EUTROPHICATION AND TROPHIC STATE CLASSIFICATION OF SEAGRASS COMMUNITIES IN THE FLORIDA KEYS

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ABSTRACT

Seagrass communities in the Florida Keys are receiving increased nutrient loadings from a variety of land-based human activities that are accelerating coastal eutrophication. We assessed relationships among total nitrogen (TN) and total phosphorus (TP) concentrations of the water column and the productivity, biomass, and epiphyte levels of the seagrasses *Thalassia testudinum* and *Halodule wrightii* along three onshore-offshore transects (Key West, Big Pine Key, and Long Key) stratified a priori into hypereutrophic (HYPER), eutrophic (EUTR), mesotrophic (MESO), and oligotrophic (OLIGO) communities with increasing distance from shore. Macroalgal biomass and alkaline phosphatase activity (APA) of macroalgae and attached seagrass epiphytes were also determined along the eutrophication gradients. *H. wrightii* was the dominant seagrass within inshore HYPER strata whereas *T. testudinum* was dominant at the EUTR, MESO and OLIGO strata. Seagrasses at the HYPER and EUTR strata had low shoot densities, low shoot production rates, low areal biomass values, low areal production rates, but high levels of attached epiphytes and mat-forming macroalgae. Seagrasses at the OLIGO strata had the highest shoot densities, highest areal biomass values, highest areal production rates, and typically the lowest or second lowest epiphyte levels of all strata. APA was lowest for macroalgae at the offshore OLIGO strata, and highest at the nutrient-enriched HYPER and EUTR strata where extensive populations of mat-forming macroalgae occurred. Microcosm studies showed that both N and P enrichment increased epiphyte levels and reduced rhizome growth rates in *T. testudinum* whereas P enrichment alone increased epiphyte levels and reduced rhizome growth rates of *H. wrightii.* Higher APA in macroalgae and attached blade epiphytes in HYPER and EUTR strata reflected increased P-limitation in these dystrophic environments resulting from high concentrations of TN relative to TP. Sustained nutrient enrichment from land-based activities results in increased biomass of attached epiphytes and macroalgae, which attenuate light, reduce dissolved oxygen, and lead to decline of *T. testudinum* and a gradient of habitat damage from nearshore to offshore waters.

Cultural eutrophication is the most frequently cited factor correlating with the marked global decline in areal extent and vigor of seagrass communities over the past two decades (Larkum, 1976; Kemp et al., 1983; Cambridge and McComb, 1984; Orth and Moore, 1984; Bourcier, 1986; Silberstein et al., 1986; Valiela et al., 1990; Giesen et al., 1990; Tomasko and Lapointe, 1991). The negative effects of cultural eutrophication result largely from nuisance algal blooms, both phytoplankton and macroalgae, which reduce light availability to seagrasses. Studies in Denmark (Borum, 1985), Australia (Silberstein et al., 1986), Mexico (Flores-Verdugo et al., 1988), Chesapeake Bay, USA (Kemp et al., 1983), Texas, USA (Dunton, 1990), and Florida, USA (Jensen and Gibson, 1986; Tomasko and Lapointe, 1991) have shown that increased water column nutrient concentrations result in greater algal epiphyte levels on seagrass blades. Nutrient-mediated increases in epiphyte and phytoplankton biomass increase light limitation of seagrasses (Sand-Jensen, 1977; Twilley et al., 1985; Silberstein et al., 1986), suggesting that chronic enrichment will decrease seagrass productivity and stress these habitats by hypoxia or anoxia. Other stress mechanisms to seagrasses may also be involved, such as direct toxicity effects due to elevated nitrate loading (Burkholder et al., 1992).

Human activities on land inevitably increase nutrient inputs to coastal waters

(Pierels et al., 1991; Turner and Rabalais, 1991), accelerating the problems associated with coastal eutrophication. Nutrient enrichment repeatedly has been identified as the single most important problem impacting nearshore waters of the Florida Keys (NOAA, 1988; Lapointe et al., 1990; Lapointe and Clark, 1992). Domestic wastewater is a major near-field source of nutrient inputs and enters coastal waters via several routes, including submarine groundwater discharge enriched by septic tanks, cesspits, and shallow injection wells (Lapointe et al., 1990). Direct surface water inputs are also associated with sewage outfalls in the Keys, including the 7 MGD City of Key West outfall; approximately 3,200 live-aboard boats discharge untreated wastewater directly into surface waters (FDER, 1987). Far-field sources of nutrients can also contribute to eutrophication in the Florida Keys region, including industrial, agricultural and domestic wastewater runoff from the south Florida mainland (Lapointe and Clark, 1992). Rivers along the entire southwest coast of Florida have phosphorus concentrations substantially higher than most rivers in North America due to high natural background levels and runoff from extensive phosphate mining (Odum, 1967). Anthropogenic nutrient inputs to the Everglades have increased over the past three decades and most nitrogen taken up by plants and microbes appears to be converted to ammonium and exported (Gordon et al., 1986; Reddy et al., 1993). Satellite imagery has shown the presence of Mississippi River water in the vicinity of the Florida Keys (Muller-Karger et al., 1991), suggesting that watershed sources as distant as the mid-western United States could conceivably contribute to eutrophication in the Florida Keys.

The purpose of our study was to assess how nutrient enrichment within the Florida Keys mediates algal epiphyte biomass, productivity, and the structure of shallow, benthic communities historically dominated by *Thalassia testudinum*. This approach provides an objective trophic state classification system for tropical seagrass communities based on known relationships between water column nutrient concentrations and epiphyte levels; by establishing these relationships in the field, seagrass communities can be used to assess water quality "integrated" over time and thus can define long term seagrass habitat requirements. This approach is similar to that of Dennison et al. (1993) who recently assessed the health of Chesapeake Bay using the habitat requirements of submersed aquatic vegetation. The specific objectives of our study included: 1) characterizing seagrass community trophic states along a eutrophication gradient by total water column nutrient concentrations and dawn dissolved oxygen, 2) determining in situ relationships among water column nutrient concentrations, biomass of mat-forming macroalgae and attached epiphytes, the degree of P-limitation of epiphytic macroalgae, and the biomass, density, and areal productivity of the seagrasses *T. testudinum* (turtle grass) and *Halodule wrightii* (shoal grass) in the different trophic states, and 3) experimentally investigating the effects of nitrogen (N) and phosphorus (P) enrichment on epiphyte growth and below-ground productivity of *T. testudinum* and *H. wrightii* in flowing seawater microcosms.

MATERIALS AND METHODS

Field Studies: Design and Data Collection.—We used the stratified-random sampling technique described by Littler and Littler (1984a) to determine locations of different seagrass trophic states, referred to as strata, along three onshore-offshore transects perpendicular to shore in the middle and lower Florida Keys (Fig. 1; Table 1). The four trophic strata along the three transects were defined a priori as hypereutrophic (HYPER), eutrophic (EUTR), mesotrophic (MESO), and oligotrophic (OLIGO) based on posited relationships between water column nutrient concentrations and macroalgal epiphyte levels on seagrasses. The HYPER strata were located at the inshore origin of all three transects and received direct impacts of wastewater nutrient discharges; nutrient sources at the HYPER strata in-
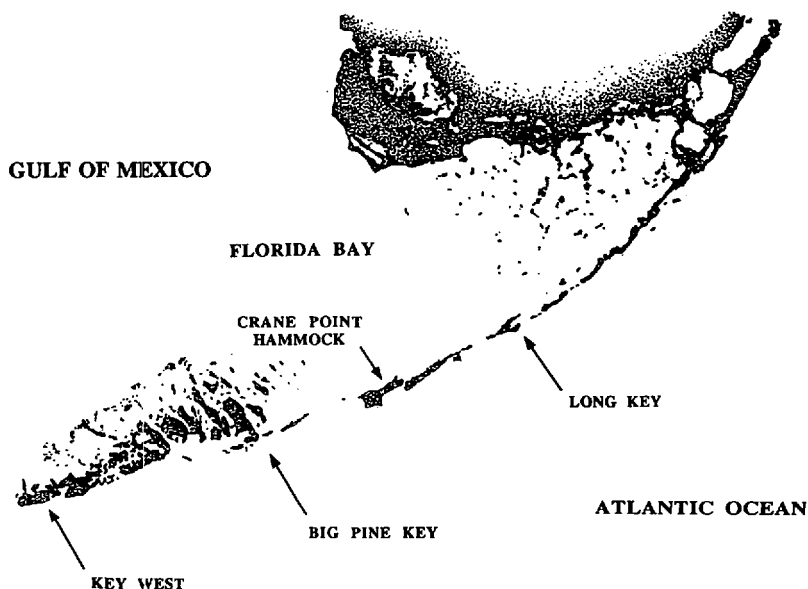
Figure 1.   Map of the Florida Keys region showing locations of sampling transects (at Key West, Big Pine Key, and Long Key) and Crane Point Hammock facilities in Marathon.

cluded live-aboard boats and stormwater runoff (Houseboat Row, Key West transect), septic tanks and cesspits (Doctor's Arm subdivision, Big Pine Key transect), and a package sewage treatment plant and surface water outfall (Fiesta Key Campground, Long Key transect). Our prior research demonstrated that dissolved nutrient concentrations in the water column, known to stimulate epiphytic macroalgal growth in the Florida Keys (Lapointe, 1987, 1989; Tomasko and Lapointe, 1991), decrease with increasing distance from shore yet occur at significant concentrations at the offshore bank reef sites (Lapointe and Clark, 1992); accordingly, the OLIGO strata were located in lagoon habitats at three bank reef sites (Alligator Reef off Long Key, Looe Key National Marine Sanctuary off Big Pine Key, and Sand Key off Key West) to ensure relatively low nutrient concentrations. Because the intent of our study was to assess the effects of elevated nutrient concentrations and epiphytes on seagrass productivity, the EUTR and MESO strata were nested within ~1 km of land; EUTR strata were chosen as areas with high coverage of mat-forming macroalgal epiphytes (e.g., >50% cover) overlying sea-grasses, whereas MESO strata had lower coverage (<20% cover) of mat-forming macroalgae but significant amounts of filamentous microalgal epiphytes attached to the seagrass blades.

