

# Nutrient content of macroalgae with differing morphologies may indicate sources of nutrients for tropical marine systems

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**ABSTRACT:** To investigate whether tissue N and P content of morphologically distinct macroalgae reflect different processes controlling nutrient availability, we measured water column nutrients and collected 5 species of algae for tissue N and P analysis from 18 stations along the southwestern coast of Puerto Rico. Nutrient content of sediments was also determined for a subset of stations. Southwestern Puerto Rico was chosen because the literature suggests that gradients in sediment type and organic matter content, advection, and terrestrial influence occur in this region. Stations were either inshore or offshore areas with 3 stations per area chosen *a priori* as High Nutrient Stations (HNS). Water column and sediment nutrient concentrations were elevated inshore and in HNS. Species were of 3 morphological forms: upright thalli with open branches, densely packed mats, and rhizophytic thalli. In the first category, *Acanthophora spicifera* had higher N content inshore compared to offshore while both *A. spicifera* and *Hypnea musciformis* had higher tissue N and P contents in HNS. In contrast, mat-forming algae (*Dictyota dichotoma* and *D. cervicornis*) had higher tissue N and P contents offshore compared to inshore. Although these species had high nutrients in many of the HNS, samples from some offshore reefs were equally high. *Halimeda incrassata*, a rhizophytic form, had greater tissue N content inshore than offshore. *H. incrassata* tissue nutrients were also elevated in some HNS, but not others. There were significant correlations between water column and sediment nutrients and the tissue N and P content of *A. spicifera* and *H. incrassata*, but not for either *Dictyota*. These results suggest algae with upright thalli and open-branching patterns may have a more direct relationship between tissue nutrient content and water column nutrient concentration than other forms. In contrast, mat-forming species may deplete nutrients within the mat, relying on strong currents found offshore to penetrate dense mats and replenish nutrients. Rhizophytic algae have access to both water column and sediment nutrients, and higher inshore tissue contents and at some HNS may reflect enhanced nutrient supplies from these sources. Our findings suggest that if other environmental factors are carefully taken into consideration, the tissue N and P content of macroalgae may prove to be an effective indicator of different nutrient sources in tropical systems.

**KEY WORDS:** Macroalgae · Tropics · Nutrients · Morphological forms · Puerto Rico

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## INTRODUCTION

Macroalgae common in shallow tropical waters have a wide diversity of morphologies including filamen-

tous, sheet-like, foliose, coarsely branched and calcified forms (Littler & Littler 1980, 1985, Steneck & Watling 1982). They occur in many habitats such as coral reefs, lagoons, seagrass beds, and associated with mangrove roots (Littler et al. 1989, Littler & Littler 2000). Productivity, growth, biomass accumulation, and the structure of tropical algal communities are controlled by many environmental factors and biotic

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interactions including temperature and light (e.g. Alves De Guimaraens & Coutinho 1996, Beach & Smith 1997, Bischoff-Baermann et al. 1997), nutrients (e.g. Littler & Littler 1985, Lapointe 1987, Lapointe & O'Connell 1989), herbivory (e.g. Carpenter 1986, Szmant 1997, Russ & McCook 1999), and competition (e.g. Hughes 1989, Tanner 1995, McCook 1999).

Limited availability of inorganic nitrogen (N) and phosphorus (P) (Lapointe 1987, Corredor et al. 1999) has been identified as important in determining distribution, productivity, and biomass accumulation of tropical algae (Hatcher & Larkum 1983, Littler & Littler 1985, Lapointe 1987, Lapointe et al. 1987, 1997, Lapointe & O'Connell 1989, Schaffelke & Klumpp 1997, 1998a,b). Traditionally, nutrient availability has been estimated by monitoring water column nutrient concentrations (Boynton et al. 1982, Howarth 1988, Cadée & Hegeman 1993, Kress & Herut 1998). Many laboratory experiments have related nutrient uptake, tissue nutrient content, and growth to water column nutrient content (e.g. Hanisak & Harlin 1978, Hanisak 1983, Rosenburg & Ramus 1984). These relationships were also documented in a combined field/laboratory experiment, where other factors such as advection were carefully controlled (Lapointe 1987), and for algae placed along an estuarine gradient (Horrocks et al. 1995). However, field studies in tropical regions suggest there is usually little correlation between ambient water column N or P concentrations and either productivity or abundance of primary producers (for a review see McCook 1999). Low correlation may be due to many nutrient processes, including pulsed nutrient supplies that are spatially and temporally variable (Genin et al. 1995, Diaz & Ward 1997), rapid nutrient uptake by primary producers (Uthicke & Klumpp 1998), and tightly coupled recycling between reef organisms (Larned & Stimson 1997, Schaffelke & Klumpp 1998b). Because of this lack of correlation, water column nutrients alone may not always provide an adequate indicator of nutrient supply or bioavailability in tropical systems (McCook 1999).

Measuring nutrient availability, especially at the scale relevant to primary producers, is difficult because availability is controlled by several complex physical and chemical processes. One physical control on nutrient supply is proximity to terrestrial sources of nutrients such as springs (Lapointe et al. 1997), fringing mangrove systems (Lapointe et al. 1987), and river mouths (Nixon et al. 1996, Corredor et al. 1999). Many activities associated with development of terrestrial systems (e.g. deforestation, agriculture, sewage disposal) increase transport of nutrients to marine habitats (Nixon et al. 1996, Jaworski et al. 1997, Vitousek et al. 1997, Corredor et al. 1999, Downing et al. 1999). Other physical processes affecting availability of N

and P are advective supply and turbulent mixing across boundary layers (Atkinson & Bilger 1992, Baird & Atkinson 1997, Larned & Atkinson 1997, Thomas & Atkinson 1997). These processes may be especially important offshore, where rapid water motion is often coupled with low water column nutrient concentrations (Larned & Atkinson 1997). Closer to shore, advection may be lower, but water column nutrient supplies may be supplemented by terrestrial or sediment sources (Lapointe & O'Connell 1989, Larned & Stimson 1997). One chemical process important in determining nutrient availability is adsorption of P to carbonate sediments; adsorption may contribute to low and variable P availability in the water column of shallow tropical areas (Delgado & Lapointe 1994, McGlathery et al. 1994, Knoppers et al. 1996, Corredor et al. 1999). However, even for carbonate sediments with large capacities for P adsorption, sustained high levels of P loading into a system will result in a proportionately higher amount of P being biologically available (McGlathery et al. 1994, Corredor et al. 1999).

Recent investigations suggest that the tissue N and P content and N:P ratio in the tissue of marine macroalgae have potential as indicators of nutrient availability (Wheeler & Björnsäter 1992, Fong et al. 1998, McCook 1999). In an early investigation, Atkinson & Smith (1983) concluded that C:N:P ratios in macroalgae and seagrasses collected from low-nutrient tropical areas were significantly different from those in high-nutrient temperate areas. Björnsäter & Wheeler (1990) linked tissue N and P content to nutrient supplies in laboratory cultures of macroalgae. Based on these results, they used the N:P ratios of field collected algae to determine seasonal changes in N and P limitation (Wheeler & Björnsäter 1992). Fong et al. (1994) developed a quantitative relationship between nutrient supply and tissue N and P content of an opportunistic green macroalga. This relationship was used to develop and test a bioindicator of nutrient supply in the field for southern California (Fong et al. 1998). Results of these studies suggested that tissue N and P contents and N:P ratios of macroalgae may be more useful than traditional water quality sampling for estimating biologically available pulses of nutrients (Horrocks et al. 1995, Fong et al. 1998).

