

Nutrient distribution across the insular shelf of La Parguera, Puerto Rico: assessment by algal tissue nitrogen

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ABSTRACT.—A survey was conducted to investigate possible presence of an inshore to shelf edge gradient in % nitrogen (%N) and stable nitrogen isotope ratio ($\delta^{15}\text{N}$; ‰ vs. air) in tissues of the rhodophyte *Acanthophora spicifera* and the ocrophytes *Lobophora variegata* and *Dictyota bartayresiana* in coastal waters of La Parguera, southwest Puerto Rico. Average %N (1.26 ± 0.08 SD to 3.25 ± 0.10) and $\delta^{15}\text{N}$ (2.06 ± 0.04 to 8.16 ± 0.14 ‰) in *Acanthophora spicifera* was highly variable along the shoreline. The highest inshore values of $\delta^{15}\text{N}$ occurred at two stations influenced by secondary sewage input and a bird rookery and lower values (0.81 ± 0.06 ‰) were observed at mid-shelf locations. *Dictyota bartayresiana* and *L. variegata* did not display clear trends in %N across the insular shelf; however, $\delta^{15}\text{N}$ for *Dictyota bartayresiana* was significantly higher inshore (3.48 ± 0.09 ‰) versus mid-shelf and shelf edge (0.00 ± 0.08 ‰) and $\delta^{15}\text{N}$ for *L. variegata* was significantly higher at mid-shelf (2.13 ± 0.25 ‰) versus shelf edge locations (0.34 ± 0.24 ‰). These results indicate that anthropogenic effects on tissue nutrients are spatially distributed across the shelf, being higher near shore and not evident at the shelf edge.

INTRODUCTION

The quantity of nutrients available to marine organisms varies both temporally and spatially and is dependent on the presence of localized sources (Larned 1998). Anthropogenic, human derived, sources are often associated with terrestrial development. Sewage, agriculture, and construction are common examples of human contributions leading to increased availability of nutrients in marine ecosystems (Fong *et al.* 2001; Lajtha and Michener 1994). All of these potentially environment altering activities occur in the town of La Parguera, Puerto Rico. West of La Parguera, a sewage treatment plant releases well-nitrified secondary sewage effluents into the adjacent mangrove community (Corredor and Morell 1994) and during the rainy season, September through October, raw sewage frequently leaks directly into the water. Additionally, large areas of land within the watershed are continually being stripped of vegetation as a result of development activities. Thus, the once oligotrophic

coastal waters off La Parguera, with its variable rainfall and anthropogenic inputs, have been exposed to an altered nutrient scenario.

Opportunistic organisms capable of rapid nutrient uptake may be favored in communities affected by nutrient enrichment (Fong *et al.* 1994). Macroalgae, cyanobacteria, and phytoplankton are primary producers that quickly extract nutrients from the water column. Macroalgae are competitive because they possess abilities to rapidly uptake several forms of nutrients simultaneously across their entire surfaces, contain large internal storage pools, and utilize currents to replenish nutrients for greater uptake (Carpenter and Capone 1983). The proposed role of fleshy algae in the decline of reef building corals within areas of coastal eutrophication has been widely discussed (Fong *et al.* 1998; Larned 1998; Littler *et al.* 1991; Miller *et al.* 1999). Kaneohe Bay, Hawaii is the first well documented example of a coral reef shifting to a macroalgal-dominated reef (Banner 1974). Nutrient

enrichment of the bay by domestic sewage, resulted in the overgrowth of coral by the chlorophyte *Dictyosphaeria cavernosa* (Forsskål) Børgesen. Szmant (2002) agreed that in many areas, anthropogenic impacts have increased the flux of nutrients into coastal waters. However, she cautions that various cases of coral reef degradation may be attributed to factors such as previous coral bleachings, massive herbivore die-offs, and a history of over-fishing because these factors increase the availability of substrata for algal colonization or increase the severity of effects from nutrient enrichment. Nevertheless, increasing eutrophication in addition to changing herbivory patterns and coral diseases, contribute to a synergism resulting in coral reef deterioration.

