TABLE 7

Amino acids, chlorophyll *a*, and tissue nitrogen in *Gracilaria edulis* after 3 days field incubation in One Tree Island ENCORE experimental patch reefs, phase 2 (high nutrient loading period).[A]

Nutrient addition	Amino acids			Tissue nitrogen	Pigment chlorophyll *a*
	Citrulline		Total amino acids		
	(nmol g_{wet}^{-1})	(% total)	(nmol g_{wet}^{-1})	(%)	(mg g_{wet}^{-1})
Control	250[a]	11	2252	1.30	1.03
+P	368[ab]	16	2280	1.26	0.90
+N	588[bc]	24	2380	1.42	1.08
+N+P	716[c]	29	2496	1.52	1.18
F-value	4.8*		0.1	2.2	3.0

[A] [abc] Values in columns for each treatment with the same letter are not significantly different at $p < 0.05$.
* $p < 0.05$.

dependent, with highest rates in summer (Stewart, unpub. data; Drew, unpub. data).

A filamentous macroalgal species common in tropical/sub-tropical waters, *Gracilaria edulis* (Rhodophyta), has been shown to be responsive to elevated nutrients (Horrocks *et al.*, 1995; Jones *et al.*, 1996). *G. edulis* was collected in Moreton Bay, Queensland (27°13′S, 153°07′E), transported to One Tree Island and incubated in the experimental patch reefs for 3 days in clear plastic containers perforated for water exchange. Following the short incubation, plants were analysed for pigment, tissue nutrient and amino acid content (Table 7). The amino acid citrulline was significantly increased under the +N and +N + P treatments. Citrulline, a 3N containing amino acid, has been invoked as a nitrogen storage compound.

Epilithic Algal Community (EAC)

The epilithic algal community is a microscopic algal biofilm, which exists on most dead limestone surfaces of coral reefs (Hatcher and Larkum, 1983). Because such surfaces are common and because the EAC is highly productive (Hatcher and Larkum, 1983), the EAC is an important contributor of food to the herbivores of coral reefs. Also because the EAC is so productive it has generally been thought that the EAC would be responsive to added nutrients (Klumpp and Mackinnon, 1992). Thus the EAC was a major focus of work during the ENCORE project.

Standing crop, growth and primary production of EAC on *Porites* coral plates were examined on 6 occasions during ENCORE. Standing crop was measured as dry weight or as chlorophyll *a*. Growth was measured as increment in dry weight over 7 days. Primary production was measured as oxygen exchange. In all experiments (across all seasons) no significant effect of nutrient enrichment was found in any of the treatments.

To test whether EAC would respond to higher nutrient levels, they were incubated in stirred nutrient-enriched seawater at two levels of nutrient enrichments: 80 μM and 200 μM NH_4Cl or KH_2PO_4 or both combined. EAC from control patch reefs were used in these experiments and showed no significant enhancement of production after 24 h incubation, either to enrichment by N or P or N+P (see Fig. 5). EAC from +N+P patch reefs were also treated in the same way and showed no response at any time.

Analyses of EAC that had been grown for 6 months in the different patch reefs showed no significant difference in the amount of chlorophyll *a* between treatments (Larkum and Koop, 1997), indicating that there was no difference in biomass of the EAC between treatment patch reefs. Based on the rates of uptake from 2 to 20 μM for each treatment from June 1994, there was a trend for uptake of ammonium to be suppressed in the EAC grown in +N patch reefs. At 20 μM, rates for +N patch reefs were 3.7×10^{-3} μM cm^{-2} min^{-1}, compared with control patch reefs (8.1×10^{-3} μM cm^{-2} min^{-1}) and +P patch reefs (4.5×10^{-3} μM cm^{-2} min^{-1})). This is consistent with the