Seagrass communities along the transects were randomly sampled within each trophic stratum at 1.5–2.0 m water depth (MLW) to avoid confounding factors from variable water depth. In addition, sediment depths were determined to be >50 cm at all strata. Various seagrass and water quality parameters (detailed below) were measured at the strata over a three week period during summer 1991 (September) and winter 1992 (February) to assess seasonal variability in relationships among nutrient concentrations, epiphyte biomass and seagrass productivity.

WATER QUALITY. Dissolved oxygen (DO), temperature, and salinity were measured at dawn using a Hydrolab Surveyor II calibrated (per manual using Winkler titration and NBS conductivity standards) to determine the minimum critical daily DO values (Lapointe and Clark, 1992). Surface and bottom measurements were averaged for each of the four strata along the three transects. Water samples from all strata were collected by hand into clean 250 ml Nalgene bottles for determination of water column nutrient concentrations. Concentrations of total N (TN) and total P (TP) were determined on a Tech-nicon Autoanalyzer II according to standard Technicon Industrial methodology using persulfate di-gestion prior to analysis of TN (D'Elia et al., 1977) and TP (Menzel and Corwin, 1965).

SEAGRASS MEASUREMENTS. Seagrass density was measured in the field studies using 25 × 25 cm quadrats placed at 10 random locations within each stratum, which generally results in coefficients of variation of less than 25% of the mean when N = 10 (Tomasko and Dawes, 1989; Tomasko and Lapointe, 1991). Species were identified in situ and numbers of shoots were determined in the field for *T. testudinum*. A 10 cm diameter core was used for *H. wrightii*, as densities were much higher for this species (Dunton, 1990; Tomasko and Dawes, 1989, 1990; Tomasko and Lapointe, 1991).

Table 1. Locations and nutrient concentrations (μM) of four trophic strata along three transects. Values represent means ± 1 SD (N = 6).

			Hypereutrophic				Eutrophic	
	Lat./Long.	Nutrient	Summer	Winter	Lat./Long.	Nutrient	Summer	Winter
Key West	24°33'30.1"N	Total N	21.29 ± 2.49	23.07 ± 2.28	24°33'20.3"N	Total N	14.40 ± 1.98	38.26 ± 4.52
	81°44'56.9"W	Total P	0.34 ± 0.18	0.37 ± 0.12	81°44'52.3"W	Total P	0.40 ± 0.09	0.58 ± 0.21
Big Pine Key	24°41'44.8"N	Total N	55.00 ± 3.73	51.13 ± 18.39	24°40'32.1"N	Total N	26.67 ± 2.27	30.90 ± 3.24
	81°21'02.0"W	Total P	0.68 ± 0.09	1.85 ± 1.27	81°20'34.5"W	Total P	0.53 ± 0.07	0.55 ± 0.16
Long Key	24°50'25.3"N	Total N	29.08 ± 2.10	42.14 ± 12.12	24°50'30.5"N	Total N	16.92 ± 0.60	19.97 ± 1.99
	80°47'42.8"W	Total P	0.37 ± 0.04	1.00 ± 0.50	80°47'48.1"W	Total P	0.30 ± 0.04	0.33 ± 0.04
Average		Total N	35.12 ± 14.69	38.78 ± 17.33	Average	Total N	19.33 ± 5.58	30.28 ± 8.12
		Total P	0.48 ± 0.19	1.03 ± 0.97		Total P	0.41 ± 0.12	0.49 ± 0.19

			Mesotrophic				Oligotrophic	
	Lat./Long.	Nutrient	Summer	Winter	Lat./Long.	Nutrient	Summer	Winter
Key West	24°33'14.0"N	Total N	14.34 ± 2.55	24.80 ± 4.79	24°27'50.5"N	Total N	10.65 ± 0.08	10.88 ± 1.72
	81°44'54.7"W	Total P	0.17 ± 0.05	0.64 ± 0.14	81°50'30.1"W	Total P	0.12 ± 0.00	0.20 ± 0.06
Big Pine Key	24°38'59.8"N	Total N	25.33 ± 15.21	20.97 ± 6.90	24°32'56.4"N	Total N	10.82 ± 1.30	12.45 ± 1.01
	81°18'49.4"W	Total P	0.16 ± 0.01	0.70 ± 0.16	81°24'24.5"W	Total P	0.12 ± 0.01	0.30 ± 0.06
Long Key	24°50'45.2"N	Total N	24.03 ± 0.82	19.02 ± 1.35	24°51'04.1"N	Total N	14.16 ± 4.53	15.12 ± 4.82
	80°47'44.7"W	Total P	0.15 ± 0.01	0.22 ± 0.10	80°37'32.9"W	Total P	0.26 ± 0.23	0.16 ± 0.01
Average		Total N	20.80 ± 10.01	21.60 ± 5.47	Average	Total N	11.66 ± 3.05	12.78 ± 3.51
		Total P	0.16 ± 0.03	0.53 ± 0.25		Total P	0.16 ± 0.19	0.21 ± 0.08

Hypodermic needles were used to mark *T. testudinum* blades for productivity measurements during the field studies. Blade bundles were marked at the blade-sheath junction of the oldest intact blade. Needle marks were displaced upwards in newer, actively growing blades. Newly identified material was separated from total blade material, and all blade material was weighed to the nearest 0.01 mg on a Mettler analytical balance after drying for at least 24 h at 65°C, as described by Tomasko and Dawes (1989, 1990) and Tomasko and Lapointe (1991). *T. testudinum* biomass did not include below-ground tissue, as rhizomes were found as deep as 50 cm in the sediment (Zieman, 1972; Zieman et al., 1989). In addition, bank reef lagoonal habitats had coral rubble mixed with sediments so that sampling was not possible without pneumatic coring devices. For *H. wrightii*, productivity and biomass were determined using the clip and reharvest method using a 10 cm diameter core as described in Dunton (1990). Shoot productivity and biomass were multiplied by shoot density normalized to a square meter to determine areal productivity and biomass. Leaf area per shoot was determined by multiplying blade lengths by the width of the second youngest blade on each shoot (Tomasko and Lapointe, 1991).

Epiphytes attached to seagrass blades included unidentified taxa of microscopic filamentous algae, calcareous species, diatoms, and other taxa that were collectively removed by scraping the blades with a razor blade. Calcareous epiphytes were brushed from the blades after drying at 65°C for 48 h; ash content of the epiphytes was determined by combustion at 400°C for 3 h. Attached epiphytes were expressed as a percent of blade weight to facilitate comparisons with other studies. Biomass of mat-forming macroalgae was estimated by randomly sampling (N = 10) loose algal biomass within each stratum with a 0.10 m² quadrat. The macroalgae were placed in plastic bags and returned to the lab where they were sorted, cleaned of epizoa and sediments, identified, dried at 65°C for 48 h, and weighed.

ALKALINE PHOSPHATASE ACTIVITY. Intact apical and distal seagrass blade tissue with attached epiphytes as well as predominant macroalgae were collected from the strata and immediately assayed for alkaline phosphatase activity (APA) as an index of P limitation of productivity (Lapointe, 1989). Three replicate measurements for each tissue type and three reagent blanks (reagent medium only to establish background APA) were used within each stratum. APA was measured by the spectrophotometric method of Kuenzler and Perras (1965) as modified for macroalgae by Lapointe (1989). Approximately 1.0 g (wet wt.) of fresh tissue was incubated in an assay medium that contained 3 ml of p-nitrophenyl phosphate (NPP stock; 1.0 g NPP substrate, 25.0 g $MgSO_4$ dissolved in 500 ml deionized water), 6 ml of 1 M Tris buffer, and brought to volume with seawater from each stratum filtered through a 1 μm filter (final volume, 100 ml of assay medium). Following 1 h of incubation in clean Nalgene bottles under midday natural irradiance (~1,500 μmol photons·m^{-2}·s^{-1}) at stable ambient temperatures, absorbances of the assay media were measured in optically-matched 1 cm-pathlength cuvettes at 410 nm in a portable Hach DR/2000 spectrophotometer. Incubated plants were placed in individual whirl-pak plastic bags and returned to the laboratory where they were dried in a lab oven at 65°C and weighed. APA was determined as μM PO_4^{3-} released·g dry wt^{-1}·h^{-1} from a standard curve based on absorbance @ 410 nm versus p-nitrophenyl concentration.