In this paper, we investigated the potential for tissue N and P content and the N:P ratio of common species of morphologically divergent macroalgae to be used as an indicator of different processes controlling nutrient availability to macroalgae across several tropical habitats. We hypothesize that nearshore habitats have greater nutrient supplies to the water column and therefore algae associated with the water column that are collected nearshore will have higher nutrient content. Sediments have been identified as an important

source of nutrients to tropical algae with bulbous holdfasts and rhizoids that anchor in sediments (Williams 1981, Larned 1998). We hypothesize that rhizophytic algae will have higher nutrient content in inshore areas with larger water column and sediment nutrient pools. In contrast, for algae that form densely compacted mats, we hypothesize that rates of flow (advection) may be especially important in controlling nutrient availability, uptake, and storage because reduction of flow within mats forms large nutrient-depleted boundary layers. Thus, mat-forming algae in high flow areas should have greater tissue nutrients than mats in low flow areas.

## METHODS

**Study area.** We chose to sample the southwestern coast of Puerto Rico (Fig. 1) because the literature suggests that gradients in sediment type and organic matter content, advection, and terrestrial influence occur in this region (Odum et al. 1959, Morelock et al. 1977, Breyer & Ehlmann 1981, Rafalska-Bloch 1985). All of our sampling stations are in the inner and middle shelf regions identified by Morelock et al. (1977); the inner shelf is further divided based on sediment types and geomorphology. Morelock et al. (1977) found that mangrove dominated coasts and shallows contain mostly fine-grained carbonates, while sediments from areas further from shore contain sandy silt to sand in seagrass beds and are very coarse-grained around reefs. Sediment organic matter also changes along an onshore-offshore gradient (Rafalska-Bloch 1985), with highest organic content along mangrove-dominated

coasts (3.15%), then abruptly decreasing as you move offshore to seagrass beds (0.62%) and reef lagoons (0.47%). Both inner and middle shelf reef sediments had very low organic content (0.16%).

Rafalska-Bloch (1985) describes southwestern Puerto Rico as an open coast because the lack of a shallow barrier reef allows relatively high water flow everywhere across the shelf. Depositional energy is very high around patch reefs, intermediate over seagrass beds and lagoons, and is low only in coastal mangrove areas (Rafalska-Bloch 1985). Early work established that advection, measured as current speed, also varied between inshore and offshore regions of equal depth (Odum et al. 1959); in a study measuring current speed in winter, spring and summer, inshore currents ranged from 0 to 4.2 m min<sup>-1</sup> while offshore speeds were 5 to 9 m min<sup>-1</sup>.

Southwestern Puerto Rico is semi-arid, with a rainy season from September through October (Morelock et al. 1977). We sampled in the dry season from 15 to 29 January 1998. There was measurable rainfall on 5 of 15 sampling days (15, 20, 25, 26, and 29 January) for a total of 0.20 cm (NOAA rainfall data 1959 to 1998 for Magueyes Island). All of January 1998 was relatively dry with a total of 0.78 cm of rain compared to the 40 yr average of 1.38 cm (NOAA rainfall data 1959 to 1998 for Magueyes Island). The prior rainy season was also relatively dry, with a total of 5.00 cm in September and October of 1997 compared to the 40 yr average of 8.84 cm (NOAA rainfall data 1959 to 1998 for Magueyes Island). As all of the major drainages for inland portions of the island are to the east of our sampling sites (Bryer & Ehlmann 1981), terrestrial runoff to inshore stations is from local sources, not inland where rainfall can be considerably higher.

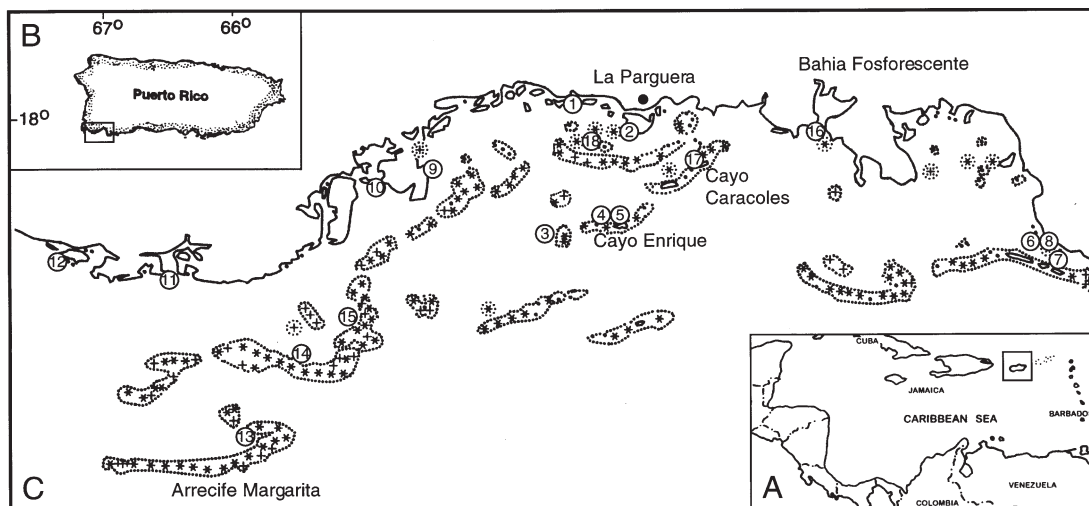


Fig. 1. Map of: (A) the Caribbean Sea outlining Puerto Rico; (B) Puerto Rico identifying the sampling area; and (C) the 18 sampling stations off the SW coast of Puerto Rico. In southwestern Puerto Rico, the prevailing wave direction is from the southeast and the prevailing current moves from east to west along the shore

We visited 18 sampling stations along the southwestern coast of Puerto Rico (Fig. 1). Sampling stations were grouped into 2 categories. Inshore stations were within 10 m of the main island in mangrove, seagrass habitats, or rubble (Stns 1, 6, 7, 8, 9, 10, 11, 12 and 16) or offshore of the main island in mangrove and seagrass habitats (Stns 2, 5, 15, 17 and 18) and shallow reefs (Stns 3, 4, 13 and 14). Stations were established both east and west of the town of La Parguera. Stations were as uniform in depth as possible (1 to 3 m), and care was taken to collect in areas that were not shaded.

Some stations were chosen haphazardly, but others were selected *a priori* as potential High Nutrient Stations (HNS). Inshore HNS included Stns 1, 11 and 16. Secondary sewage effluent is released into percolation ponds adjacent to the fringing mangroves in La Parguera (Corredor & Morell 1994). Stn 1 was within 10 m of this potential nutrient source. Stn 11 was inside an embayment that had lower carbonate content compared to our other inshore areas (Jacobson & Fong unpubl. data); these sediments may be a source of terrestrial nutrients. Stn 16 was at the mouth of Bahia Fosforescente; the benthic communities in the fringing areas of this bay were found to be highly productive (Odum et al. 1959) and have sediments with high organic content (2.79%; Rafalska-Bloch 1985) suggesting this may be an area with enhanced nutrient supply. Stations chosen as potential offshore HNS included Stns 15, 17, and 18. Stn 15 was a mangrove island used extensively by local fishermen as a fish cleaning station. Refuse is dropped into the water on a daily basis, and was observed in various states of decay on the benthos at the time of collection. We hypothesize that degradation processes released significant amounts of nutrients at this station. Stn 17 was in the lee of an island where several live-aboard sailboats anchor. Typically these boats release untreated wastewater directly into the ocean. Stn 18 was off an island with a large bird rookery. Dense aggregations of birds have been documented to be a nutrient source to nearby shallow marine habitats (Powell et al. 1989).