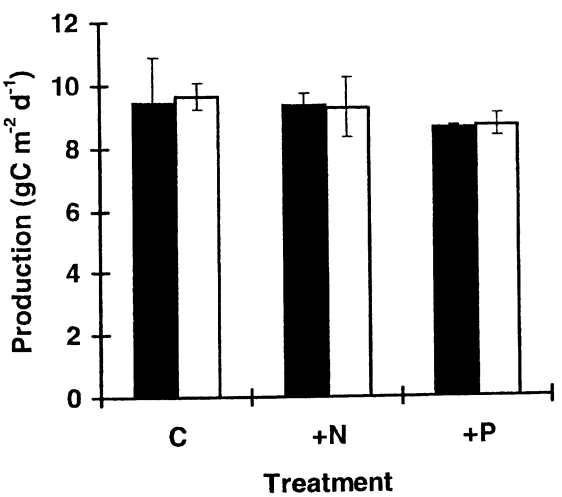

Fig. 5 Effect of short-term enrichment with 80 M NH_4Cl on primary production (g Cm^{-2} day^{-1}) of 12-month-old coral blocks from fertilized patch reefs (+N = 10 μM NH_4Cl; +P = 2 μM KH_2PO_4) and control (C) patch reefs. N = 6SD. Production was measured as oxygen evolution over 30 min. (minus dark treatments) using chambers as described in Larkum and Koop (1997). Dark bars are unenriched EAC, light bars are from 80 μM NH_4Cl incubations.

hypothesis that algae conditioned to higher concentrations of ammonium in the +N patch reefs, would show a lower capacity for ammonium uptake when exposed to episodic increases in concentrations of ammonium (Fujita, 1985).

Rhodoliths

Crustose coralline algae were a conspicuous component of the algae of the experimental patch reefs. These algae are important calcifiers in reef environments contributing to the calcium carbonate reserves and acting as important consolidators of the reef structure (Borowitzka and Larkum, 1986). Phosphate has been suggested to be an inhibitor of calcification in algae (Simkiss, 1964). Kinsey and Davies (1979) reported inhibition of calcification as a result of enrichment of a patch reef at One Tree Reef with urea and phosphate, although the calcifying agent was not identified. There is one report of enhanced levels of phosphate inhibiting the calcification of tropical coralline algae in the field (Björk *et al.*, 1995). These algae are difficult to work with experimentally because they encrust the substratum and other organisms. Rhodoliths (Larkum *et al.* (in press)) were therefore chosen as the experimental organism for this work since they are discrete semi-spherical bodies comprising of a single species (in this case *Lithophyllum kotchyanum*). Replicate rhodoliths were set out on plastic supports and monitored throughout ENCORE. Growth of replicate rhodoliths ($n = 12$) in each experimental patch reef was measured by increase in buoyant weight over periods of 2–4 months throughout ENCORE (Larkum *et al.* (in press)). No effect of enrichment by N or P was found at any time ($p > 0.05$ ANOVA; see Fig. 6 for four seasonal observations).

Calcification was measured by the alkalinity anomaly method at 3 seasons (March and June 94, August 95) and showed no significant effect of nutrient enrichment ($p > 0.05$; ANOVA; Larkum *et al.* (in press)). Growth rates (summer: 0.125 mg g^{-1} day^{-1} and winter 0.076 mg g^{-1} day^{-1}), primary production (6–14 g C m^{-2} day^{-1}), gross calcium carbonate increase (0.36 g g^{-1} yr^{-1} or $\sim$ 1.15 kg m^{-2} yr^{-1}) and calcification rates (70–180 mg $CaCO_3$ g (buoyant weight)$^{-1}$ h^{-1}, with summer rates $\sim$ twice those of winter) were all comparable with other work for tropical crustose coralline algae (Chisholm, 1988; Matsuda, 1989).

Animals

Different aspects of the biology of five major groups of animals were studied as part of the ENCORE project. These were: stomatopods, fish, reef-building corals, soft corals and giant clams. Though not all-inclusive, these groups represent a major proportion of the animal life present in the ENCORE patch reefs. Each of these groups is dealt with as a separate section below. Because of the importance of the symbiotic dinoflagellates (zooxanthellae) associated with many of these animals, one section is devoted to the responses of zooxanthellae within the experiment.