Experimental Microcosm Studies: Design and Operation.—A 2 × 2 factorial experiment, utilizing two levels of N and P enrichment (low level as ambient seawater, high level achieved by known additions of reagent grade nutrients), was used to determine the main effects and interactions of N and P enrichment on blade epiphyte levels and below ground productivity (rhizome growth). Three replicate microcosms (120 liter) were used per treatment, resulting in a total of 12 microcosms. The microcosms were maintained at Crane Point Hammock in Marathon (Fig. 1) where seawater was continuously pumped from adjacent surface waters via a plastic impeller pool pump and metered with PVC ball valves. High seawater turnover rates of 50 volume exchanges/day minimized "enclosure effects" (Lapointe and Ryther, 1979) and ensured water quality in the microcosms was representative of nearshore water quality.

T. testudinum and *H. wrightii* were collected from shallow waters near Marathon, FL, transplanted into the microcosms, and acclimated for 2 weeks prior to initiation of the experiment. Only plants with intact rhizome apices and at least three short shoots were used to enhance survivorship (Tomasko et al., 1991). Grazing organisms were added to all microcosms after sieving a heavily epiphytized grass bed with a fine mesh net for the collection of micrograzers (Tomasko and Lapointe, 1991). Plants were tagged just in front of the second youngest short shoot, and the distance between the tag and the end of the rhizome apex recorded.

During the microcosm experiments water flow was terminated in all 12 aquaria at 1800 daily, and the aquaria enriched by known addition of N (as NH_4^+, from reagent grade NH_4Cl solutions) and/or soluble reactive phosphate (SRP, from reagent grade NaH_2PO_4 solutions) to produce nutrient concentrations of 10 μM NH_4^+ and 1 μM SRP. These concentrations are typical of waters of the Keys impacted by wastewater discharges (Lapointe et al., 1990; Lapointe and Clark, 1992). Low seagrass biomass levels and gentle aeration in the microcosms insured that critically low dissolved oxygen concentrations did not occur during the no-flow nightime hours. Following overnight nutrient enrich-

ment, seawater flow to the microcosms was restored at 0800. We have previously used this "pulsed" nutrient-enrichment technique as a bioassay for the type and degree of nutrient-limited productivity of macroalgae in the Florida Keys and Caribbean (Lapointe, 1987; Lapointe et al., 1987). New rhizome growth of the seagrasses was determined after 4 weeks by subtracting the amount of rhizome originally present (Tomasko and Lapointe, 1991). Levels of epiphytes were determined among the experimental treatments as described above.

Statistical Analyses.—Water quality data (nutrients and dissolved oxygen) collected from the three transects during the field studies were analyzed by two-way (between seasons and strata) ANOVA. Pairwise *t*-tests were used for comparisons of water quality data between specific locations within each season. The seagrass data from two of the transects in the field studies were confounded by the fact that *H. wrightii* dominated the HYPER strata at Long Key and Key West whereas all other strata including the entire Big Pine Key transect was dominated by *T. testudinum*; accordingly, we chose the Big Pine Key transect database to assess statistical significance for the main effects and interaction of strata (HYPER, EUTR, MESO, OLIGO) and season (winter, summer) on the seagrass variables with two-way ANOVA. One-way ANOVA was also used to assess the main effect of season on epiphyte levels from the field studies. Two-way and one-way ANOVA as well as pairwise *t*-tests were used to analyze the APA data for macroalgae and seagrass blade epiphytes. The effects of N and P enrichment on seagrass growth and epiphyte levels during the microcosm factorial studies were assessed with two-way ANOVA. Duncan's multiple range test was also used to determine differences among treatment means. Statistical significance reported below indicates the probability of the null hypothesis is <0.05.

RESULTS

Field Study: Water Quality.—Two-way ANOVA of seawater TN data from all strata, transects, and for both seasons revealed significant effects of season ($F = 4.79$, $P = 0.03$) but highly significant effects of strata ($F = 36.09$, $P < 0.0001$); for TP, the effects of both season ($F = 15.98$, $P = 0.001$) and strata ($F = 12.30$, $P < 0.0001$) were also significant. TN and TP concentrations decreased with increasing distance from shore such that HYPER < EUTR < MESO < OLIGO strata (Table 1). During winter, TN averaged 38.78 ± 17.33 μM, 30.28 ± 8.12 μM, 21.60 ± 5.47 μM, and 12.78 ± 3.51 μM at the HYPER, EUTR, MESO, and OLIGO strata, respectively; TP concentrations decreased from 1.03 ± 0.97 μM, 0.49 ± 0.19 μM, 0.53 ± 0.25 μM, and 0.21 ± 0.08 μM along this same eutrophication gradient (Table 1).

During the summer, dawn DO concentrations averaged 1.55 ± 0.53 mg·liter^{-1} at the HYPER strata, 2.2 ± 1.16 mg·liter^{-1} at the EUTR strata, 3.28 ± 2.04 mg·liter^{-1} at the MESO strata, and 5.44 ± 0.47 mg·liter^{-1} at the OLIGO strata. Salinity ranged from 32.5 to 37.0‰ among the various strata during summer, averaging 34.7 ± 1.28‰. The lowest salinities (~32.5–33.5‰) occurred at the HYPER strata at Key West and Long Key both of which were influenced by land-based inputs of fresh water derived from stormwater runoff and domestic wastewater. During winter, dawn DO concentrations averaged 2.38 ± 0.41 mg·liter^{-1} at the HYPER strata, 2.90 ± 0.91 mg·liter^{-1} at the EUTR strata, 4.30 ± 0.64 mg·liter^{-1} at the MESO strata, and 6.07 ± 1.44 mg·liter^{-1} at the OLIGO strata. Salinity during winter ranged from 36.1 to 37.6‰ and averaged 37.1 ± 0.27‰.

SEAGRASS MEASUREMENTS. *H. wrightii* was the only seagrass species at the HYPER strata on the Long Key and Key West transects wheras *T. testudinum* was the dominant seagrass species at all other strata along the three transects. *H. wrightii* at the HYPER strata had the highest shoot densities during this study (Fig. 2). Two-way ANOVA of the Big Pine Key transect data showed that shoot densities of *T. testudinum* were significantly affected by strata ($F = 266$, $P = 0.0001$), season ($F = 6.56$, $P = 0.0125$), and the season × strata interaction ($F = 19.80$, $P = 0.0001$). For the strata where *T. testudinum* was dominant, densities increased from HYPER < EUTR < MESO < OLIGO strata; shoot densities at
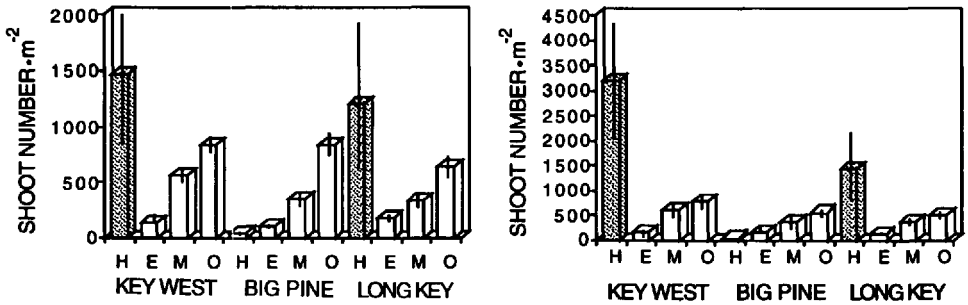
Figure 2.   Shoot density of dominant seagrass species during the summer (1991, left) and winter (1992, right) field studies in hypereutrophic, eutrophic, mesotrophic, and oligotrophic strata along three offshore transects. Shaded bars represent *Halodule wrightii* and unshaded bars represent *Thalassia testudinum*. Values represent means ± 1 SD (N = 10).

OLIGO strata during summer were typically 5 to 10 times higher than those at EUTR strata.

Shoot biomass of *T. testudinum* along the three transects was generally greater than that of *H. wrightii* at the HYPER strata (Table 2). Two-way ANOVA of the Big Pine Key data for *T. testudinum* showed that the main effects of strata (F = 29.79, P < 0.0001), season (F = 4.59, P = 0.035), and the season × strata interaction (F = 10.40, P = 0.001) all significantly affected shoot biomass. In the Big Pine Key transect data, shoot biomass increased from HYPER to OLIGO strata (Table 2). However, shoot biomass for *T. testudinum* was not significantly different between the EUTR and OLIGO strata at Key West and Long Key during either summer or winter.

T. testudinum had higher shoot productivity along the three transects compared to *H. wrightii* at the HYPER strata at Key West and Long Key (Table 2). Two-way ANOVA of the Big Pine Key data indicated that strata (F = 11.64, P = 0.0001) and season (F = 10.63, P = 0.0017) significantly affected shoot productivity of *T. testudinum*. Shoot productivity increased significantly from the HYPER to OLIGO strata at Big Pine Key, but was statistically similar between the EUTR and OLIGO strata at Key West and Long Key. For all three transects, shoot productivity was at least 5-fold higher at the OLIGO compared to the HYPER strata (Table 2).