A predictive indicator of bioavailable nutrient enrichment that provides an accurate and early warning to changes in the nutrient conditions has become highly desirable for management of sensitive coral reef ecosystems (Costanzo *et al.* 2000; Fong *et al.* 1998; Larned 1998). Historically, the principle method for measuring nutrient availability has been via water column concentrations of ambient nitrogen (N) and phosphorous (P). Recent literature suggests that this method alone may not be an accurate indicator of nutrient availability because ambient N and P concentrations in the water column fail to indicate the fraction which is bioavailable (Maher and Norris 1990) and do not necessarily correlate with production or abundance of primary producers (Fong *et al.* 1998; Larned 1998). Not all nutrients reaching the marine environment can be utilized metabolically. Approximately 80% of the total dissolved nitrogen (N) in urban/suburban runoff and 20-60% from forests and pastures is bioavailable (Seitzinger *et al.* 2002). The rapid uptake ability of macroalgae and the close recycling of nutrients within coral reef ecosystems also diminish the effectiveness of water column concentrations as nutrient enrichment indicators (Fong *et al.* 1998; Fong *et al.* 2001).

Nutrients from anthropogenic sources are usually supplied in pulses to inshore waters by runoff and periodic sewage leaks.

Water quality fluctuations were observed by Otero and Carbery (2005) in La Parguera evidencing the presence of transient peaks of nutrient levels in the water column. Within a community of opportunistic autotrophs which rapidly strip the water of nutrients, nutrients cannot accumulate unless the loading rates are extremely high, thus direct periodic sampling of the water column will often under or overestimate the nutrient conditions, dependent on whether the "peaks" are hit or missed (Costanzo *et al.* 2000; Fong *et al.* 1994). The nitrogen concentration within tissue of macroalgae may be a more useful indicator of nutrient availability because particular species respond to nutrient enrichment by taking in nutrients, growing, and storing excess (Fong *et al.* 1998). Costanzo *et al.* (2000) reported that water column concentrations of nutrients returned to ambient conditions rapidly after a pulse, but the effects on the tissue nitrogen content of *Gracilaria sp.* remained for days. This indicates that brief nutrient pulses which may not be detected by traditional sampling methods, nevertheless continue to influence biological activities after the water column nutrient event has passed.

Macroalgal tissue is responsive to changes in the amount of nutrients and integrates the changes over a period of time. Species specific maximum uptake volume, determined by the size of storage vacuoles and rates of assimilation, determine the period of time over which algae can integrate the response. Using algal tissue nutrients as bioindicators of enrichment allows for less frequent sampling than would be necessary for the same accuracy in direct water column sampling (Fong *et al.* 1994). Algal tissue nutrients may also be effective in identifying the location of the nutrient source because the percent change in tissue nitrogen concentration measured among several locations allows for the assessment of a net increase or decrease in nutrient enrichment between these areas (Fong *et al.* 1998).

While studies dealing with the quantity of nutrients introduced into coastal waters have focused on total %N within algal tissue, fewer studies on the source

of nutrients have evaluated tissue $\delta^{15}\text{N}$. The stable nitrogen isotope signature ($\delta^{15}\text{N}$) in algal tissue can be used to determine local sources of enrichment because terrestrial sources of nutrients generally produce higher $\delta^{15}\text{N}$ values than ambient seawater and rainwater (McClelland and Valiela 1998; Umezawa *et al.* 2007). This difference is created during physical and chemical processes where a selection against or for a particular isotope occurs (i.e., ^{15}N versus ^{14}N). Agricultural practices increase $\delta^{15}\text{N}$ by increasing oxidation of soil organic N (Kreitler and Browning 1983). Denitrification performed by many soil inhabiting microbes results in an increase of $\delta^{15}\text{N}$ in the soil that may be transported to the ocean in runoff (McClelland and Valiela 1998). Costanzo *et al.* (2001) performed experiments using levels of $\delta^{15}\text{N}$ in marine plants to map the effects of multiple sewage outfalls on an Australian bay. High values of $\delta^{15}\text{N}$ (ca. 10‰) were found around river mouths and decreased with distance from the source. Measurements greater than 3‰ were described as having been influenced by sewage.

