing local coral recruitment.

Porites astreoides larvae were deterred from recruiting to the tile by the presence of *Dictyota*, *Lyngbya* and *Lobophora* species. We observed species-specific effects on multiple components of recruitment including total number of recruits, recruitment location and recruit survival. *D. pulchella* and *Lobophora variegata* decreased overall recruitment compared to the macrophyte mimic, while *D. pinnatifida*, *Lyngbya polychroa* and *L. confervoides* affected the location of recruitment, suggesting that larvae avoid these species. Since benthic cover by *Dictyota* spp. approaches 50% on many reefs in the Florida Keys, the potential for these algae to reduce coral recruitment is considerable.

In a previous study examining the effects of cyanobacteria on scleractinian coral recruitment in Guam, Kuffner & Paul (2004) found that the presence of *Lyngbya majuscula* had a negative effect on larval survival for *Acropora surculosa* (broadcast spawner) and recruitment of *Pocillopora damicornis* (brooder). They also observed an avoidance behavior in the brooding species similar to that we observed in *Porites astreoides*. In our study, *L. majuscula* reduced survival and settlement of *Briareum asbestinum* larvae, and both *L. polychroa* and *L. confervoides* affected the recruitment location of *P. astreoides*, but not larval survival or total recruitment. The species-specific effects revealed in this study illustrate the complexity of algae–coral interactions and their potential impact on community dynamics.

Larvae are known to respond to both positive chemical cues associated with crustose coralline algae (Morse et al. 1988, Heyward & Negri 1999) and negative physical cues such as sediment (Hodgson 1990, Babcock & Davies 1991, Gilmour 1999) and ultraviolet radiation (Kuffner 2001, Gleason et al. 2006). Our results suggest that *Porites astreoides* larvae may use certain macroalgae and cyanobacteria as negative

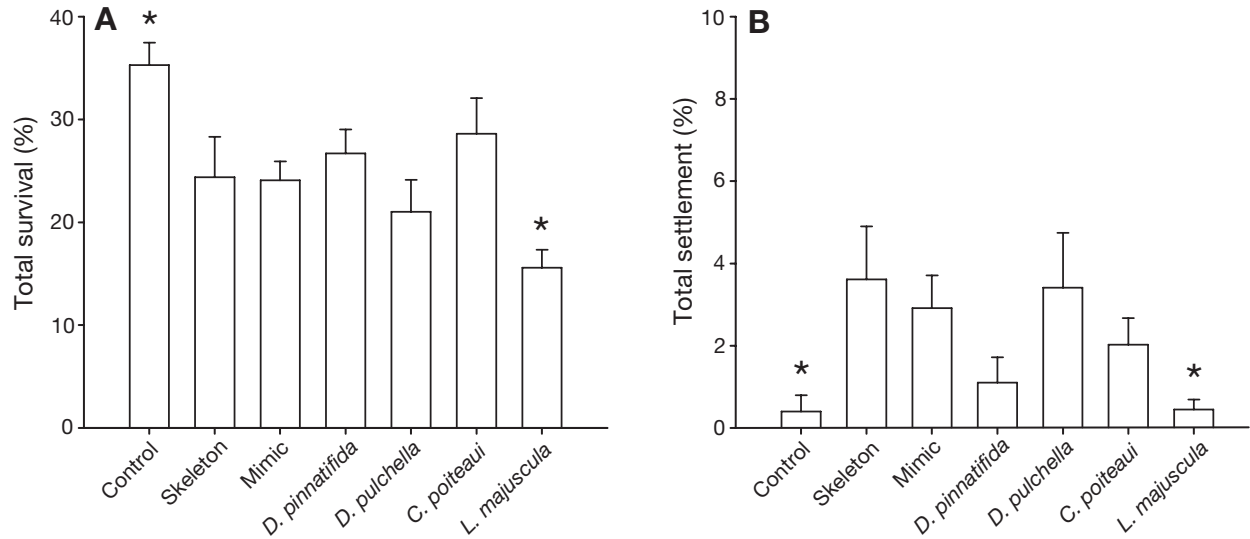


Fig. 6. *Briareum asbestinum*. Expt 6. Test of hypothesis that macrophytes affect settlement, showing mean (+1 SE) percent of larvae in chambers ($n = 10$, except for *Chondrophycus poiteaui* where $n = 11$, and *Lyngbya majuscula* where $n = 9$) that (A) survived (swimmers + settlers; *: significant difference between negative control and skeleton [$p = 0.008$], and macrophyte mimic and *L. majuscula* [$p = 0.043$]), and (B) settled (*: significant difference between negative control and skeleton [$p = 0.011$] and macrophyte mimic and *L. majuscula* [$p = 0.029$]). *D. pinnatifida*: *Dictyota pinnatifida*; *D. pulchella*: *Dictyota pulchella*

settlement cues. Baird & Morse (2004) showed similar effects of *Lobophora* sp. on the brooded planulae of *Acropora palifera* and *Stylophora pistillata* and suggested allelopathy as the mechanism. Similarly, Maida et al. (1995) showed allelopathic effects for 2 species of soft corals on scleractinian recruitment. However, an alternative to allelopathy is the hypothesis that pathogens associated with the algae are responsible for the observed negative effect (Nugues et al. 2004b).