Two-way ANOVA showed that the effects of strata (F = 10.66, P = 0.0001) and season (F = 12.47, P = 0.0007) significantly affected blade epiphytes on *T. testudinum* along the Big Pine Key transect. Blade epiphytes were highest at the HYPER (Key West and Long Key) and EUTR strata (Big Pine Key; Table 2). However, epiphyte levels were frequently higher at OLIGO compared to MESO strata due to dense growths of calcareous epiphytic algae (*Melobesia membranaceae* and *Fosliella farinosa*) that have high mass:volume ratios at the OLIGO strata; diatoms and fleshy filamentous taxa were more common at the more enriched HYPER and EUTR strata. Blade epiphyte levels were significantly higher in the winter compared to summer (Table 2).

Results of two-way ANOVA indicated that strata (F = 4.16, P = 0.023) but not season significantly affected biomass of mat-forming macroalgae. During both seasons, epiphytic macroalgae were most abundant at the HYPER and EUTR strata, where biomass levels ranged from 13.7 to 291 g dry wt·m^{-2} and averaged 101.8 g dry wt·m^{-2} (Table 2). Intermediate values, ranging from 6.1 to 58.4 g dry wt·m^{-2}, occurred at the MESO strata and the lowest values (<10.1 g dry

Table 2. Seagrass variables along three transects containing four trophic strata in the Florida Keys. Values for shoot biomass (mg dry wt.·shoot⁻¹), shoot productivity (mg dry wt.·shoot⁻¹·day⁻¹), seagrass epiphytes (ash-free % of blade wt.), macroalgal biomass (g dry wt.·m⁻²) and turnover time (days) represent means ± 1 SD (N = 10). All data based on *Thalassia testudinum* except where an asterisk (*) indicates *Halodule wrightii*.

| Transect location	Shoot biomass		Shoot productivity		Seagrass epiphytes		Macroalgal biomass		Turnover time	
	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer	Winter
Key West										
Hypereutrophic	17.76 ± 2.69*	17.12 ± 10.51*	0.82 ± 0.24*	0.21 ± 0.08*	20.46 ± 8.17*	28.59 ± 12.84*	13.7 ± 7.4	53.2 ± 18.2	27.6*	80.56*
Eutrophic	270.53 ± 55.01	131.01 ± 62.60	5.42 ± 1.24	2.31 ± 1.29	15.10 ± 7.77	48.35 ± 27.07	89.3 ± 35.2	71.0 ± 20.2	50.1	56.27
Mesotrophic	193.46 ± 36.63	169.04 ± 41.89	3.63 ± 0.82	3.37 ± 1.01	10.75 ± 3.06	13.81 ± 3.93	6.1 ± 3.9	58.4 ± 18.4	53.2	50.44
Oligotrophic	217.59 ± 52.38	209.20 ± 80.45	5.07 ± 1.40	3.27 ± 0.99	18.33 ± 3.77	11.52 ± 3.47	1.2 ± 0.8	1.1 ± 1.5	42.9	63.94
Big Pine										
Hypereutrophic	96.03 ± 39.44	111.92 ± 26.34	1.52 ± 0.50	1.37 ± 0.27	10.90 ± 6.25	97.28 ± 71.18	55.9 ± 30.3	60.9 ± 35.4	66.5	79.3
Eutrophic	237.62 ± 37.64	158.63 ± 41.89	5.16 ± 1.34	1.89 ± 0.64	31.39 ± 30.09	26.30 ± 11.89	70.2 ± 13.9	122.4 ± 41.7	45.9	84.34
Mesotrophic	408.95 ± 93.22	244.09 ± 113.53	6.76 ± 1.23	4.42 ± 1.91	3.06 ± 0.91	11.05 ± 5.28	11.9 ± 6.7	8.0 ± 4.1	60.4	55.19
Oligotrophic	191.70 ± 66.25	275.02 ± 97.29	10.89 ± 9.63	5.57 ± 1.87	6.93 ± 1.46	12.06 ± 3.94	1.1 ± 0.9	1.3 ± 1.7	17.6	49.45
Long Key										
Hypereutrophic	14.98 ± 3.71*	8.66 ± 2.02*	0.55 ± 0.29*	0.46 ± 0.37*	29.13 ± 12.16*	57.86 ± 11.44*	291.2 ± 125.0	115.8 ± 38.5	27.4*	18.83*
Eutrophic	292.24 ± 64.92	163.85 ± 43.47	5.51 ± 3.21	1.87 ± 1.33	2.87 ± 0.93	12.83 ± 4.57	75.4 ± 29.3	202.9 ± 3.3	53.3	87.29
Mesotrophic	290.16 ± 75.92	164.85 ± 39.49	6.67 ± 3.92	1.99 ± 0.63	2.33 ± 1.15	8.87 ± 5.90	55.5 ± 23.0	33.9 ± 13.5	43.5	83.02
Oligotrophic	215.81 ± 43.24	232.77 ± 70.14	5.70 ± 1.88	4.53 ± 1.69	8.70 ± 1.98	17.10 ± 5.48	10.1 ± 5.3	7.0 ± 4.2	37.8	51.46
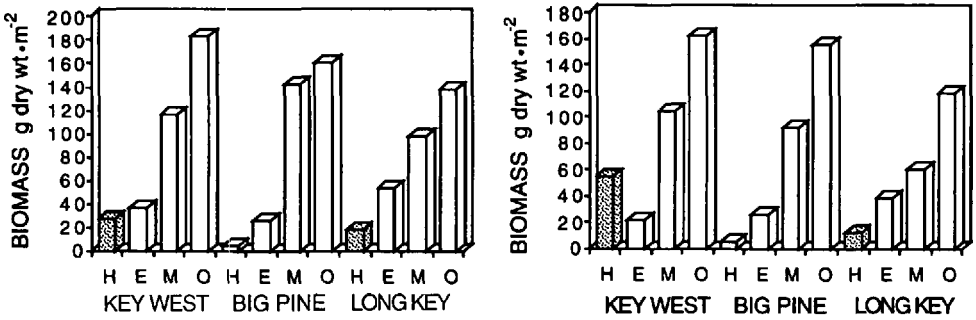
Figure 3.   Areal biomass of dominant seagrass species during the summer (1991, left) and winter (1992, right) field studies in hypereutrophic, eutrophic, mesotrophic, and oligotrophic strata along three offshore transects. Shaded bars represent *Halodule wrightii* and unshaded bars represent *Thalassia testudinum*.

wt·m^{-2}) consistently occurred at the OLIGO strata (Table 2). Blade epiphyte levels were significantly higher in the winter compared to summer (Table 2).

Two-way ANOVA showed that the effects of strata (F = 14.71, P = 0.0001), season (F = 17.34, P = 0.0001), and the strata × season interaction (F = 7.20, P = 0.0003) all significantly affected blade turnover times. During summer, turnover times were lowest at the Key West and Long Key HYPER strata where *H. wrightii* was the dominant seagrass species (Table 2). In contrast, the highest blade turnover times for *T. testudinum* along the Big Pine Key transect occurred at the HYPER stratum. During winter, blade turnover times were higher for *H. wrightii* at the HYPER stratum at Key West, but lower for *H. wrightii* at Long Key (Table 2). During both seasons at Big Pine Key and Long Key, blade turnover times of *T. testudinum* generally decreased from the EUTR to OLIGO strata.

During both summer and winter, areal biomass increased from HYPER to OLIGO strata; biomass values at OLIGO strata were 4–6 fold higher than at the HYPER strata for all three transects (Fig. 3). Areal productivity also increased from HYPER to OLIGO strata during both seasons, with the highest values during summer (Fig. 4). Areal productivity at the OLIGO strata off Big Pine Key ranged up to ~ 9 g dry wt·m^{-2}·d^{-1} in summer compared to ~3 g dry wt·m^{-2}·d^{-1} in winter. Areal productivity was ~4 fold higher at the OLIGO compared to the HYPER strata (Fig. 4).
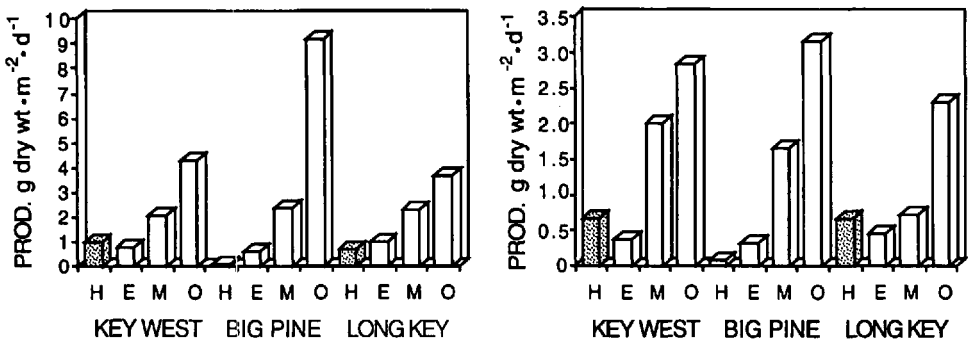