**Water column and sediment nutrients.** Water samples were collected at each of the 18 stations concurrent with algal collections. Care was taken to collect water samples near the benthos, just above the algae. Water samples were placed on ice in a dark cooler immediately after collection, returned to the lab within 3 h, filtered with Whatman GF/C filters, and frozen. Frozen samples were packed in dry ice in a cooler and sent to DANR Analytical Lab. at the University of California Davis. We confirmed with the lab that samples arrived the next day, and were still frozen. Water samples were analyzed for Total Kjeldahl Nitrogen (TKN),  $\text{NO}_3$ ,  $\text{NH}_4$ , and total phosphate (TP). The TKN method is based on the wet oxidation of nitrogen using sulfuric acid and digestion

catalyst. The procedure converts organic nitrogen to the ammonium form and subsequent determination of ammonium. The procedure does not quantitatively digest nitrogen from heterocyclic forms (bound in a carbon ring), or from oxidized forms such as nitrate and nitrite. Ammonium and nitrate are measured by the diffusion-conductivity method as described by Carlson (1978) based on the gaseous diffusing of ammonia ( $\text{NH}_3$ ) across a gas permeable membrane in the presence of excess base (KOH) and subsequent conductivity detection. Total phosphorous in seawater is measured utilizing a nitric acid/hydrogen peroxide microwave digestion (Johnson & Ulrich 1959). Subsequently, the analyte concentration is determined by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) with a vacuum spectrometer (Franson 1985). These automated methods have detection limits of 3.57  $\mu\text{M}$  for N and 1.56  $\mu\text{M}$  for P. Many of the 54 samples taken were below these detection limits. Number of samples below detection were 2 for TKN, 49 for  $\text{NH}_4$ , 3 for  $\text{NO}_3$ , and 19 for TP.

Sediment samples were taken at 8 of the 18 stations in Sep 1998, 8 mo after the water and algal samples were collected. Ideally, sediment samples would have been taken concurrently with other sampling. However, as this was not possible at the time, we used these later samples for comparison. Sediment cores (5 cm deep) were taken with 60 ml syringes (inner diameter = 2.5 cm) with the end cut off and edges sharpened. Ten sediment cores were taken from each site; cores were haphazardly located within each collection site. Cores were stored on ice in the dark, returned to the lab within 3 h, dried in a forced air oven ( $<60^\circ\text{C}$ ) to constant wt (24 to 48 hr), ground with a mortar and pestle, and analyzed for TKN, total P, and P-Olsen content at DANR Analytical Lab. Total Kjeldahl Nitrogen in sediment is determined by measuring total reduced nitrogen after the wet oxidation of sediment organic matter using standard Kjeldahl procedure with sulfuric acid and digestion catalyst (Issac & Johnson 1976, Carlson 1978). Total P in sediments was determined by microwave acid digestion/dissolution of sample (Sah & Miller 1992) followed by atomic emission spectroscopy. The detection limit of this method is  $<0.010\%$  P, resulting in 48 of 76 samples being below limits. P-Olsen is extractable phosphate based on alkaline extraction by 0.5 Normal  $\text{NaHCO}_3$ . It measures available phosphate for sediments with pH greater than 6.5 by ascorbic acid reduction of phosphomolybdate complex and measurement by spectrophotometry (Olsen et al. 1954).

**Algal collections.** Five common species of algae were collected at as many of the 18 stations as they occurred. Algae were considered common based on visual observations of presence/absence during initial surveys over a wide variety of areas. Two congeneric species of brown algae (Division Chromophyta), *Dictyota dichotoma*

*toma* and *Dictyota cervicornis*, were collected. Thalli are thin, dichotomously branched flattened blades that are often formed into very dense, tightly packed mats on hard bottom, rubble, or coarse sand. For this study we only collected thalli of *Dictyota* that formed dense mats. Two species, *Acanthophora spicifera* and *Hypnea musciformis*, are red algae (Division Rhodophyta) with open-branching patterns compared to the mat-forming *Dictyota*. *A. spicifera* has a coarsely branched morphology with upright portions extending into the water column. It was often found growing epiphytically on the roots of mangroves. When collecting from mangrove roots, samples were only taken from the outermost, sunlit roots. In seagrass or reef habitats, *A. spicifera* was found growing on rubble or hard substrate. We only collected specimens that had open branches extending into the water column. *H. musciformis* is a more finely branched species that frequently epiphytized algae attached to rubble or hard bottoms. The fine branches usually extend into the water column above its host, and we restricted our collections to these samples. The fifth species, *Halimeda incrassata* is a psammophytic green alga (Division Chlorophyta) that is anchored in sandy substrates by a bulbous rhizoidal system that allows access to sediment nutrients (Littler et al. 1991). It also has an openly branched and upright thallus that is heavily calcified and extends up into the water column.

At each of the 18 stations we collected algal samples at 3 sites to test for within station variability. Each of the sites in a station was within 10 m of each other. At each site we attempted to collect 5 replicate samples of each alga. However, not all species of algae were found at each station or within each site in sufficient abundance to accomplish this goal. Because sample sizes were not always consistent within a station, we included sample sizes in every figure and table.

Within 3 h of collection, algal samples were carefully sorted to species and cleaned of sediments, macroinvertebrates, and epiphytes. Tissue was briefly rinsed in freshwater to remove salts, dried in a forced air oven (60°C) to constant wt (24 to 48 h), ground with mortar and pestle or a Wiley Mill, and analyzed for tissue N and P content. The heavy  $\text{CaCO}_3$  deposits in *Halimeda incrassata* result in less living tissue per gram wt. Because tissue N and P content is determined as a percentage of dry wt, tissue N and P values for *H. incrassata* should be lower than for the other species of algae collected in this study.

Algal samples were analyzed for tissue N and P content at DANR Analytical Lab using standard methods. The method for total N quantitatively determines the amount of nitrogen in all forms (ammonium, nitrate, protein and heterocyclic nitrogen) in botanical materials using an induction furnace and a thermal conductiv-

ity detector. In this method, samples are ignited in a quartz combustion tube containing a helium and oxygen environment placed in an induction furnace at approximately 900°C. An aliquot of the combustion gases is passed through a copper catalyst to remove oxygen and convert nitrous oxides to  $\text{N}_2$ . The aliquot of combustion gases is then scrubbed of moisture and carbon dioxide. Finally, the nitrogen content is determined by thermal conductivity (Sweeney 1989). The method for total phosphorous quantitatively determines the concentration of phosphorous in botanical materials utilizing a nitric acid/hydrogen peroxide microwave digestion (Johnson & Ulrich 1959). Subsequently, the analyte concentration is determined by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES) with a vacuum spectrometer (Franson 1985).

**Data analysis.** Data were tested to ensure that they conformed to the assumptions of parametric statistics; no transformations were required. Nested ANOVA did not find significant differences in mean N or P contents among sites within stations for any of the species of algae, so the nested factor (site within stations) was dropped from all subsequent analyses.

Single-factor ANOVA (factor = species) was used to identify differences in tissue N, P, and N:P ratios among the 5 species of macroalgae. All replicate samples for all stations for all algal species were used in this analysis. After a statistically significant ANOVA, differences in mean values among species were identified using a Protected Least Significant Difference (PLSD) test. This test is a conservative multiple comparison technique as it controls experiment-wise error; the unprotected test allows this error to increase with each comparison.

We used *t*-tests to determine if there were significant differences between mean water column nutrients, sediment nutrients, and N, P and N:P ratios in samples collected from inshore (Stns 1, 6, 7, 8, 9, 10, 11, 12 and 16) vs offshore (Stns 2, 3, 4, 5, 13, 14, 15, 17 and 18). For water and sediment samples, each independent sample (water bottle or sediment core) was used as a replicate, regardless of station. Statistical analyses were not conducted on sediment total P as only 3 of 28 samples collected offshore had total P contents above detection limits. In the tissue nutrient analysis, we compared all samples collected inshore to all samples collected offshore for each algal species separately, regardless of station. Inshore/offshore patterns were analyzed for 4 of 5 species of algae because *Hypnea musciformis* was only found in 1 offshore station, which we did not consider to be sufficient representation of offshore sites. For all comparisons there were greater sample sizes inshore compared to offshore. However, the relative proportion of samples from HNS were similar inshore and offshore as algae were usually abundant in these locations.



Single-factor ANOVA was used to determine differences in water column and sediment nutrients among stations. Single-factor ANOVA (factor = station, with all samples collected within each station used as replicates) was also used to compare mean N and P contents and N:P ratios for 3 of the species (*Acanthophora spicifera*, *Dictyota dichotoma*, and *Halimeda incrassata*). This analysis was also conducted for *Hypnea musciformis* and *Dictyota cervicornis*, though, unlike the above analysis, these results are only reported in a table. We did not feel these results warranted both table and graph as these species were found with replication in less than half of the stations (8 and 5 stations, respectively). After a statistically significant ANOVA, differences among means were identified using a PLSD test. Finally, correlations between water column, sediment and algal tissue nutrients were determined.