Our study also showed evidence of positive settlement cues; *Porites astreoides* larvae clearly preferred the biologically conditioned tile to the chamber sides, even though the tile comprised only 10% of the total area available. A positive effect of shade was also suggested by our experiments. A higher proportion of larvae recruited to the bottom of the tile in the controls compared to the mimic (which provided shade) in Expts 1 and 3, and in both experiments using chambers (Expts 1 and 2), larvae that recruited to the chamber sides were overwhelmingly found in the shade provided by the chamber lid (data not shown). It is unclear why we did not see a positive effect of the mimic in Expt 2, but it is possible that irradiance levels were lower during this experiment due to increased turbidity (reversal of tidal direction during the day) or cloud cover. Coral larvae are known to recruit to the shaded undersides of recruitment tiles in shallow, high-irradiance conditions (Birkeland et al. 1981, Maida et al. 1994). In addition, in our study there was a significant positive effect of gorgonian skeleton on *Briareum asbestinum* settlement, similar to the documented effect of crustose coralline algae on sclerac-

tinian larvae (Morse et al. 1988, Heyward & Negri 1999). Evidence for the complexity and selectiveness of larval choice in substrate habitat continues to mount, showing that coral recruitment patterns are species-specific and coral larvae are highly attuned to their preferred habitat (Baird et al. 2003).

Because of the difficulty in finding very small juvenile corals in the field and the logistics of long-term recruitment studies, there is a gap in our knowledge regarding post-recruitment survival. In this study, survival of *Porites astreoides* recruits was significantly reduced (compared to the plastic flagging tape macrophyte mimic) when the recruits were placed in contact with *Dictyota menstrualis* and *Lyngbya confervoides*. Thus, it appears that these species can kill coral recruits using mechanisms other than simple shading or abrasion. As the juvenile coral reaches a certain threshold size, defense mechanisms on the part of the coral may come into play, such as the use of mesenterial filaments in reducing the growth and damaging the thalli of neighboring algae (Nugues et al. 2004a). It remains unknown how old corals have to be to employ these tactics, but apparently adult *P. astreoides* do not use this defense mechanism against algae (Nugues et al. 2004a). Lirman (2001) found that the growth of adult *P. astreoides* and *Montastrea annularis* was reduced when caged with *Dictyota* spp. Similarly, an algal clearance experiment demonstrated that coral growth (*Acropora cuneata* and *A. palifera*) and fecundity (*A. palifera*) were increased by algal removal (Tanner 1995). Thus, it is clear that coral-algae interactions can be metabolically expensive for some species of coral.

Available data on *Dictyota* spp. and cyanobacteria show that these taxa can colonize large areas on reefs. *Dictyota* spp. in the Florida Keys can propagate rapidly by vegetative fragmentation resulting from storms (Vroom et al. 2005) and fish foraging (Herren et al. 2006). Under certain conditions, benthic cyanobacteria can also spread rapidly and form blooms covering >50% of the substrate (Dennison et al. 1999, Thacker & Paul 2001). Species of *Dictyota* and *Lyngbya* are chemically rich and are generally unpalatable to most generalist grazers (Thacker et al. 1997, Nagle & Paul 1998, Paul et al. 2001), probably facilitating the formation of blooms on coral reefs. Many secondary metabolites have multiple functions (Becerro et al. 1997, Paul et al. 2001), so it is conceivable that anti-herbivory compounds are responsible for the inhibition of coral recruitment and survival observed in this study.

Further investigation into the mechanisms behind the effects observed in this study are a critical next step in determining if allelopathic interactions between benthic primary producers and coral larvae could be perpetuating phase shifts on coral reefs. While studies have shown that coral mortality, reduced herbivory and increased nutrient availability can act in concert to cause phase shifts, few have investigated the mechanisms responsible for the maintenance of high algal cover on disturbed reefs. By showing that algae and cyanobacteria can inhibit coral recruitment, we have identified 1 mechanism that could be preventing the recovery of coral reef communities.

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