Figure 4.   Areal productivity during the summer (1991, left) and winter (1992, right) field studies in hypereutrophic, eutrophic, mesotrophic, and oligotrophic strata along three offshore transects. Shaded bars represent *Halodule wrightii* and unshaded bars represent *Thalassia testudinum*.
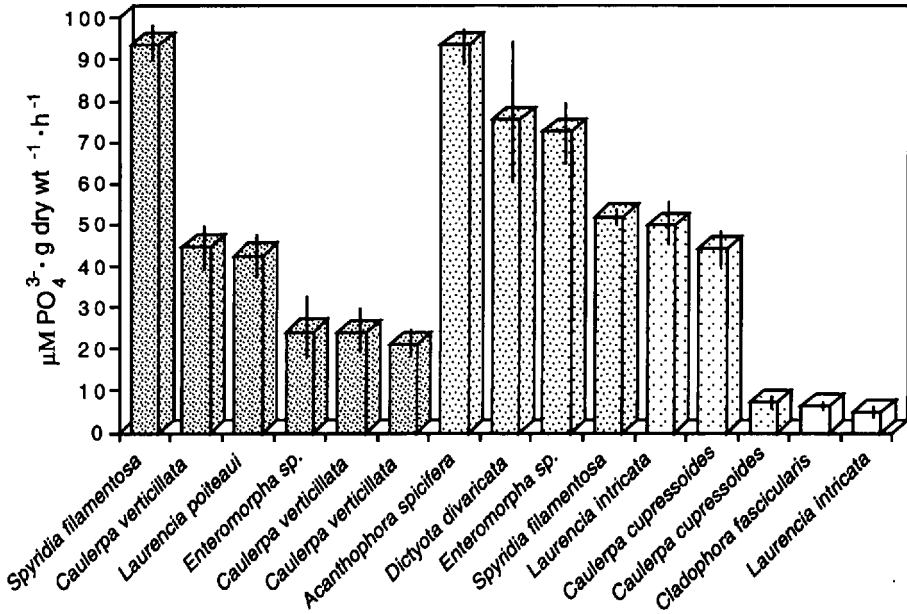
Figure 5. Alkaline phosphatase activity of predominant macroalgae from hypereutrophic (dark shading), eutrophic (intermediate shading), mesotrophic (light shading), and oligotrophic (unshaded) strata during the summer (1991) field studies. Values represent means ± 1 SD (N = 3).

ALKALINE PHOSPHATASE ACTIVITY. Two-way ANOVA of the APA data for macroalgal epiphytes showed significant effects of strata (F = 22.12, $P < 0.0001$) and the strata × season interaction (F = 6.239, $P = 0.0006$) but insignificant effects of season. The highest macroalgal APA occurred in the HYPER and EUTR strata and the lowest in the OLIGO strata (Figs. 5, 6). The rhodophytes *Spyridia filamentosa* and *Acanthophora spicifera* in HYPER and EUTR strata had the highest APA values in summer (>80 μM PO_4^{3-} released·g dry $wt^{-1} \cdot h^{-1}$; Fig. 5) while the chlorophytes *Enteromorpha* sp. in HYPER strata had the highest APA during winter (>100 μM PO_4^{3-} released·g dry $wt^{-1} \cdot h^{-1}$; Fig. 6). The phaeophytes *Dictyota divaricata* and *D. dichotoma*, the rhodophytes *Laurencia intricata* and *L. poiteaui*, and the siphonaceous chlorophytes *Caulerpa cupressoides* and *C. verticillata* all had relatively high APA within the HYPER and EUTR strata during both summer and winter (Figs. 5, 6). *L. poiteaui* and *L. intricata* from MESO strata had intermediate APA (20–50 μM PO_4^{3-} released·g dry $wt^{-1} \cdot h^{-1}$) whereas these same species, as well as the phaeophytes *Padina vickersiae* and *D. dichotoma* and the chlorophytes *C. cupressoides* and *Cladophora fascicularis* from OLIGO strata had low APA (<15 μM PO_4^{3-} released·g dry $wt^{-1} \cdot h^{-1}$; Figs. 5, 6). Within the species *L. poiteaui* in the winter studies, APA decreased significantly from EUTR > MESO > OLIGO strata (Fig. 6).

Two-way ANOVA of apical seagrass blade APA showed significant effects of strata (F = 7.37, $P = 0.0003$) and season (F = 3.928, $P = 0.049$). For the more highly epiphytized distal blade tissue, the effects of strata (F = 12.41, $P < 0.0001$), season (F = 8.95, $P = 0.0039$), and the strata × season interaction (F = 8.29, $P < 0.0001$) were all significant. One-way ANOVA showed that the most significant effects of season on distal tissue APA was in the EUTR and MESO strata; APA of the distal seagrass tissue averaged 53.7 and 35.9 μM PO_4^{3-} re-
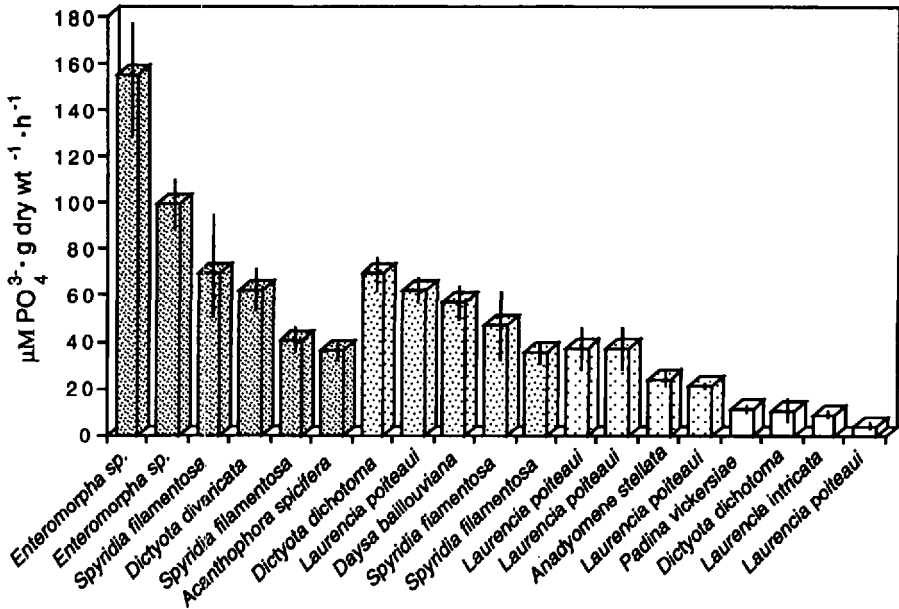
Figure 6. Alkaline phosphatase activity of predominant macroalgae from hypereutrophic (dark shading), eutrophic (intermediate shading), mesotrophic (light shading), and oligotrophic (unshaded) strata during the winter (1992) field studies. Values represent means $\pm$ 1 SD (N = 3).

leased·g dry $wt^{-1}\cdot h^{-1}$ in the EUTR and MESO strata during summer compared to lower values of 15.6 and 13.7 μM PO_4^{3-} released·g dry $wt^{-1}\cdot h^{-1}$ during winter, respectively (Figs. 7, 8). The highest APA of both apical and distal blade tissue consistently occurred in the HYPER and EUTR strata with the lowest in the OLIGO strata (Figs. 7, 8).

APA of *T. testudinum* and *H. wrightii* blade tissue was higher on distal versus apical sections (in 19 out of 24 pairwise t-tests, $P < 0.05$; Figs. 7, 8). APA of apical seagrass tissue ranged up to 35 μM PO_4^{3-} released·g dry $wt^{-1}\cdot h^{-1}$, compared to values ranging up to 100 μM PO_4^{3-} released·g dry $wt^{-1}\cdot h^{-1}$ for more heavily epiphytized distal seagrass tissue in the HYPER and EUTR strata. The highest APA values for distal tissue of *H. wrightii* and *T. testudinum* were at the Long Key HYPER stratum in winter (Fig. 8; ~80 μM PO_4^{3-} released·g dry $wt^{-1}\cdot h^{-1}$) and the Key West EUTR stratum in summer (Fig. 7; ~70 μM PO_4^{3-} released·g dry $wt^{-1}\cdot h^{-1}$), respectively.

Microcosm Studies.—Two-way ANOVA indicated significant enrichment effects of N (F = 10.28, P = 0.002), P (F = 53.84, P = 0.0001), and N × P interaction (F = 17.70, P = 0.0001) on increased epiphyte biomass of *H. wrightii* (Fig. 9). Both the controls and N enrichment treatments had significantly lower epiphyte levels than the P and N + P treatments, indicating a primary importance of P enrichment in stimulating epiphyte growth on this seagrass (Fig. 9). For *T. testudinum,* epiphyte biomass was significantly increased by enrichment with both N (F = 8.91, P = 0.005) and P (F = 3.76, P = 0.049; Fig. 10); no significant differences in epiphyte levels were found between the N, P, and N + P treatments (Fig. 10).