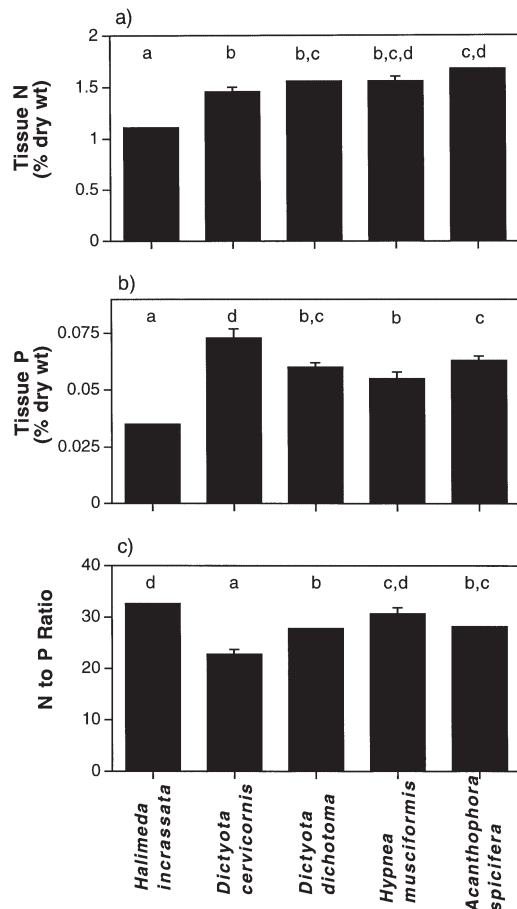


Fig. 2. Tissue (a) N, (b) P, (c) N:P ratio in different species of algae collected in southwestern Puerto Rico (error bars = SE). Letters above the bars show the results of the PLSD; different letters distinguish significantly different means ( $p < 0.05$ ). Samples size for *Acanthophora spicifera* = 210, *Halimeda incrassata* = 150, *Dictyota dichotoma* = 133, *Dictyota cervicornis* = 67, and *Hypnea musciformis* = 39

## RESULTS

### Comparisons among species

Tissue N differed significantly among the 5 species of algae collected (Fig. 2a; ANOVA  $p = 0.0001$ ). As expected because of  $\text{CaCO}_3$  deposits (i.e. less living tissue per g of wt), tissue N was lowest in *Halimeda incrassata* (PLSD  $p = 0.0001$  for all comparisons). There were slight differences in tissue N contents among the other 4 species of algae, with *Dictyota cervicornis* lowest and *Acanthophora spicifera* highest.

Differences in mean P contents were also significant among the algal species (Fig. 2b; ANOVA  $p = 0.0001$ ). There were proportionally larger differences among mean tissue P contents than N contents for the 5 species of algae. P content of *Halimeda incrassata* was lower than all other species (PLSD  $p < 0.001$  for all comparisons). P content of *Dictyota cervicornis* was the highest (PLSD  $p < 0.003$  for all comparisons), about double that of *H. incrassata*. Among the 3 species of algae with intermediate P contents, *Acanthophora spicifera* ranked highest and *Hypnea musciformis* lowest. Tissue P content of *Dictyota dichotoma* was between these two, and not significantly different from either.

N:P ratios also differed among algal species (Fig. 2c; ANOVA  $p = 0.0001$ ). The N:P ratio of *Dictyota cervicornis* was lower than all others (PLSD  $p = 0.0001$ ) due to a high tissue P content. There was considerable overlap among the other 4 species with *Halimeda incrassata* ranked the highest and *Dictyota dichotoma* the lowest.

There were significant correlations between tissue N and P content for *Acanthophora spicifera* ( $R = 0.847$ ,  $p = 0.0001$ ), *Dictyota dichotoma* ( $R = 0.752$ ,  $p = 0.012$ ), *Dictyota cervicornis* ( $R = 0.828$ ,  $p = 0.0058$ ), and *Halimeda incrassata* ( $R = 0.829$ ,  $p = 0.0005$ ). Tissue N and P content did not correlate in *Hypnea musciformis*.

### Inshore versus offshore comparisons

Mean TKN in water samples collected inshore was significantly higher than offshore (Table 1). No other measure of water column nutrients differed between inshore and offshore samples, though  $\text{NO}_3$  was close to significant. TKN in sediments inshore was more than triple that offshore (Table 1) while mean P-Olsen in sediments inshore was nearly double the offshore content. Only 3 offshore samples had TP above detection limits, so statistical analysis was not conducted. However, the low number of samples with measurable TP offshore compared to inshore suggest there is less P in offshore sediments.

*Acanthophora spicifera* and *Halimeda incrassata* had similar inshore/offshore patterns in tissue nutrient content (Table 1). Samples of both algae collected inshore had significantly higher tissue N than samples collected offshore around mid-shelf islands or reefs. This reflected elevated N in both the water and sediments inshore. Although extractable P in sediments was higher inshore, mean tissue P contents were not different between onshore and offshore stations for either species. Thus, N:P ratios for both species were higher inshore due to greater tissue N of inshore samples ( $t$ -test  $p = 0.0001$  for both species).

Inshore/offshore patterns of N and P contents for the 2 species of *Dictyota* were similar to each other, but different from the other algal species (Table 1). Both species of *Dictyota* had higher N content offshore compared to inshore. Tissue P was also almost twice as high offshore compared to inshore for *D. cervicornis*. While mean tissue P was higher offshore for *D. dichotoma*, this difference was not significant. Mean N:P ratio for *D. cervicornis* was higher inshore than offshore. There were no differences in N:P ratios for *D. dichotoma*. Neither tissue N nor P contents reflected water column or sediment concentrations.

### Comparisons among stations

There were no significant differences in water column TKN among stations (Table 2; ANOVA  $p = 0.0781$ ). With the exception of the High Nutrient Stations (HNS), TKN was usually lower offshore than inshore. There were no patterns in mean water column TP among stations. However, there were significant differences in  $\text{NO}_3$  among stations (ANOVA  $p = 0.0004$ ). Stn 1 (sewage ponds) and Stn 11 (low-carbonate bay) had higher  $\text{NO}_3$  than any of the other stations (PLSD  $p < 0.007$  for all comparisons). These were inshore HNS. Bahia Fosforescente, another HNS, did not have elevated  $\text{NO}_3$ . Although not statistically different due to high variability,  $\text{NO}_3$  concentrations in offshore HNS were higher than in other stations that were not HNS.

There was a significant difference in sediment TKN, TP, and P-Olsen (ANOVA  $p = 0.0001$  for all 3 comparisons) among stations (Table 3). Three of 5 inshore sta-