Stomatopods

Gonodactyloid stomatopods are benthic reef crustaceans that typically inhabit shallow reef flats and seagrass communities and live in cavities in hard substrata such as dead coral rubble. Recent studies have demonstrated a sensitive response of gonodactyloid stomatopod assemblages to marine pollution, including eutrophication. They have shown a reduction in abun-

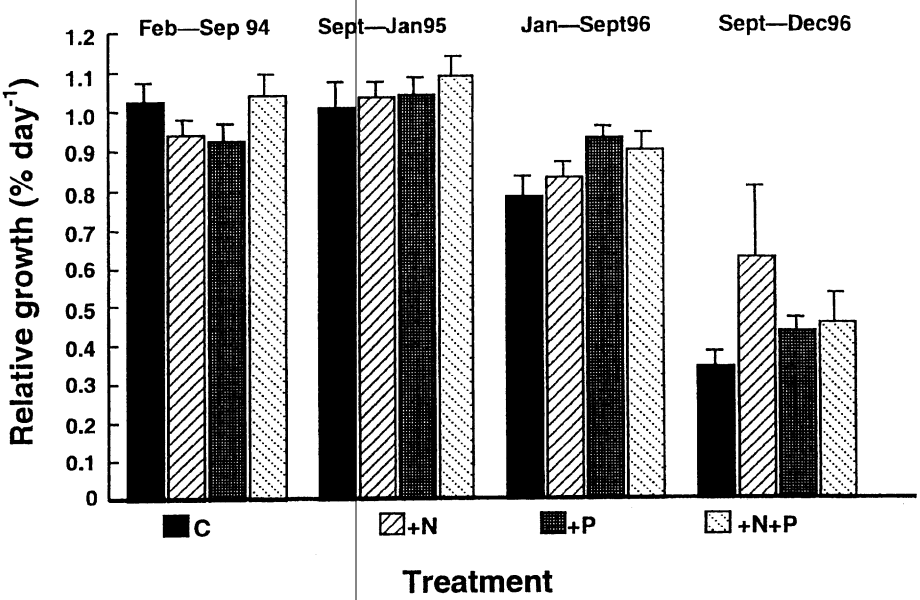

Fig. 6 Effect of nutrient treatments and season on mean relative growth (% day^{-1}) of rhodoliths. Error bars are standard errors of means ($n = 12$).

dance and species richness and apparent recruitment failure with increasing pollution (Steger and Caldwell, 1993; Erdmann and Caldwell, 1997). In this study, the effect of nutrient enrichment on stomatopod recruitment was examined by adding suitable stomatopod habitat (in the form of sun-dried, tagged coral rubble pieces) to the experimental patch reefs, and later collecting the rubble and extracting all animals which had recruited to the rubble during the experiment. A preliminary survey of the surrounding reef flats and patch reefs indicated that the shallow water stomatopod fauna of One Tree Island is dominated by four species (*Gonodactylaceus mutatus, Gonodactylinus viridis, Gonodactylus childi*, and *Haptosquilla glyptocercus*), and only these species were considered in this experiment. Additionally, only those animals which had clearly recruited to the rubble during the experiment (conservatively, those animals ≤ 18 mm total length) were counted. This experiment was conducted during the high-loading phase of ENCORE, from May, 1995 through January 1996.

Results indicated that recruitment of gonodactyloid stomatopods is negatively affected by nutrient enrichment (Fig. 7). One-way ANOVA shows that the mean recruit densities in the 4 nutrient treatments were significantly different ($F = 5.85; df = 3,8; p = 0.02$). Multiple comparisons of the 4 means using Tukey's studentized range test (controlling procedure-wise Type I error rate at $p < 0.05$) revealed that rubble from the combined +N+P treatment patch reefs had significantly lower stomatopod recruit densities than rubble from the control, +P and +N patch reefs.