P enrichment significantly (F = 5.91, P = 0.020) decreased rhizome growth rates of *H. wrightii* whereas the effects of N and N + P enrichment were insig-
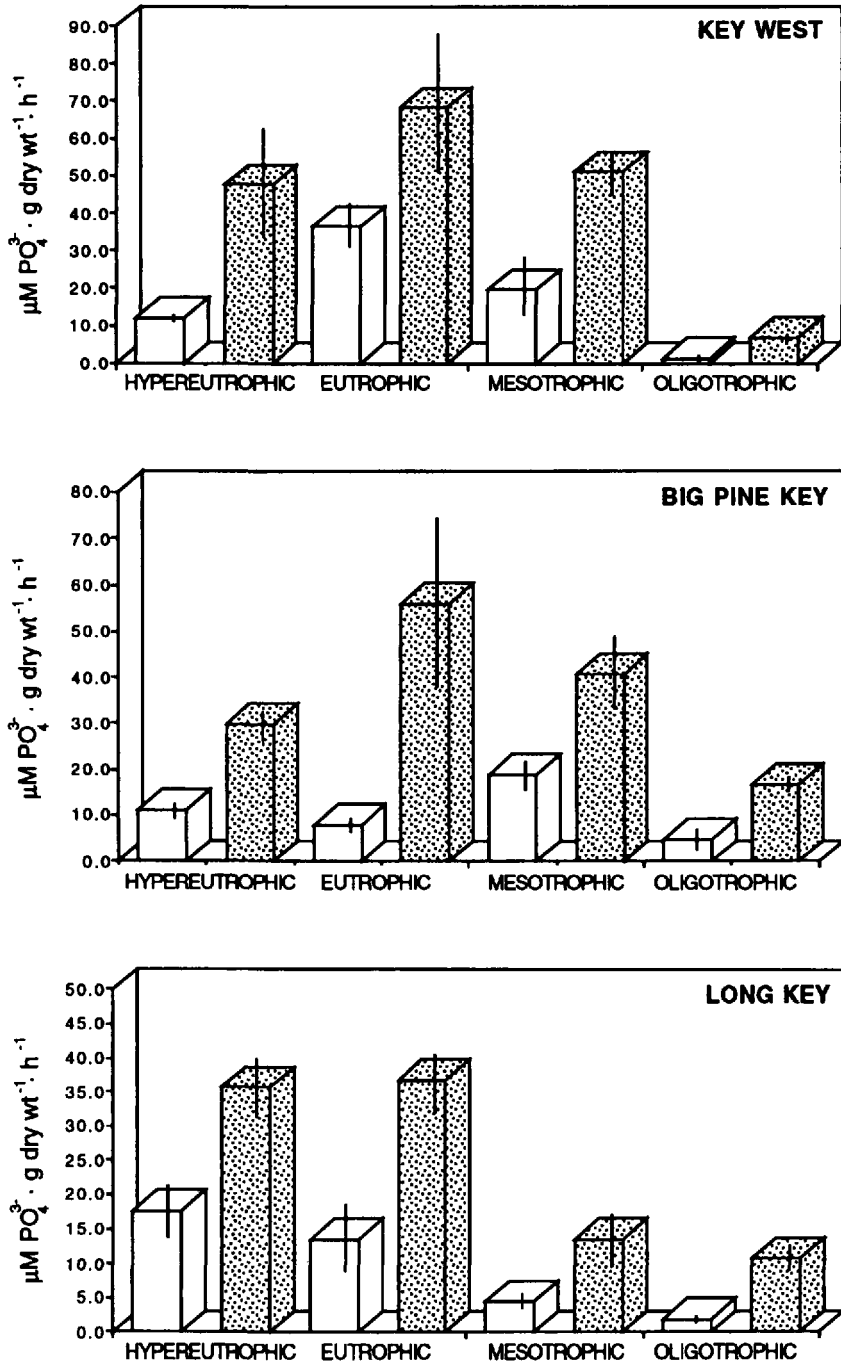
Figure 7.   Alkaline phosphatase activity localized for apical (unshaded) and distal (shaded) seagrass blade tissue from hypereutrophic, eutrophic, mesotrophic, and oligotrophic strata during the summer (1991) field studies. Values represent means ± 1 SD (N = 3).
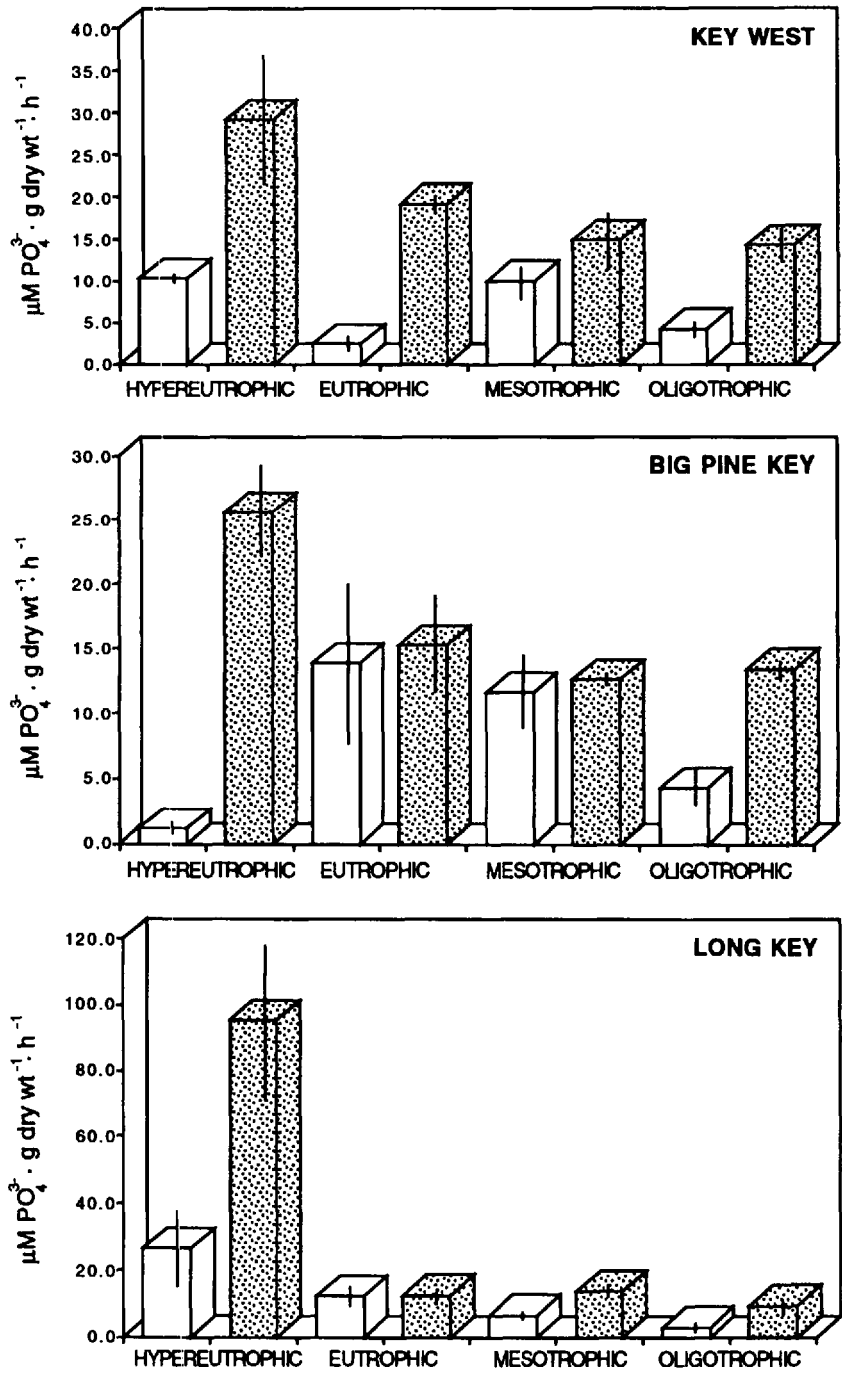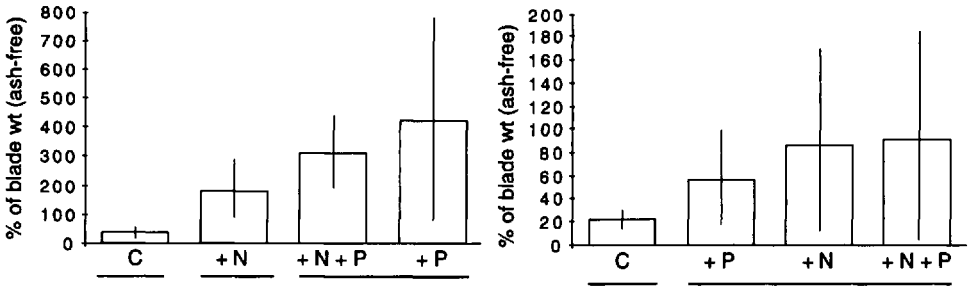
Figure 8. Alkaline phosphatase activities localized for apical (unshaded) and distal (shaded) seagrass blade tissue from hypereutrophic, eutrophic, mesotrophic, and oligotrophic strata during the winter 1992 studies. Values represent means ± 1 SD (N = 3).