Table 1. Patterns of water column nutrients, sediment nutrients, and tissue N, P, and N:P ratio between inshore and offshore samples. Numbers are means (SE).  $p$ -values are results of  $t$ -tests. \*Identifies significant differences and number in bold identify the larger values. – Indicates no statistical analysis due to insufficient sample size

	n	Inshore	n	Offshore	p
Water column nutrients					
TKN	25	<b>45.71 (4.10)</b>	27	28.04 (4.64)	0.0066*
$\text{NO}_3$	23	7.52 (1.45)	27	5.02 (0.26)	0.0745
TP	14	1.876 (0.114)	20	1.750 (0.080)	0.3590
Sediment nutrients					
TKN	45	<b>0.21 (0.02)</b>	28	0.06 (0.01)	0.0001*
TP	34	0.031 (0.003)	3	0.023 (0.003)	–
P-Olsen	48	<b>33.719 (2.787)</b>	28	18.429 (2.450)	0.0004*
<i>Acanthophora spicifera</i>					
Tissue N	122	<b>1.76 (0.05)</b>	88	1.61 (0.04)	0.0155*
Tissue P	122	0.063 (0.002)	88	0.067 (0.002)	0.3173
N:P ratio	122	<b>30.35 (0.773)</b>	88	24.959 (0.50)	0.0001*
<i>Halimeda incrassata</i>					
Tissue N	100	<b>1.18 (0.3)</b>	50	0.99 (0.04)	0.0002*
Tissue P	100	0.036 (0.001)	50	0.035 (0.0020)	0.6119
N:P Ratio	100	<b>33.57 (0.44)</b>	50	30.06 (0.91)	0.0001*
<i>Dictyota cervicornis</i>					
Tissue N	33	1.28 (0.06)	34	<b>1.63 (0.05)</b>	0.0001*
Tissue P	32	0.053 (0.005)	33	<b>0.092 (0.0050)</b>	0.0001*
N:P Ratio	32	26.92 (1.23)	33	<b>18.77 (0.85)</b>	0.0001*
<i>Dictyota dichotoma</i>					
Tissue N	70	1.47 (0.06)	63	<b>1.74 (0.04)</b>	0.0004*
Tissue P	69	0.059 (0.004)	61	0.067 (0.002)	0.0533
N:P Ratio	69	27.41 (0.70)	61	27.97 (1.34)	0.7016
<i>Hypnea musciformis</i>					
Tissue N	35	1.61 (0.04)	4	1.09 (0.06)	–
Tissue P	34	0.054 (0.003)	3	0.067 (0.003)	–
N:P Ratio	34	31.88 (1.08)	3	16.70 (1.23)	–

tions (Stns 1, 9 and 11) had significantly higher TKN contents than other stations (PLSD  $p < 0.006$  for all comparisons). In contrast, the 2 offshore stations that were not HNS (Stns 3 and 14) were an order of magnitude lower in TKN (PLSD  $p < 0.05$ ). The offshore HNS (Stn 17) had significantly higher TKN than other offshore stations (PLSD  $p < 0.05$  for both comparisons), but ranked with inshore stations that were not HNS. Sediment TP contents of offshore stations that were not HNS (Stns 3 and 14), as well as one inshore station (Stn 6), were always below detection limits (Table 3). Although the 2 HNS inshore (Stns 1 and 11) had high TP contents, another inshore station (Stn 9) that was not chosen *a priori* as a HNS also had high TP (PLSD  $p < 0.006$  for all comparisons). For extractable P (P-Olsen), values ranked highest in the inshore HNS (1 and 11). In the rest of the stations, extractable P decreased from the offshore HNS (Stn 17), to the inshore stations (Stns 6, 9 and 12), with the lowest values found in the offshore stations (Stns 3 and 14) (PLSD  $p < 0.05$  for all comparisons).

Table 2. Water column nutrient content for stations in southwestern Puerto Rico during Jan 1998. Where  $n < 3$ , other measures are below detection limits. High Nutrient Stations (HNS) are indicated by \*

Location	Stn	TKN ( $\mu\text{M}$ )	n	TP ( $\mu\text{M}$ )	n	$\text{NO}_3$ ( $\mu\text{M}$ )	n
Inshore*	1	54.76 (2.38)	3	–		15.00 (5.77)	3
Inshore*	11	52.38 (8.59)	3	2.345	2	18.33 (5.24)	3
Inshore*	16	30.95 (2.38)	3	1.560	1	4.53 (0.24)	3
Inshore	6	52.38 (8.59)	3	2.190	1	4.05 (0.24)	3
Inshore	7	42.86 (18.90)	3	1.560	2	3.93	2
Inshore	8	59.52 (17.17)	3	1.560	1	5.00 (0.41)	3
Inshore	9	52.38 (15.51)	3	1.773 (0.107)	3	3.93	2
Inshore	10	21.43	1	2.190	1	3.57	1
Inshore	12	28.57 (8.25)	3	1.880 (0.000)	3	4.29 (0.41)	3
Offshore*	15	50.00 (10.91)	3	1.667 (0.000)	3	6.56 (0.84)	3
Offshore*	17	35.71 (25.08)	3	1.720	2	6.45 (0.83)	3
Offshore*	18	54.76 (22.71)	3	1.560	2	6.25 (1.00)	3
Offshore	2	9.52 (2.38)	3	2.190 (0.479)	3	4.05 (0.48)	3
Offshore	3	26.19 (12.60)	3	1.560	2	4.05 (0.24)	3
Offshore	4	26.19 (2.38)	3	1.773 (0.107)	3	4.76 (0.47)	3
Offshore	5	9.52 (2.38)	3	1.720	2	5.00 (0.00)	3
Offshore	13	16.67 (2.38)	3	1.667 (0.107)	3	4.05 (0.24)	3
Offshore	14	23.81 (4.76)	3	–		4.05 (0.24)	3

There were differences in tissue N contents of *Acanthophora spicifera* among stations (ANOVA  $p = 0.0001$ ). HNS ranked high in tissue N regardless of

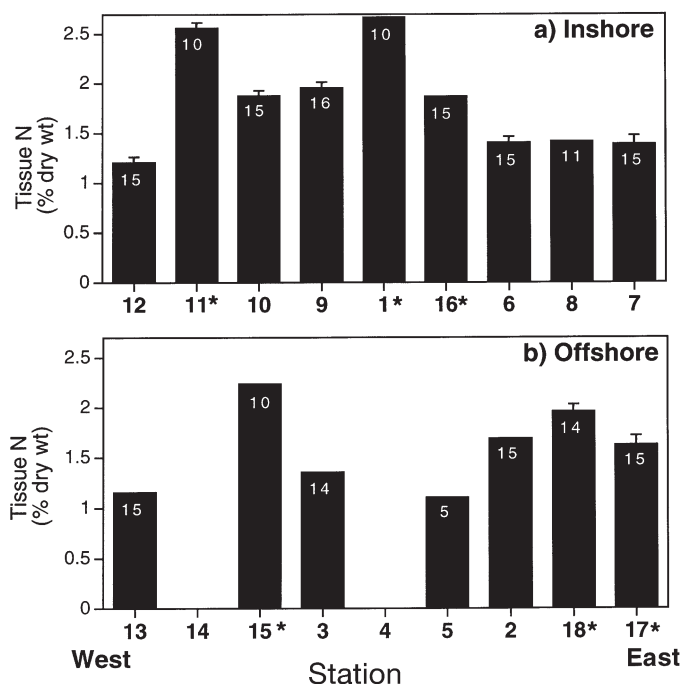


Fig. 3. Tissue N content of *Acanthophora spicifera* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. High Nutrient Stations (HNS) are indicated by \*

whether they were inshore or offshore (Fig. 3). Inshore HNS, Stn 1 (sewage ponds) and Stn 11 (low-carbonate bay), were significantly higher than all other samples (Table 4; PLSD  $p < 0.0005$  for all comparisons). Tissue N contents of *A. spicifera* from Stns 1 and 11 were 2.67% dry wt and 2.56% dry wt, respectively. These 2 stations also had high water column TKN and  $\text{NO}_3$  (Table 2) as well as sediment TKN (Table 3). The next highest tissue N (2.24% dry wt) was in algae collected in Stn 15, the offshore HNS at fishermen's island (PLSD  $p < 0.0005$ ) that also had high water column TKN and intermediate  $\text{NO}_3$  values (Table 2). The 4 means ranked next highest in tissue N grouped together (Table 4) with mean tissue N in station 18  $> 9 > 10 > 16$ . Of these stations, 2 were identified *a priori* as HNS; Stn 18 is the offshore bird rookery and Stn 16 is an inshore station at the mouth of Bahia Fosforescente. Stn 18 had elevated TKN and intermediate  $\text{NO}_3$  while concentrations of both forms of N were lower in Stn 16. Although inshore Stns 9 and 10 were not identified *a priori* as HNS, they were the stations closest downcurrent from the sewage ponds and had elevated water column and sediment nitrogen (Tables 2 & 3). With the exception of Stn 10, where  $n = 1$ , these tissue N rankings were the same as water column TKN and  $\text{NO}_3$  concentration rankings (Table 2). Sediment TKN was also high in Stn 9. Tissue N contents of Stns 17 and 2 also grouped together ( $p < 0.02$  for all comparisons); Stn 2 was offshore of Isla Magueyez Island, which houses the marine lab, while Stn 17 was one of our offshore HNS, the sailboat anchorage. Intermediate tissue N values were found for inshore stations upcurrent from the town of La Parguera (Stns 6, 7 and 8) and an



offshore reef (Stn 3). The lowest mean N contents (1.10 to 1.21 % dry wt) were found at the inshore station most distant from the town (Stn 12) and 2 offshore reefs (Stns 5 and 13).