In order to use algae to identify sources of nutrient enrichment, a low level of fractionation within the alga is desired to avoid altering the $\delta^{15}\text{N}$ signal of the source. Alteration in $\delta^{15}\text{N}$ of the source will not be observed if algal kinetics are unsaturated and adequate agitation of the algal tissue boundary layer occurs (Lajtha and Michener 1994). Thus, in a tropical marine environment where N is considered a limiting nutrient and moderate water energy is observed, it is likely that algae do not fractionate N to a large degree and thus can be used as bioindicators of $\delta^{15}\text{N}$ in coastal waters (Cohen and Fong 2005; Lajtha and Michener 1994).

Measurements of algal tissue nutrients can provide information about nutrient conditions in the marine environment. Tissue total %N indicates the quantity of nutrients available and $\delta^{15}\text{N}$ identifies potential sources of the nutrients. Thus, objectives of this study were to determine: algal tissue %N and $\delta^{15}\text{N}$ of benthic marine algae in the vicinity of La Parguera, possible cross-shelf patterns in distribution of %N and $\delta^{15}\text{N}$ in

algal tissue, and candidate species for indicators of nutrient inputs.

METHODS

La Parguera is located on the southwest coast of Puerto Rico (17°57'N, 67°02'W). A range of hills within 2 km of the coast isolates the local watershed from farther inland sources. Mangrove forests occupy much of the coastline as well as colonize small cays leeward of patch reefs in the inner and mid-shelf. The insular shelf extends approximately 10 km from shore, reaches an average depth of 20 m and supports spur and groove reef formations along the perimeter. The prevailing currents flow east to west and a sewage treatment plant is located less than 1 km downstream from the town center of La Parguera. A total of 21 sites within the La Parguera region were included in this study (Fig. 1). Sampling was conducted within three consecutive days starting on July 11th 2007. No rainfall occurred in La Parguera for 21 days prior to sampling (Caricom data provided by Ernesto Weil, personal communications).

Three algal species were selected for nutrient analysis on the basis of possessing a relatively high capacity for N storage within tissue (Larned 1998), year-round abundance and broad distributions across the insular shelf. Thus this work focused on *Dictyota bartayresiana* Lamouroux (Ocrophyta) which possessed the broadest cross-shelf distribution, being common from adjacent to shore to the edge of the insular shelf; *Lobophora variegata* (Lamouroux) Womersley ex Oliveira (Ocrophyta) which is common in habitats of various light and depth conditions at mid-shelf and outer-shelf locations; and *Acanthophora spicifera* (Vahl) Børgesen (Rhodophyta) which has a rapid growth rate and prevalence from near shore to mid-shelf reefs. *Acanthophora spicifera* was additionally selected because total tissue N of red algae has been demonstrated to have closer correlation to water column concentration than the tissue of green or brown algae (Bird *et al.* 1982; Björnsäter and Wheeler 1990; D'Elia and DeBoer 1978; Jones *et al.* 1996). All collected algae were neither