Figure 9 (left).   Epiphyte levels on *Halodule wrightii* in response to nitrogen (N), phosphorus (P), and N + P enrichment of the water column; controls (C) received no enrichment. Shared underlines denote a lack of significant difference at P < 0.05. Values represent means ± 1 SD (N = 9).

Figure 10 (right).   Epiphyte levels on *Thalassia testudinum* in response to nitrogen (N), phosphorus (P), and N + P enrichment of the water column; controls (C) received no enrichment. Shared underlines denote a lack of significant difference at P < 0.05. Values represent means ± 1 SD (N = 9).

nificant (Fig. 11). P enrichment and N + P enrichment decreased rhizome growth rates of *H. wrightii* below that of the controls (Fig. 11). In contrast, significant effects of N (F = 4.74, P = 0.037), P (F = 4.70, P = 0.04), and N + P enrichment (F = 4.38, P = 0.044) reduced rhizome growth rates of *T. testudinum* (Fig. 12). Rhizome growth rates were not significantly different among the three nutrient enrichment treatments but those three treatments had rhizome growth rates lower than that of the controls (Fig. 12).

DISCUSSION

Trophic State Classification of Seagrass Communities.—Our study initiates the application of trophic state classification as a method to gauge the structural and functional characteristics of tropical seagrass communities in relation to water column nutrient concentrations. The word eutrophy, derived from the German adjective eutrophe and in general referring to "nutrient-rich" (Wetzel, 1975), is an often ambiguous term; we use the word eutrophication to refer to a process in which increased nutrient concentrations resulting from human activities results in
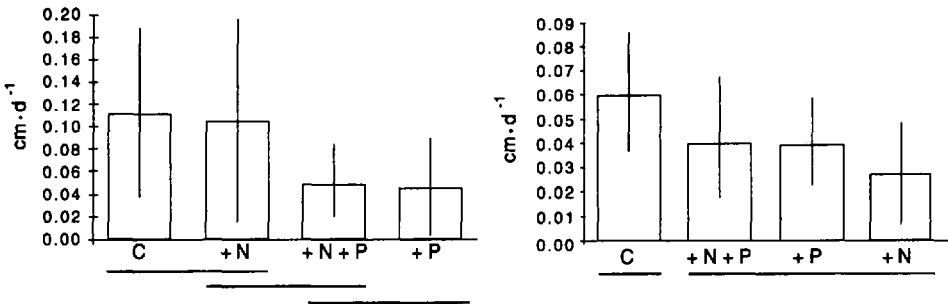

Figure 11 (left).   Rhizome growth rates of *Halodule wrightii* in response to nitrogen (N), phosphorus (P), and N + P enrichment of the water column; controls (C) received no enrichment. Shared underlines denote a lack of significant difference at P < 0.05. Values represent means ± 1 SD (N = 9).

Figure 12 (right).   Rhizome growth rates of *Thalassia testudinum* in response to nitrogen (N), phosphorus (P), and N + P enrichment of the water column; controls (C) received no enrichment. Shared underlines denote a lack of significant difference at P < 0.05. Values represent means ± 1 SD (N = 9).

"nuisance" algal growth. Paerl (1988) defines "nuisance" algal categories to include perceptible water quality degradation, including trophic changes. Our study identified several species of macroalgae that fit the nuisance category for the various trophic states in seagrass communities of the Florida Keys region that can be used as indicators of the relative trophic status. An objective method of trophic state classification has long been sought by limnologists to rank lakes with different structural and trophic characteristics (Naumann, 1919; Thienemann, 1925). A similar classification method is a useful conceptual framework for scientists and managers alike to not only better characterize the water quality requirements of seagrass habitats but also to help justify improved management policies aimed at reducing the impacts of cultural eutrophication on seagrass productivity and the fisheries dependent on these habitats.

Most trophic state classification methods for fresh water lakes rely on water column variables, such as nutrient concentrations, phytoplankton biomass, transparency, and primary productivity (Goldman, 1988) although some studies have considered aquatic macrophyte biomass (Canfield et al., 1983). Water column nutrient concentrations, chlorophyll *a,* and enhanced growth of macroalgae have also been used to gauge the onset of eutrophication in coral reef regions (Bell, 1992). Because dissolved inorganic nutrient concentrations are highly variable and do not include particulate or dissolved organic nutrient pools, the use of total N and P pools appears to be the best single nutrient index of eutrophication as this measurement includes all nutrient pools and is also a proxy for water transparency (Lapointe and Clark, 1992). Results of the present study showed that total N and P concentrations decreased linearly from HYPER to OLIGO strata and that average concentrations of ~25 µM N and 0.45 µM P represent levels above which seagrass meadows dominated by *T. testudinum* change to hypereutrophic communities dominated by *H. wrightii* and opportunistic macroalgae. Powell et al. (1991) also noted that *H. wrightii* replaced *T. testudinum* in habitats receiving sustained guano enrichment in Florida Bay. We define the term hypereutrophication as a state when nutrient concentrations rise above critical threshholds and initiate change in the relative dominance of benthic community structure (sensu Littler and Littler, 1984b for tropical reef communities). Although we assume that light limitation was the primary mechanism causing die-off of *T. testudinum* in our HYPER strata, direct effects of nutrient enrichment (such as toxicity, see Burkholder et al., 1992) cannot be ruled out. Other biotic and abiotic factors affected by nutrient enrichment, including changes in grazing (e.g., loss of grazers by low DO), water temperature (e.g., increased temperatures with increased chlorophyll), etc., could also contribute to stress and decline of *T. testudinum.*

Along the trophic gradient in our study, *T. testudinum* typifies an "oligotrophic species" and *H. wrightii* a "eutrophic species." In their natural state prior to human development, protected shallow waters of the Florida Keys, including dredged canal systems, had extensive oligotrophic seagrass communities dominated by *T. testudinum,* which is typical of similar areas of the Caribbean (Phillips and Menez, 1987). Inshore waters of the Florida Keys have been noticeably impacted by wastewater nutrient inputs from land-based residential and tourist development over the past two decades and this enrichment has spatially spread to more offshore strata (Lapointe and Clark, 1992). As a result, inshore HYPER and EUTR seagrass meadows now differ from more offshore MESO and OLIGO meadows in that inshore communities have greater diversity of primary producers, including macroalgae, attached seagrass epiphytes, phytoplankton, and medusae (*Cassiopeia*) with symbiotic zooxanthellae.

Nutrient-enhanced productivity of macroalgae and attached epiphytes leads not

only to decreased productivity of *T. testudinum,* but also to reduced dissolved oxygen levels that can result in significant habitat damage prior to actual die-off. The present study, and others (Lapointe and Clark, 1992), have shown that eutrophication in seagrass meadows in the Florida Keys and Florida Bay results in predawn hypoxia (<2.0 mg·liter^{-1} DO) or anoxia (<0.1 mg·liter^{-1}), especially during warm, rainy periods. Decreased oxygen levels result from both increased light-limitation (due to shading by macroalgae, attached epiphytes and phytoplankton), increased community respiration resulting from high macroalgal biomass (Valiela et al., 1990), and increased sediment oxygen demand associated with mineralization of organic matter (Mee, 1988). McClanahan (1992) reported low predawn oxygen concentrations that were negatively correlated with species richness and diversity of epibenthic gastropods in Florida Bay compared to higher oxygen waters in Hawk Channel offshore Key Largo; he noted that low oxygen stress resulting from eutrophication reduced the abundance of higher trophic levels resulting in dominance of ancestral forms not adapted to predation but tolerant of stress. Low oxygen levels could also exacerbate the algal epiphyte problems by elimination of important groups of algal grazers.

Macroalgae as Mediators of Eutrophication.—Our observation that blooms of mat-forming macroalgae and/or attached epiphytes significantly correlated with the decline of *T. testudinum* and increase in *H. wrightii* was also made by Reyes and Merino (1991) for Bojorquez Lagoon, Cancun, Mexico, an area like the Florida Keys that has been impacted by eutrophication from tourism development and associated sewage inputs. In Bojorquez Lagoon, blooms of the chlorophytes *Chaetomorpha* and *Acetabularia* formed mats that covered *T. testudinum* prior to die-off (Reyes and Merino, 1991). On a smaller scale, guano enrichment on the lee side of Man-of-War Cay on the Belize barrier reef resulted in blooms of the mat-forming macroalgae *Ulva lactuca, Chaetomorpha linum, Enteromorpha* spp., and *Acanthophora spicifera* that overgrew and outcompeted *T. testudinum* (Lapointe et al., 1994).

Thus, persistent blooms of macroalgae, epiphytes, and phytoplankton mediate the decline of *T. testudinum* meadows impacted by sustained nutrient inputs. In the present study, spatial and temporal patterns of APA illustrated the variability of nutrient inputs and biological cycling that sustain such algal blooms. APA is an exoenzyme produced by bacteria and algae that hydrolyzes dissolved organic phosphorus (DOP) compounds and releases metabolically active SRP (Kuenzler and Perras, 1965); because APA increases with P-limitation in algae (Kuenzler, 1965) it can be used as a proxy for the relative growth limitation by P. Our present study showed that the APA of seagrass blade tissue is localized primarily on the distal, older and more epiphytized tissue; we also found that these distal APA values were higher in summer than winter for the EUTR and MESO strata.