There were significant differences in tissue P contents of *Acanthophora spicifera* among stations (ANOVA  $p = 0.0001$ ). As for tissue N, HNS had high tissue P regardless of whether they were inshore or offshore (Fig. 4). However, there was less statistical separation among means for tissue P than tissue N for the lower values (Table 1). With a few exceptions, similar groupings of stations with high P occurred as with high N. Stn 1 had the highest tissue P (0.120 % dry wt; PLSD  $p < 0.05$ ), followed by Stns 15 and 11 (0.111 and 0.102 % dry wt, respectively; PLSD  $p < 0.0001$ ); these were all identified *a priori* as HNS and had high sediment total and extractable P (Table 3) where measured. However, water column concentrations did not co-vary with tissue P (Table 2). Mean tissue P in algae

Table 3. Sediment nutrient content for sites in southwestern Puerto Rico during Sep 1998. Where  $n < 10$ , other measures are below detection limits. High Nutrient Stations (HNS) are indicated by \*

Location	Site No.	TKN (% dry wt)	n	TP (% dry wt)	n	P-Olsen (ppm)	n
Inshore*	1	0.24 (0.02)	10	0.035 (0.004)	10	44.48 (5.20)	10
Inshore*	11	0.36 (0.07)	10	0.033 (0.003)	10	48.37 (8.35)	10
Inshore	6	0.06 (0.00)	10	–		27.94 (4.05)	10
Inshore	9	0.24 (0.04)	8	0.046 (0.003)	7	21.00 (1.72)	10
Inshore	12	0.18 (0.00)	9	0.012 (0.001)	9	26.12 (2.52)	9
Offshore*	17	0.11 (0.02)	9	0.025	2	34.39 (1.94)	10
Offshore	3	0.03 (0.00)	9	–		7.32 (0.69)	9
Offshore	14	0.03 (0.00)	9	–		11.36 (0.51)	9

from Stns 18, 16 and 17 grouped together (PLSD  $p < 0.0005$ ) with tissue P values ranging from 0.075 to 0.081 % dry wt. With the exception of Stn 12, the remaining tissue P contents graded together, with means ranging from 0.042 to 0.061 % dry wt. Mean tissue P content of *A. spicifera* from Stn 12 was significantly lower (0.026 % dry wt; PLSD  $p < 0.003$ ) than all other stations.

Table 4. Results of Protected Least Significant Difference tests after statistically significant ANOVA. Station numbers are listed in order of increasing mean (left to right) algal tissue nutrient or ratio. Bars connect means that are not significantly different from each other

Tissue N <i>Acanthophora spicifera</i>															
5	13	12	3	7	6	8	17	2	16	10	9	18	15	11	1
Tissue P <i>Acanthophora spicifera</i>															
12	5	8	13	3	6	2	10	9	7	17	16	18	11	15	1
Tissue N <i>Dictyota dichotoma</i>															
12	10	6	5	7	2	11	15	13	4	3	1				
Tissue P <i>Dictyota dichotoma</i>															
12	10	6	3	11	7	5	4	2	15	13	1				
Tissue N <i>Halimeda incrassata</i>															
18	16	12	9	15	8	2	13	6	7	17	1	11			
Tissue P <i>Halimeda incrassata</i>															
18	16	9	8	12	7	13	15	6	17	1	2	11			

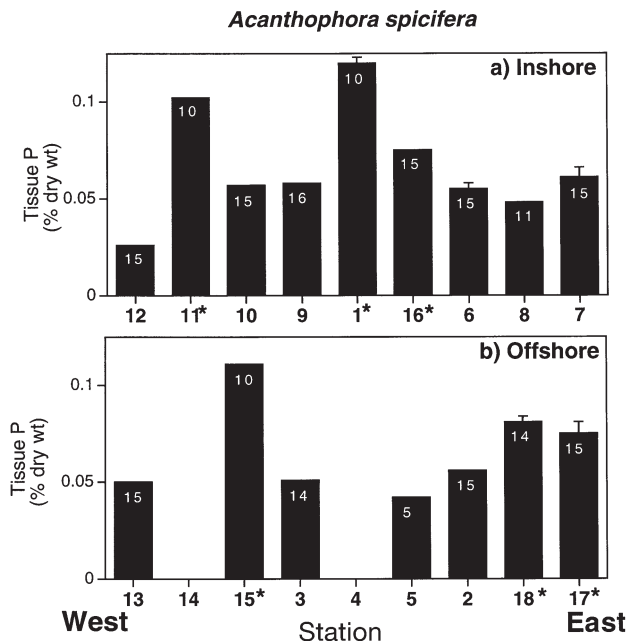


Fig. 4. Tissue P content of *Acanthophora spicifera* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located from along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. High Nutrient Stations (HNS) are indicated by \*

Mean tissue N contents for *Dictyota dichotoma* varied among stations (ANOVA  $p = 0.0001$ ), ranging from 0.81 to 2.16% dry wt (Fig. 5). Among station patterns for *D. dichotoma* were very different than for *Acanthophora spicifera* (Table 4). Although *D. dichotoma* tissue from Stn 1 ranked the highest in N content, it was not significantly higher than tissue collected at Stn 3, one of the offshore reefs (PLSD  $p > 0.05$ ). The 7 stations ranked as highest in tissue N content (highest to lowest: Stns 1, 3, 4, 13, 15, 11 and 2) included all 3 of the HNS where *D. dichotoma* was collected with enough replication for statistical analysis (HNS underlined). However, they also included 3 of the 5 reef stations that were not HNS (Stns 3, 4 and 13). In addition, 4 of the 5 lowest ranked means were from inshore stations (Stns 12, 10, 6 and 7). Among these means, Stn 12 was the lowest (0.810% dry wt).

P contents of *Dictyota dichotoma* also varied among stations (ANOVA  $p = 0.0001$ ), ranging from 0.020 to 0.110% dry wt (Fig. 6, Table 4). Of the stations with enough replication for statistical analysis, Stns 1 (sewage ponds) and 13 (offshore reef) had the highest P (PLSD  $p < 0.01$  for all comparisons). Stations 2 (Isla Magueyez) and 15 (Fisherman's Island) had the next highest P contents (PLSD  $p < 0.001$ ); both of these were

also offshore sites. P contents among the rest of the stations were all lower than 0.060% dry wt. As with tissue N, Stn 12 had the lowest P.

*Halimeda incrassata*, a psammophytic alga, was found more often inshore than offshore (Fig. 7). There were significant differences in tissue N contents among stations (ANOVA  $p = 0.0001$ ), although the range in tissue N (0.74 to 1.65% dry wt) was smaller than for other species. The 3 highest ranked stations for tissue N of *H. incrassata* were HNS (Table 4). Stn 11 (low-carbonate bay) was significantly higher than other stations (PLSD  $p < 0.0001$ ). Stns 1 (sewage ponds) and 17 (sailboat anchorage) grouped together as the next highest means (PLSD  $p < 0.001$  for all comparisons). However, other HNS such as Stns 18 (bird rookery), 16 (Bahia Fosforescente) and 15 (Fisherman's Island) had some of the lowest tissue N. Stns 18 and 15 had high water column TKN while  $\text{NO}_3$  values were the same as in non-HNS (Table 2). No sediment data were taken for these stations (Table 3). It is possible that the water column, but not the sediments, had elevated nutrients. Stns 12 and 8 had low tissue N for *H. incrassata*, similar to results for other species. However, Stns 6 and 7 were relatively high in tissue N for *H. incrassata* whereas they were low for the other species.