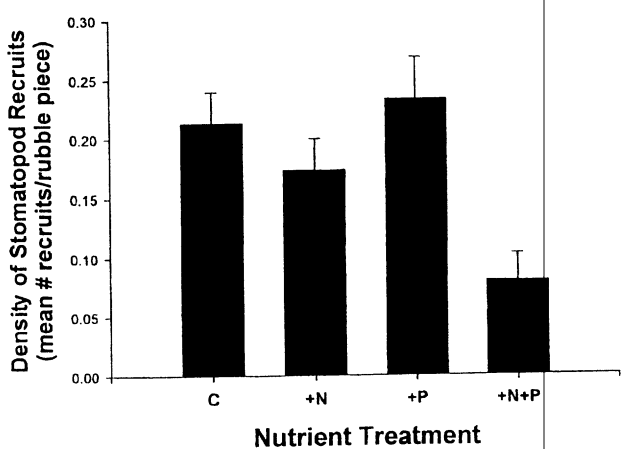

Fig. 7 Mean recruit density of stomatopods (average number of recruits per rubble piece) for each nutrient treatment. Bars indicate standard error.

Fish

Earlier studies at One Tree Island (Hatcher and Larkum, 1983) suggested that grazing fishes remove large quantities of epilithic algae, and so may mask the effects of nutrient enrichment on algal communities. Grazing fish assemblages may respond to nutrient enhancement by changes in: (1) fish density, (2) individual grazing rates, and (3) nesting and egg production.

Fish grazing rates. The majority of roving grazers were small (< 10 cm) parrotfishes, in groups which entered patch reefs with the rising tide (Fig. 8, see also Hawkins, 1995; Booth, 1997, 1998). Densities of fish varied but they could not be related to nutrient treatment (Booth unpub. data). Overall, small scarids removed 1.32 g algae m^{-2} day^{-1} in summer and 0.35 g m^{-2} day^{-1} in winter (Table 8; Booth, 1998). In contrast, territorial damselfish removed 2.0 g algae m^{-2} day^{-1} in summer and 1.0 g m^{-2} day^{-1} in winter. Although the species composition of the EAC differed significantly

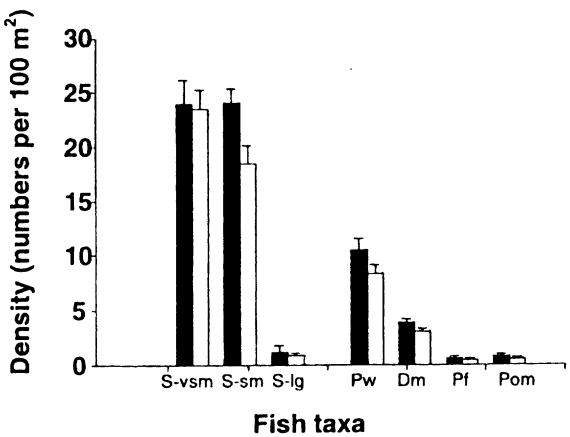

Fig. 8 Density of grazing fish taxa on patch reefs in One Tree Island lagoon in summer (dark bars) and winter (light bars) censuses ($n = 4$ censuses in winter, $n = 5$ censuses in summer, SE shown). S-vsm: *Scarus* spp (< 6 cm TL); S-sm; *Scarus* spp (6–10 cm TL); S-lg: Scarids, acanthurids, siganids (> 10 cm TL), Pw: *Pomacentrus wardi*; Dm; *Dischistodus melanotus*; Pf: *Pomacentrus flavicauda*; Pom; other territorial pomacentrids. (from Booth, 1998).

TABLE 8

Summary of grazing pressure and its components for scarids and pomacentrids (*P. wardi*) on reef tops in One Tree Island lagoon (summer–winter mean values indicated) (from Booth, 1998).

Taxon	Coverage (%)	Feeding rate (bites day^{-1})	Food intake (mg DW $bite^{-1}$)	Density (m^{-2})	Grazing pressure (g m^{-2} day^{-1})
P. wardi	21–24	3514–1366	0.6	0.95–1.25	2.0-1.0
Scarids	89–86	8400–3168	0.05	0.29–0.30	0.48–0.12
(< 6 cm TL) Scarids	89–86	6600–2100	0.4	0.32–0.27	0.84–0.23
(6–10 cm TL) Large scarids	89–86	*ca* 1000	2	*ca* 0.01	*ca* 0.02