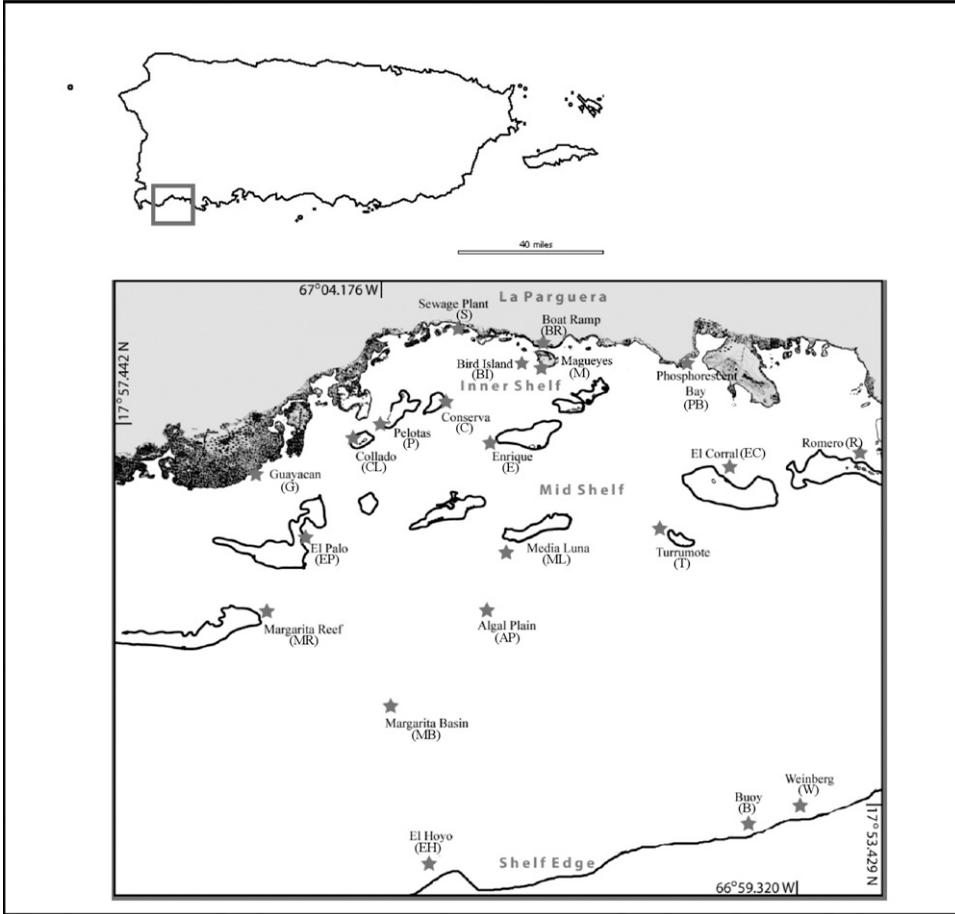


FIG. 1. Map of Puerto Rico and La Parguera study sites.

in layered mats nor growing in shaded habitats as organisms that are not continuously undergoing a high rate of photosynthesis have an increased opportunity for storage of nutrients (Fong *et al.* 2001).

Acanthophora spicifera occurred at 12 sites across the inshore and mid-shelf reefs. *Dictyota bartayresiana* occurred at 16 sites reaching across the inshore, mid-shelf and edge of the insular shelf and *Lobophora variegata* occurred at 8 sites from the mid-shelf to the edge of the insular shelf. Where available, at least six replicates of each alga were collected. Distance between replicates varied among sites due to frequency of species occurrence. In most cases, replicates were at least 5 m apart.

Algal samples were collected by SCUBA and snorkeling, transported back to lab in

seawater-filled plastic bags kept on ice, briefly rinsed in fresh water to remove salts and nutrients from the surface (Fong *et al.* 1994, 1998, 2001; Jones *et al.* 1996), blotted with paper towels to remove excess water, placed in glass scintillation vials, and stored at -20°C until freeze-dried (Virtis Model 25SL Freeze Drier). Following freeze drying, algae were ground with a mortar and pestle (Cohen and Fong 2005). Samples were analyzed for total %N and $\delta^{15}\text{N}$ utilizing a CHN analyzer coupled to an isotope-ratio mass spectrometer at the Cornell University Isotope Laboratory and the University of California, Davis Stable Isotope Facility. Delta ^{15}N is defined as:

$$\delta^{15}\text{Nitrogen} (\%) = [(R_{\text{sample}}/R_{\text{air}}) - 1] \times 10^3$$

where R is equal to the atomic $^{15}\text{N}/^{14}\text{N}$ ratio. Non parametric multiple comparison analysis with t-distribution was performed using the Unistat 5.5 statistical package for Excel to distinguish significantly different values among sites. Unless otherwise stated, significance was at the 5% level.