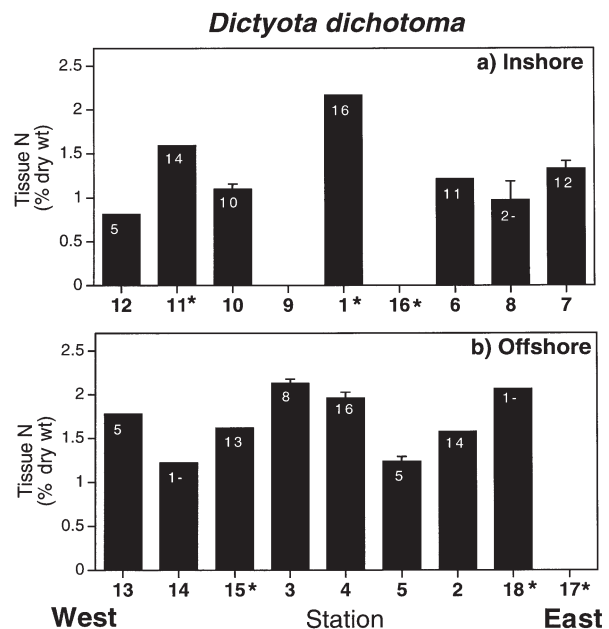


Fig. 5. Tissue N content of *Dictyota dichotoma* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located from along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. Within column — indicates stations with sample sizes too low to include in the ANOVA.

High Nutrient Stations (HNS) are indicated by \*

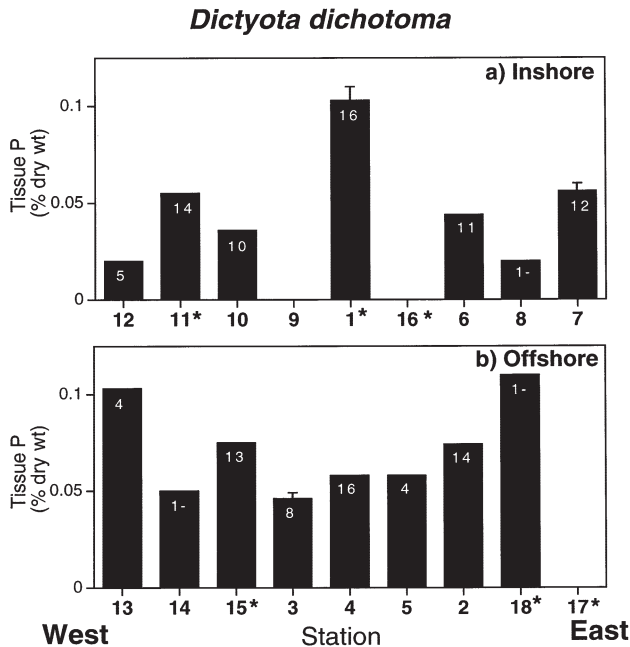


Fig. 6. Tissue P content of *Dictyota dichotoma* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located from along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. Within column -: indicates stations with sample sizes too low to include in the ANOVA. High Nutrient Stations (HNS) are indicated by \*

Tissue P contents of *Halimeda incrassata* (Fig. 8) varied among stations (ANOVA  $p = 0.0001$ ), with a smaller range (0.03 to 0.05 % dry wt) than other species. There was more overlap in means among stations for *H. incrassata* tissue P than for other species (Table 4). Mean P contents in some HNS were ranked high (Stns 11, 1 and 17) and these corresponded to stations with elevated extractable P in sediments. Other HNS ranked

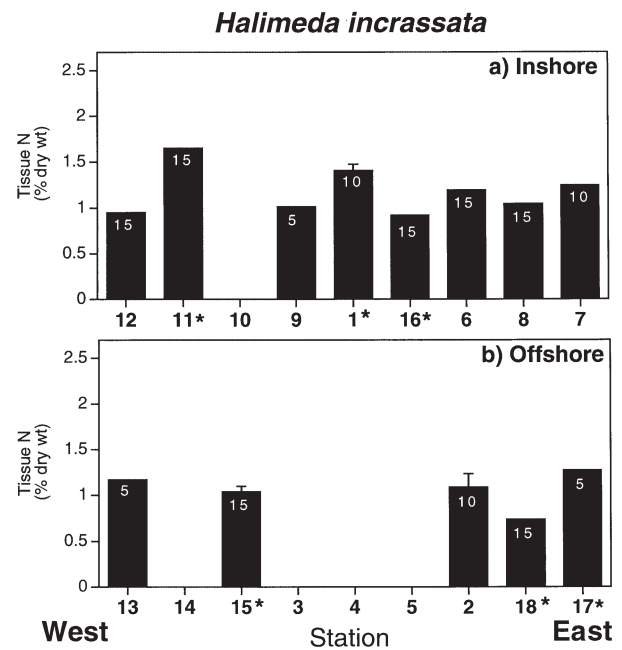


Fig. 7. Tissue N content of *Halimeda incrassata* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located from along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. High Nutrient Stations (HNS) are indicated by \*

intermediate (Stn 15) or low (Stn 18 and 16). Stn 2, off Isla Magueyez, also had high tissue P content for *H. incrassata*, though not for other species of algae. Sediment nutrients were not measured for these stations.

There were significant differences in tissue N of *Dictyota cervicornis* among stations (Table 5; ANOVA  $p = 0.0001$ ); patterns were similar to *D. dichotoma*. Although tissue N contents of samples from Stns 15, 11

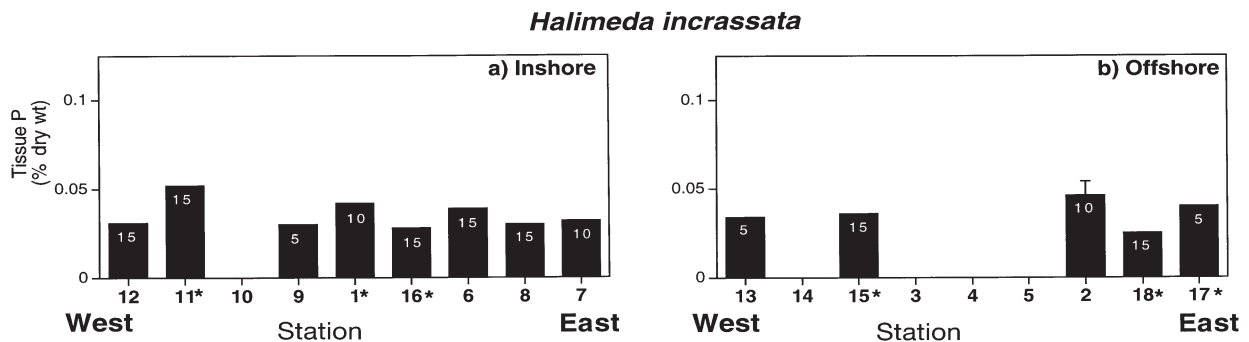


Fig. 8. Tissue P content of *Halimeda incrassata* samples collected from each station. Stations are grouped into (a) inshore and (b) offshore sites. For both inshore and offshore groups, stations are located from along the x-axis from east to west. White numbers within columns represent sample sizes (n) for each station and error bars = SE. High Nutrient Stations (HNS) are indicated by \*



## DISCUSSION

Our results support the hypothesis that tissue nitrogen content of erect algae with open branches is a good indicator of water column nitrogen concentration. *Acanthophora spicifera* had higher N content inshore compared to offshore, reflecting water column TKN and NO<sub>3</sub> concentrations. *A. spicifera* also had higher tissue N content in samples collected at High Nutrient Stations (HNS) with elevated water column nitrogen concentrations. In addition, both water column and tissue nitrogen were high in a station that had not been identified *a priori*, but was just downcurrent of a nutrient source. Although replication was low, *Hypnea musciformis*, another species with open branches, followed the same pattern. Similar results have been found in both laboratory and field studies. In a laboratory experiment, Pedersen & Borum (1997) found that a branching red alga took up nutrients in proportion to water column concentration across a wider range of N supply than slower growing, more fleshy forms of algae. Field studies suggest tissue nutrients in excess of minimum concentrations for growth in branching red algae can be used to assess water quality (Lyngby 1990, Lyngby & Mortensen 1994). Fong et al. (1998) related nutrient supply to the tissue N content of floating mats of green macroalgae tethered along a nutrient gradient in an estuary.