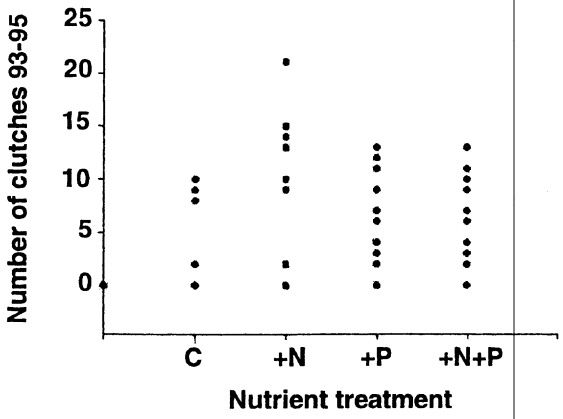

Fig. 9 Total number of clutches (•) received by individual males on patch reefs with different nutrient-enrichment treatments as part of the ENCORE study (from Beretta and Booth, 1998).

inside and outside damsel fish territories (*Pomacentrus wardi*), standing crop was similar (Booth, 1997, 1998).

Fish reproduction. P. wardi males attracted females to lay clutches of eggs between new moon and full moon (peaking at 3/4 moon) in November/December 1993, December/January 1994/95 and December 1995 (Beretta and Booth, 1998). Some males attracted more females and hence guarded significantly more clutches than others, but there were no apparent nutrient-treatment effects (Fig. 9). Lipid analyses from eggs from different nutrient treatments showed no differences (Booth and Beretta unpub. data).

Reef-building corals

For most experiments, reef-building corals were collected and transplanted into the ENCORE patch reefs. Corals were collected as either entire colonies or large portions of colonies, or colonies were broken into small sub-colonies ('nubbins'; Spencer Davies, 1989) that were deployed on plastic racks within the experimental patch reefs. Nubbins were 5–10 cm in diameter, while other colonies ranged in size up to 30–40 cm in diameter.

Coral mortality. Coral mortality was studied by monitoring survivorship among coral colonies or nubbins introduced into the ENCORE patch reefs. No differences in survivorship between treatments were detected during the initial low-loading phase of the ENCORE project. Adding nutrients, however, increased the mortality of some coral species during the second, high-loading phase. Mortality rates of two morphotypes of *P. damicornis* ('brown' and 'pink' = pocilloporin containing), (Takabayashi and Hoegh-Guldberg, 1995; Dove *et al.*, 1995) were significantly higher in patch reefs that received nutrients ($p = 0.007$) and were highest in patch reefs that received a combination of both ammonium and phosphate (271% and 211% of control mortality for brown and pink morphotypes, respectively) (Fig. 10). The mortality of nubbins of the branching coral *A. longicyathus* was not significantly different in the nutrient treatments ($p > 0.05$; Hoegh-Guldberg, unpub. data) although it was generally about 10–20% higher in +N and +P patch reefs.