Over 10 cm of rainfall in the days following the sampling period (Caricomp data provided by Ernesto Weil, personal communications) allowed for a unique opportunity to test for change in algal tissue nutrient concentration resulting from runoff. *Acanthophora spicifera* was sampled on July 20th, 2007 at both Magueyes and a nearby boat ramp to compare nutrient concentration pre- and post-rain. Single-factor ANOVA was used to detect significance in pre- and post-rain differences at both sites.

RESULTS

Inshore measurements of %N in *Acanthophora spicifera* varied greatly ($1.27\% \pm .17$

to $3.25\% \pm .24$) (Table 1). Higher values were associated with well developed mangrove stands and lower values corresponded with lesser developed mangrove areas. Percent N means at mid-shelf locations were lower than most means along the shoreline; however, multiple comparison analysis formed 10 subsets of nonsignificantly different sites (Fig. 2) and three of these subsets contain locations from both the shoreline and mid-shelf.

Percent N of *Dictyota bartayresiana* and *Lobophora variegata* also did not display clear trends across the insular shelf (Table 1). Four of the 5 nonsignificantly different subsets for *D. bartayresiana* included locations near the shoreline and at the shelf edge (Fig. 3). *Lobophora variegata* produces values with a similar lack of a gradient. Two of the 4 nonsignificantly different subsets included locations at the mid-shelf and shelf edge (Fig. 4). The lowest value of *L. variegata* ($0.64\% \pm .07$) was found at a mid-shelf location.

Similar to %N, $\delta^{15}\text{N}$, inshore measurements of *Acanthophora spicifera* varied greatly

TABLE 1. Algal tissue % Nitrogen and $\delta^{15}\text{N}$ Nitrogen for *Acanthophora spicifera*, *Dictyota bartayresiana* and *Lobophora variegata* across the insular shelf at La Parguera, Puerto Rico.

Shelf Location	Station	% Nitrogen			$\delta^{15}\text{N}$ Nitrogen		
		<i>Acanthophora spicifera</i>	<i>Dictyota bartayresiana</i>	<i>Lobophora variegata</i>	<i>Acanthophora spicifera</i>	<i>Dictyota bartayresiana</i>	<i>Lobophora variegata</i>
Inshore	Phosphorescent Bay	$1.58 \pm .27$	$2.08 \pm .39$		$3.05 \pm .21$	$1.52 \pm .22$	
	Bird Island	$2.67 \pm .41$	3.08		$5.91 \pm .48$	7.71	
	Magueyes	$1.68 \pm .55$	$1.72 \pm .18$		$3.96 \pm .62$	$3.48 \pm .09$	
	Boat Ramp	$2.09 \pm .36$	$1.89 \pm .44$		$3.42 \pm .63$	$3.03 \pm .54$	
	Sewage Plant	$2.29 \pm .27$			$8.16 \pm .34$		
	Guayacan	$2.80 \pm .22$			$2.06 \pm .10$		
Mid-Shelf	Romero	$1.27 \pm .17$	$1.87 \pm .17$		$2.60 \pm .09$	$2.40 \pm .12$	
	El Corral		$1.26 \pm .31$			$2.30 \pm .19$	
	Enrique	$1.52 \pm .21$	$1.83 \pm .19$		$2.32 \pm .23$	$2.02 \pm .14$	
	Conserva	$1.35 \pm .11$	$1.78 \pm .26$		$1.77 \pm .22$	$1.77 \pm .27$	
	Pelotas	$3.25 \pm .24$	$2.72 \pm .38$		$2.35 \pm .18$	$2.72 \pm .38$	
	Collado	$3.09 \pm .54$			$2.37 \pm .45$		
	Turumote		$1.44 \pm .40$			$1.71 \pm .21$	
	Media Luna	$1.38 \pm .15$	$1.88 \pm .27$	$1.37 \pm .20$	$2.06 \pm .12$	$1.90 \pm .40$	$1.85 \pm .44$
	El Palo	$1.71 \pm .13$	$2.00 \pm .25$		$1.86 \pm .24$	$1.84 \pm .23$	
	Algal Plain	0.81 ± 0.6		$0.64 \pm .07$	$2.05 \pm .20$		$2.13 \pm .25$
	Margarita Reef		$2.13 \pm .25$			$1.67 \pm .18$	
Margarita Basin		$1.68 \pm .12$	$1.22 \pm .05$		$1.02 \pm .13$	$1.49 \pm .11$	
Shelf Edge	Weinberg			$1.16 \pm .11$			$0.50 \pm .21$
	Buoy		$2.23 \pm .20$	$1.35 \pm .11$		$0.00 \pm .08$	$0.35 \pm .24$
	El Hoyo		$1.67 \pm .18$	$1.23 \pm .14$		$0.22 \pm .17$	$0.70 \pm .33$