In contrast to the pattern found for tissue N, tissue P of *Acanthophora spicifera* did not reflect water column P concentrations. Although tissue P was higher inshore than offshore, and highest in HNS, water column P concentration did not follow this pattern. Evidence is accumulating that P may limit primary productivity and growth of tropical marine algae in shallow carbonate systems (Lapointe et al. 1987, 1992, Littler et al. 1991). If so, then P may be rapidly depleted from the water by biological uptake as well as adsorption to the sediment, resulting in low water column concentrations despite variability in supply. Fong et al. (1994) found a strong relationship between nutrient supply, water column nutrients, and tissue nutrients for the nutrient (in this case, N) that was not in limiting supply. This suggests that

Table 6. Correlations between water column nutrients, sediment nutrients and tissue N and P. Numbers are R, values in bold are significant ( $p < 0.05$ ). For values denoted with \*  $p$  is between 0.05 and 0.10. Number in parentheses =  $n$

Water column and sediment nutrients			
	Water TKN	Water TP	Water NO <sub>3</sub>
Sediment TKN	0.650* (8)	0.557 (6)	<b>0.771</b> (8)
Sediment TP	<b>0.888</b> (5)	0.053 (4)	0.220 (5)
Sediment P-Olsen	0.704* (8)	0.605 (6)	<b>0.822</b> (8)
Water column nutrients and tissue nutrients			
	Water TKN	Water TP	Water NO <sub>3</sub>
<i>Acanthophora spicifera</i>			
Tissue N	<b>0.525</b> (16)	0.340 (15)	<b>0.759</b> (16)
Tissue P	<b>0.520</b> (16)	0.073 (15)	<b>0.736</b> (16)
<i>Halimeda incrassata</i>			
Tissue N	0.122 (13)	<b>0.589</b> (12)	<b>0.730</b> (13)
Tissue P	0.123 (13)	<b>0.843</b> (12)	<b>0.650</b> (13)
<i>Dictyota dichotoma</i>			
Tissue N	0.107 (15)	0.273 (13)	0.345 (15)
Tissue P	0.061 (15)	0.215 (13)	0.286 (15)
<i>Dictyota cervicornis</i>			
Tissue N	0.141 (10)	0.089 (8)	0.300 (10)
Tissue P	0.068 (9)	0.650 (7)	0.036 (9)
<i>Hypnea musciformis</i>			
Tissue N	0.642 (5)	0.861 (4)	0.764 (5)
Tissue P	0.228 (5)	0.024 (4)	0.805* (5)
Sediment nutrients and tissue nutrients			
	Sediment TKN	Sediment TP	P-Olsen
<i>Acanthophora spicifera</i>			
Tissue N	<b>0.826</b> (8)	0.652 (5)	<b>0.799</b> (8)
Tissue P	0.580 (8)	0.491 (5)	<b>0.768</b> (8)
<i>Halimeda incrassata</i>			
Tissue N	0.529 (6)	0.239 (5)	<b>0.943</b> (6)
Tissue P	0.445 (6)	0.108 (5)	<b>0.917</b> (6)
<i>Dictyota dichotoma</i>			
Tissue N	0.115 (6)	0.937 (3)	0.128 (6)
Tissue P	0.324 (6)	0.860 (3)	0.492 (6)
<i>Dictyota cervicornis</i>			
Tissue N	0.133 (5)	0.995* (3)	0.170 (5)
Tissue P	0.047 (5)	0.784 (3)	0.089 (5)

P may be limiting primary productivity in southwestern Puerto Rico.

Our results provide some evidence to support the hypothesis that rhizophytic algae have higher tissue P where sediment P content is higher. *Halimeda incrassata* had higher tissue P inshore and in stations where extractable P was highest, resulting in a significant correlation between these measures. It has been documented that *Caulerpa cupressoides*, a siphonous green alga with rhizoids, translocates sediment nutrients to photosynthetic portions of the thallus (Williams 1981). Rhizophytic algae from inshore areas of Kaneohe Bay with high sediment nutrient content were able to sus-



tain positive growth despite low water column nutrient concentrations (Larned & Stimson 1997). Others have found that sustained high levels of nutrient loading into a tropical system resulted in enriched sediments and more nutrients being biologically available (McGlathery et al. 1994, Corredor et al. 1999). However, more quantification of sediment nutrients is needed in order for our study to support this hypothesis fully, as we had little data for sediments with low P content. In addition, in the stations where sediment P was low (offshore), *Halimeda incrassata* was absent. Although high tissue P content in *Halimeda incrassata* corresponded with extractable sediment P, tissue N and P content was also related to water column nutrient content. This suggests that *H. incrassata* may also rely on water column nutrient sources. However, high water column and sediment N and P co-varied in HNS, potentially confounding the relationships. It is possible that transplant experiments may aid in understanding the relative importance of these 2 sources of nutrients.

Tissue nutrients in algae that form tightly compacted mats were not higher in HNS, and did not show any patterns relating to any measure of either water column or sediment nutrients. Tissue N and P contents for both species of *Dictyota* were higher offshore, where sediment and water column nutrient concentrations were lowest. In another study, dense mats of a benthic green alga were found to strip all nutrients from the water within the mat, resulting in nutrient depletion of upper layers of the mat (McGlathery et al. 1997). *Dictyosphaeria cavernosa*, a mat-forming species of green algae common in Kaneohe Bay, Hawaii, requires high rates of advection to sustain growth in offshore sites with low water column and sediment nutrient sources (Larned & Atkinson 1997). We suggest that the pattern of higher tissue nutrients in mat-forming algae collected offshore may be explained by higher rates of advection.

Our data suggest N:P ratios alone may not be a good indicator of nutrient supply. The same pattern of higher N:P ratio inshore was found for *Acanthophora spicifera*, *Halimeda incrassata*, and *Dictyota cervicornis*. Using only N:P ratio as an indicator of nutrient supply, one would conclude that N was more plentiful inshore than offshore for all species. However, the source of this inshore/offshore pattern differed among the species of algae. For *A. spicifera* and *H. incrassata*, higher N:P ratios were due to high N contents inshore, supporting the hypothesis of an inshore N source. In contrast, for *D. cervicornis* both N and P contents were higher offshore, suggesting an offshore source of both nutrients. The higher N:P ratio inshore was simply due to the much greater differential between P values compared to N values inshore vs offshore. Thus, N:P ratios must be used in conjunction with tissue values to determine nu-

trient status of macroalgae (Björnsäter & Wheeler 1990, Wheeler & Björnsäter 1992, Fong et al. 1994, 1998).

Our results suggest that the tissue N and P content of macroalgae may be a good indicator of the various sources of nutrients biologically available to primary producers in tropical marine systems. To be a good indicator, tissue N and P content must be a plastic trait that is determined, at least in part, by nutrient availability. This seems to be true for all species collected. Tissue N and P varied between inshore and offshore areas and among specific stations, suggesting nutrient content is controlled by environmental factors. Although a suite of environmental factors such as light and temperature are known to influence the tissue N and P content of macroalgae, our data show that nutrient availability is also very important, as tissue N and P varied with water column and sediment supplies for non mat-forming species. Others have demonstrated that nutrient availability can be estimated by macroalgal tissue N and P content in temperate environments (Lyngby 1990, Lyngby & Mortensen 1994, Fong et al. 1998) provided that variance in light and temperature are carefully controlled. Our findings suggest that if other environmental factors are taken into consideration, tissue N and P contents may prove to be effective indicators in tropical systems as well.

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