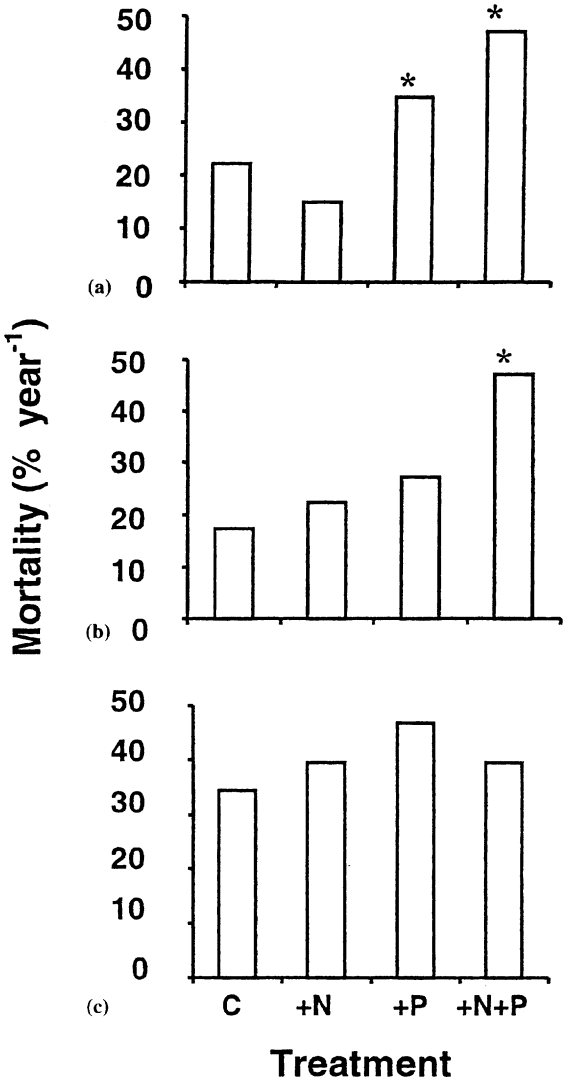

Fig. 10 Mortality rates (percent of colonies dying per year) of (a) *Pocillopora damicornis* (brown morphotype), (b) *P. damicornis* (pink) and (c) *Acropora longicyathus* after nine months in different treatments of the ENCORE project at One Tree Island reef. Shown are means and 95% confidence intervals. Asterisks indicated differences significant at $p = 0.05$. Adapted from Hoegh-Guldberg (1999).

Mortality was lower in larger coral colonies (Ward, 1997; Bucher unpub. data; Steven unpub. data). Aside from cyclone damage, larger portions of *A. longicyathus* and *A. aspera* (up to 5 kg in weight) that were transplanted into patch reefs for reproduction and growth studies suffered little mortality. Some predation by *Drupella* sp, folliculinids (a protist) and mortality from bleaching and disease were limited to single patch reefs and could not be linked with nutrient treatments.

TABLE 9

Summary of coral growth responses to the high-loading phase of the ENCORE nutrient treatments.

Parameter measured	Species used	Response		References
		Ammonium	Phosphate	
Linear extension	*Acropora longicyathus*	Reduced	Increased	Bucher and Harrison (unpub. data)
	A. palifera	No effect	Increased	
	Stylophora pistillata	Increased	Increase with ammonium	Steven (unpub. data) Takabayashi (1996)
Injury repair	*A. longicyathus*	Reduced	No effect	Bucher and Harrison (unpub. data)
Calcification (buoyant weight increments)	*A. longicyathus*	Increased[a]	Increased	Bucher and Harrison (unpub. data)
	A. aspera	No effect	No effect	
	A. palifera	Decreased	Increased	Steven (unpub. data)
	S. pistillata	No effect	Decreased	
	Pocillopora damicornis	Decrease	Decreased	Takabayashi (1996) Hoegh-Guldberg (unpub. data)
	A. longicyathus	No effect	No effect	Hoegh-Guldberg (unpub. data)
Skeletal density				
Bulk density	*A. longicyathus*	Increased	Reduced	Bucher and Harrison (unpub. data)
Micro-density	*A. longicyathus*	Increased	Increased	Bucher and Harrison (unpub. data)
Tissue				
Morphology				
Mucus cell density	*A. longicyathus*	No effect	Reduced	Bucher and Harrison (unpub. data)
Free body wall thickness	*A. longicyathus*	No effect	Increased	Bucher and Harrison (unpub. data)

[a] Increased over year but seasonal (increased in winter and spring but decreased in summer).

Coral growth. A summary of coral growth responses is presented in Table 9. Three teams within the ENCORE project independently measured a number of growth parameters in a range of coral species. The species included massive, columnar, densely branched and open staghorn growth forms. Several trends in growth response were consistent across these studies. Few growth responses were detected in any of the nutrient treatments during the initial, low-loading phase of ENCORE. Marked seasonal and clonal but not treatment variability was noted for *P. damicornis* (Hoegh-Guldberg and Moreno unpub. data; Hoegh-Guldberg *et al.*, 1997) and *A. longicyathus* (Bucher and Harrison, unpub. data) during the low-loading phase. In some seasons significant differences in calcification were measured in *A. longicyathus* between nutrient treatments. There were no significant differences, however, when calcification was integrated over a full year (Bucher and Harrison, unpub. data).