(1.77‰ ± 0.22 to 8.16‰ ± .34) (Table 1). However, a reverse trend was observed with higher values associated with lesser developed mangrove areas (including the town of La Parguera) and lower values corresponding with well developed mangrove stands. The highest inshore values of δ¹⁵N occurred at two stations influenced by secondary sewage input (Sewage Plant: 8.16‰ ± 0.34) and a bird rookery (Bird Island: 5.91‰ ± 0.48). The seven nonsignificantly

different subsets created by multiple comparison analysis show a clear progression from higher δ¹⁵N values along the eastern portion of the shoreline to lower values at mid-shelf and the western portion of the shoreline (Fig. 5). A single anomaly to this trend occurred at the inshore location Conserva, where the lowest δ¹⁵N value (1.77‰ ± 0.22) was found.

Delta ¹⁵N ratios in *Dictyota bartayresiana* and *Lobophora variegata* also display a

Site	AP	R	C	ML	E	PB	M	EP	BR	S	BI	G	CL	P
%N	.81	1.27	1.35	1.38	1.52	1.58	1.68	1.71	2.09	2.29	2.67	2.80	3.09	3.25

FIG. 2. Nonsignificantly different subsets produced by multiple comparison analysis of %N in tissue of *Acanthophora spicifera* across the insular shelf (See Fig. 1 for location of sites).

Site	EC	T	EH	MB	M	C	BR	E	ML	R	EP	PB	MR	B	P	BI
%N	1.26	1.44	1.67	1.68	1.72	1.78	1.89	1.83	1.88	1.87	2.00	2.08	2.13	2.23	2.72	3.08

FIG. 3. Nonsignificantly different subsets produced by multiple comparison analysis of %N in tissue of *Dictyota bartayresiana* across the insular shelf (See Fig. 1 for location of sites).

Site	AP	W	MB	EH	ML	B
%N	0.64	1.16	1.22	1.23	1.37	1.35

FIG. 4. Nonsignificantly different subsets produced by multiple comparisons analysis of %N in tissue of *Lobophora variegata* across the insular shelf (See Fig. 1 for location of sites).

Site	C	EP	AP	G	ML	E	P	CL	R	PB	BR	M	BI	S
δ ¹⁵ N	1.77	1.86	2.05	2.06	2.06	2.32	2.35	2.37	2.60	3.05	3.42	3.96	5.91	8.16

FIG. 5. Nonsignificantly different subsets produced by multiple comparison analysis of δ¹⁵N in tissue of *Acanthophora spicifera* across the insular shelf (See Fig. 1 for location of sites).

clear trend across the insular shelf (Table 1). Nine subsets of *D. bartayresiana* indicate a decrease in $\delta^{15}\text{N}$ from shoreline to shelf edge (Fig. 6). Two anomalies to the pattern exist at near shore locations, Conserva and Phosphorescent Bay, where values are similar to those at mid-shelf locations. The four subsets of *L. variegata* follow a similar decreasing trend from mid-shelf to shelf edge (Fig. 7).