These distinct seasonal and spatial patterns in APA of seagrass epiphytes and macroalgae are consistent with previous studies of nutrient-limited algal productivity in the Keys. Cage culture bioassays with the rhodophyte *Gracilaria tikvahiae* in Pine Channel adjacent to Big Pine Key showed higher P limitation of productivity during the summer wet season when seasonally maximum nutrient input occurs (Lapointe, 1987), a finding corroborated by the higher APA of seagrass epiphytes during summer months in the present study. Nutrient inputs from septic tank leachate in the Keys also have high N:P ratios due to selective adsorption of P onto calcium carbonate minerals during migration of wastewater plumes through limestone groundwaters (Lapointe et al., 1990). This phenomenon results in elevated water column DIN concentrations (both NH_4^+ and NO_3^-) and

N:P ratios in nearshore waters and a gradient of decreasing P limitation but increasing N limitation with increasing distance from shore (Lapointe and Clark, 1992). Our present study confirmed this pattern by demonstrating a gradient of decreasing APA in predominant macroalgae with increasing distance from shore.

The first-order effects of nutrient enrichment on the long term productivity and structure of *T. testudinum* meadows were evident from our study. Communities dominated by *T. testudinum* at HYPER strata had low shoot densities (except where *H. wrightii* was present), low shoot production rates, low shoot biomass values, high epiphyte levels, low areal biomass values, and low areal production rates. In general, seagrass shoot densities, areal biomass values, and areal production rates consistently increased with increasing distance from land-based nutrient input along each transect. The only strata where *H. wrightii* was dominant were the HYPER strata at Key West and Long Key. MESO strata were typically more similar to OLIGO strata than HYPER or EUTR strata. Shoot densities, areal biomass values, areal production rates, epiphyte levels, and APA values of epiphytic algae were most similar between the MESO and OLIGO sites. Despite the broad similarity between MESO and OLIGO strata, areal production rates of *T. testudinum* were highest in OLIGO rather than MESO strata; maximum values of ~10 g dry wt·m^{-2}·d^{-1} and ~3 g dry wt·m^{-2}·d^{-1} occurred at the OLIGO stratum offshore Big Pine Key in the summer and winter, respectively. OLIGO strata from all three transects had the highest shoot densities, areal biomass values, and areal production rates but the lowest epiphyte levels and APA values for epiphytic macroalgae in the entire study. Because densities of benthic invertebrates and fishes that inhabit seagrass meadows are positively correlated with seagrass density and productivity (Stoner, 1983; Sogard et al., 1987), eutrophication of nearshore seagrass meadows will also reduce secondary production.

Nutrient Enrichment and Macroalgal Growth.—The high APA of the attached seagrass epiphytes and mat-forming macroalgae allows them to efficiently recycle and utilize the DOP pool in the high N inshore waters of the Florida Keys. For example, the rhodophytes *Spyridia filamentosa, Acanthophora spicifera,* and *Dasya baillouviana,* and the phaeophytes *Dictyota* spp., and the chlorophytes *Cladophora fasicularis, Enteromorpha* spp., and *Caulerpa verticillata* all had high APA and were common in HYPER and EUTR strata. Symbiotic medusae (*Cassiopeia*) were also abundant in these nutrient-enriched areas, possibly in response to the elevated NH$_4^+$ that is utilized by the medusae zooxanthellae (Muscatine and Marian, 1982). In contrast to the nutrient enriched areas, calcareous species, including *Melobesia membranacea* and *Fosliella farinosa,* dominated the attached epiphytic community at the OLIGO strata, where the predominant macroalgae had low APA. These findings corroborate the conclusions of Lapointe (1989) and Tomasko and Lapointe (1991) that water column DOP is an important nutrient source regulating the development of nearshore blade epiphyte communities. Burkholder and Wetzel (1990) suggested that under oligotrophic conditions, freshwater macrophytes themselves serve as a significant P source to their algal epiphytes.

Nutrient enrichment from land-based sewage inputs can have significant effects on seagrass productivity for considerable distances from shore. Concentrations of SRP decrease to lower concentrations (<0.20 µM) within 1–2 km from shore in the Florida Keys, but DOP, the larger pool of P in nearshore waters of the Keys, is present at higher concentrations (~0.50 µM) to a distance of 5–6 km from shore (Lapointe and Clark, 1992). The DOP pool is recognized by oceanographers to be labile and an important source of P in oligotrophic waters (Jackson and

Williams, 1985). The higher epiphyte levels on seagrasses at the Key West OLIGO stratum compared to the Big Pine and Long Key OLIGO strata during summer supports previous reports (Lapointe and Clark, 1992) of high DOP (and other nutrients) on reefs adjacent to Key West. These observations suggest that nutrient enrichment from Key West are impacting seagrasses at Sand Key some 7 km from shore. Key West is the most populated island in the Florida Keys and the offshore reefs are impacted by nutrient enrichment from Key West's 7 MGD sewage outfall, stormwater runoff, and submarine discharge of enriched groundwaters. Studies in Australia have shown that unusually heavy growths of epiphytes on seagrass occur up to 6 km distance from a wastewater outfall (Neverauskas, 1987).

The microcosm studies showed that both N and P increase epiphyte growth and decrease productivity of seagrass communities, especially those dominated by *T. testudinum.* P was the primary nutrient that increased epiphyte levels and decreased rhizome growth rates of *H. wrightii,* in general agreement with previous studies showing strong P limitation of macroalgal productivity in nearshore waters of the Keys (Lapointe, 1987). Reduced rhizome growth rates probably resulted from reduced photosynthetic rates due to light attenuation by epiphyte communities (Sand-Jensen, 1977). In contrast to *H. wrightii,* however, both N and P enrichment increased epiphyte levels and decreased rhizome growth rates of *T. testudinum,* indicating that both N and P contribute to decline of *T. testudinum* meadows. The high N:P ratio of watershed nutrient inputs from groundwaters enriched by septic tank drainfields and injection wells (Lapointe et al., 1990) result in P limitation through stoichiometric dystrophy (i.e., unnaturally high N:P ratios) that increase APA, P cycling, and epiphyte productivity but decrease seagrass productivity. Direct enrichment effects of N are also important as demonstrated by in situ nutrient pulsing studies with the rhodophyte *Gracilaria tikvahiae* (Lapointe, 1987). Genera of mat-forming red macroalgae common in N-enriched waters of the Florida Keys contain N-rich phycobiliprotein complexes and flourish in water with high N availability (Lapointe, 1981).

Results of the present study are the first to suggest that large-scale, long term eutrophication is also likely to have initiated the ongoing decline of *T. testudinum* and increase in *H. wrightii* in Florida Bay (Robblee et al., 1991). Several lines of evidence support this contention. First, the average water column nutrient concentrations in Florida Bay are now well above the levels where we observed decline of *T. testudinum* in the Florida Keys. Fourqurean et al. (1993) reported average total N and P water column concentrations for Florida Bay of 45.6 and 0.58 μM, respectively, values that are 82 and 29% greater than those of our wastewater impacted EUTR strata. Extensive populations of macroalgae were also documented in Florida Bay in the early 1980's, when the rhodophyte *Laurencia* spp. was the second most abundant plant species in Florida Bay, surpassed only by *T. testudinum* prior to its die-off; seagrasses adjacent to the Everglades mainland during that period were also described as "highly epiphytized" (Zieman and Fourqurean, 1985). Thus, the chronology of events in Florida Bay is quite similar to that of the Florida Keys and Cancun Lagoon (Reyes and Merino, 1991) where land-based nutrient enrichment led to increased macroalgal and phytoplankton biomass, seagrass epiphytization, hypoxia, decline of *T. testudinum,* and colonization by *H. wrightii.*

Management Implications.—The results of this study suggest that chronic N and P enrichment of surface waters throughout the Florida Keys will further degrade seagrass communities and food webs dependent on these habitats. Although previous studies in the Keys stressed the primary role of P in limiting growth of

macroalgae, the importance of N was also noted (Lapointe, 1987, 1989). Both N and P have previously been targeted as priority pollutants in clean-up strategies for domestic wastewaters in fresh as well as coastal receiving waters. Monroe County has enacted an ordinance aimed at eliminating cleaning agents with high levels of P, an action that could significantly reduce P inputs at the source. Chemical removal of P from wastewaters via flocculation/sedimentation by ferric chloride (10 to 15 mg·liter^{-1} of Fe^{3+}) is frequently used, but this results in additional problems of sludge handling and disposal (Thomas, 1973). Furthermore, reduction of N loadings needs to be considered in context with an overall nutrient removal strategy for water quality protection. Considering the costs and limitations associated with N removal via conventional denitrification systems (Clark et al., 1977), natural biological systems (e.g., advanced integrated pond systems) that can remove both N and P simultaneously and allow nutrient and water reclamation are likely to be the most cost effective. When properly designed, advanced pond systems virtually eliminate sludge disposal, minimize power use, require less land than conventional ponds, and are much more reliable and economical than mechanical systems of equal capacity (Oswald, 1990).

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