Mean post-rain %N increased non significantly from 1.68 to 1.87 ($p=.53$) at Magueyes and decreased non significantly from 2.09 to 2.05 ($p=.81$) at Boat Ramp. Mean post-rain $\delta^{15}\text{N}$ increased from 3.96 to 4.55 ($p=.14$) at Magueyes but was only significant at Boat Ramp where it increased from 3.42 to 4.31 ($p=.022$).

DISCUSSION

Despite high variability, %N in *Acanthophora spicifera* tissue indicates a weak nitrification pattern across the inner and mid-shelf; however, *Dictyota bartayresiana* and *Lobophora variegata*, are also variable for %N and do not clearly indicate cross shelf patterns of nitrification. High values of %N for *A. spicifera* in developed mangrove stands may be due to higher rates of productivity in the immediate area,

including a large invertebrate community that is capable of supplying nutrients in the form of excretory wastes (Bracken *et al.* 2007). Nitrogen recycling within the mangrove community may also be responsible for the elevated observed values (Carpenter and Capone 1983). The lower values found in some mid-shelf locations may indicate lower nutrient concentrations. The range of %N in *A. spicifera* reported in this study (0.81-3.25%) is larger than that reported by Fong *et al.* (2001) for approximately the same study area (1.10-2.67%).

Several explanations may clarify the marked variability of %N in *Dictyota bartayresiana* and *Lobophora variegata* across the insular shelf. The first is that the pattern is representative of truly variable nutrient conditions across the insular shelf. This lack of a trend in %N can also occur if algae are N limited and thus can not accumulate N in storage vacuoles, preventing a reflection of different nutrient regimes. Secondly, Otero (unpubl.) indicated that previous experimentation on nutrient limitation within the sampling area indicated a co-limitation of nitrogen, phosphorus, and iron and these limitations may be responsible for the variations of algal tissue %N. A third possibility is saturation of N in algal tissue across the insular shelf. Within

Site	B	EH	MB	PB	MR	T	C	EP	ML	E	EC	R	P	BR	M	BI
$\delta^{15}\text{N}$	0.00	0.22	1.02	1.52	1.67	1.71	1.77	1.84	1.90	2.02	2.30	2.40	2.72	3.03	3.48	7.71

FIG. 6. Nonsignificantly different subsets produced by multiple comparison analysis of $\delta^{15}\text{N}$ in tissue of *Dictyota bartayresiana* across the insular shelf (See Fig. 1 for location of sites).

Site	B	W	EH	MB	ML	AP
$\delta^{15}\text{N}$	0.35	0.54	0.70	1.49	1.85	2.13

FIG. 7. Nonsignificantly different subsets produced by multiple comparison analysis of $\delta^{15}\text{N}$ in tissue of *Lobophora variegata* across the insular shelf (See Fig. 1 for location of sites).

a system experiencing low levels of N, sufficient water energy may provide access to more N than can be taken up or stored in internal storage vacuoles (Larned and Atkinson 1997). Nitrate releasing cyanobacterial symbionts living within Caribbean reef sponges can also provide a significant amount of nitrate to algae (Corredor *et al.* 1988). Percent N means reported here for *D. bartayresiana* are similar to means for *D. cervicornis* and *D. dichotoma* reported by Fong *et al.* (2001) for the coastal waters of La Parguera, which were approximately 1.5% and 1.6% for *D. cervicornis* Kütz. and *D. menstrualis* (Hoyt) Schnetter, Hornig & Weber-Peukert (= *D. dichotoma* in Fong *et al.* op cit.) respectively. The samples, taken in January 1998, depicted higher %N offshore compared to inshore for both *D. cervicornis* and *D. dichotoma*, a trend opposite to that of all other species examined. Results of the present study and those of Fong *et al.* (2001) suggest that algal tissue %N in *Dictyota spp.* is not a clear indicator of nutrient loading in La Parguera. However, Fong *et al.* (2001) did find a decrease in %N from the shoreline to mid-shelf in the tissue of other algal species, including *Acanthophora spicifera*. Their sampling took place in January, a month typically characterized by calm seas and low wind speeds. July, the month sampled in the current study was very windy, with large swells and currents. Increased water energy provided in July could elevate nutrient flux from sediments or sufficiently increase the supply of N to algal cells, effectively saturating intercellular stores, and thus disrupting any spatial detectable nutrient trends within the water column.

Gradients in $\delta^{15}\text{N}$ and %N do not necessarily co-fluctuate in the marine environment. This is to be expected because high values of %N can occur with low values of $\delta^{15}\text{N}$ (-2 to 2‰) when ammonium based fertilizers are used for farming. Low values of %N can occur with high values of $\delta^{15}\text{N}$ (10-20‰) when sewage effluents become diluted in aquifers or other large masses of water (McClelland and Valiela 1998). The latter scenario may explain the presence of a $\delta^{15}\text{N}$ gradient occurring with a lack of a %N gradient in the La Parguera region.

Algal tissue $\delta^{15}\text{N}$ values reported in this study range from -0.39 to 8.05‰. This represents the combined $\delta^{15}\text{N}$ signature of all algal intracellular nitrogen. Trends of $\delta^{15}\text{N}$ for *Acanthophora spicifera*, *Dictyota bartayresiana*, and *Lobophora variegata* indicate significant anthropogenic impacts in spatially limited areas along the developed shoreline (up to 8.05‰), moderate to low effects at mid-shelf (2-3‰), and no detectable effect at the shelf edge (< 2‰). The highest $\delta^{15}\text{N}$ values for *A. spicifera* identify two potential point sources of allochthonous nutrients; a sewage treatment plant and a bird rookery inhabited primarily by cattle egrets, land feeding birds.

In the present study, $\delta^{15}\text{N}$ provides a more defined nutrient trend than %N across the insular shelf of La Parguera. The trend defined by $\delta^{15}\text{N}$ does not indicate the quantity of nutrients in the water, as will %N, but implies that anthropogenic impacts are strongest at locations along the shoreline and may not reach the local shelf edge. In areas where algae are not N limited, $\delta^{15}\text{N}$ would probably provide better evidence for allochthonous nutrient inputs than %N.

During and after the rain event, sediment plumes in the area of Boat Ramp were evident for more than 24 hours, however, measures of %N did not indicate an increase in nutrient input. It is possible that N stores within the algae were already saturated from previously high nutrient concentrations at both Boat Ramp and Magueyes. Measures of $\delta^{15}\text{N}$ indicate a significant increase in allochthonous N at Boat Ramp but not at Magueyes. Algae sampled from Magueyes are located in an area shielded from the visible sediment plume by a land mass and currents that direct the plume away from the site while algae at the boat ramp are directly exposed to run-off and potential sewage spills. In this particular sampling event, $\delta^{15}\text{N}$ was also a better indicator than %N of new nutrient influx. Delta $\delta^{15}\text{N}$ may perform better as an indicator of nutrient pulses when algae are sampled from locations often saturated with N.

In conclusion, high levels of algal tissue $\delta^{15}\text{N}$ (ca. 8‰) in the inshore waters of La Parguera are indications of moderate anthropogenic influence that may be originating

from sewage input and/or land development activities. Low levels of $\delta^{15}\text{N}$ (less than 0‰) in algal tissues observed at the edge of the insular shelf are typical of nutrient depleted oceanic water (Kreitler 1979), demonstrating that the influence of anthropogenic effects visible in inshore waters did not have an effect on offshore waters. Results from this study strongly support the use of algal tissue $\delta^{15}\text{N}$ as an indicator of anthropogenic influence in coastal waters. Conclusions presented here are based on a limited sampling period. Future sampling should be designed to take into account seasonal differences in environmental conditions.

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