During the second, high-loading phase of ENCORE, *A. longicyathus* and *A. palifera* had higher extension rates in the presence of elevated phosphate and reduced extension in ammonium treatments (Fig. 11; Bucher and Harrison, unpub. data; Steven unpub. data). In contrast to these studies, Takabayashi (1996) reported no effect of nutrient treatment on the linear extension rates of small (5–10 cm diameter) colonies of *S. pistillata* during the period of higher nutrient loading.

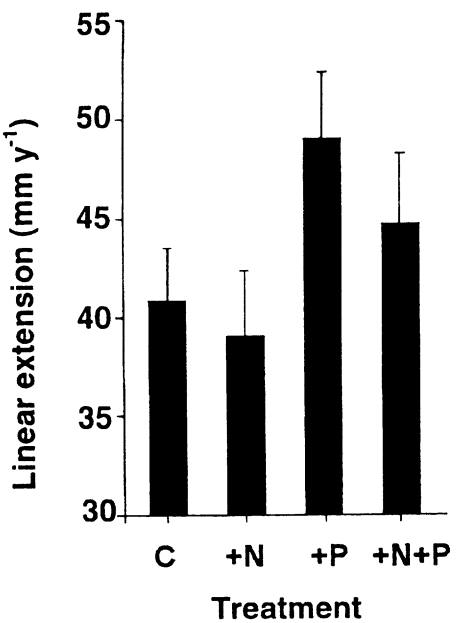

Fig. 11 Mean linear extension rates of *A. longicyathus* branches during the high dose period. Orthogonal analyses of variance showed significantly greater extension ($p < 0.001$) in the presence of phosphate and no significant effect ($p = 0.06$) of ammonium on linear extension ($n = 45$ branches per treatment, error bars are standard errors).

Changes in the weight of calcium carbonate in coral skeletons were measured by changes in the buoyant weight of colonies. Skeletal material represents the majority of any coral's buoyant weight and consequently changes in buoyant weight are primarily due to skeletal growth (Bak, 1973, 1976; Jokiel *et al.*, 1978). This non-destructive method has been used to measure small changes in growth rate in corals during exposure to 'adverse' conditions (Davies, 1989, 1990, 1995). The effect of nutrient addition on calcification, as measured by buoyant weight increments, was both species and nutrient specific. Rates of change in buoyant weight decreased in the presence of N and/or P in small (5–10 cm diameter) colonies of *P. damicornis* but not in *A. longicyathus* after nine months of the high-loading phase (Hoegh-Guldberg unpub. data). Ammonium enrichment also led to a decrease in the rate of calcification of *A. palifera* (Steven and Broadbent, 1997; Fig. 12) but had no effect on *A. aspera* (Bucher and Harrison unpub. data) or *S. pistillata* (Takabayashi, 1996). The effect of ammonium on larger (> 20 cm in diameter) colonies of *A. longicyathus* was dependent on season, with increased calcification in winter and spring but decreased rates in summer (Bucher and Harrison, unpub. data). Integrated over a full year, an overall increase in calcification in this species was found in these larger colonies. This was not the case in the smaller colonies (Hoegh-Guldberg unpub. data). When combined with a reduced rate of linear extension in the larger colonies (Fig. 11), this not only produced higher bulk density than untreated *A. longicyathus* but may also explain the reduced ability of *A. longicyathus* in ammonium treatments to overgrow lesions (Table 10).

The calcification rate of both *A. longicyathus* and *A. palifera* increased in +P treatments (Bucher and Harrison unpub. data; Steven unpub. data). In contrast, +P treatments had no effect on *A. aspera* (Bucher and Harrison, unpub. data) and tended to decrease calcification in *S. pistillata* (Takabayashi, 1996; Fig. 13) and the pink form of *P. damicornis* (Hoegh-Guldberg unpub. data; Fig. 14). In *A. longicyathus*, the changes in linear extension and calcification led to a significant reduction in skeletal bulk density in phosphate treatments (Bucher and Harrison, unpub. data; Fig. 15). Scanning electron microscopy of ENCORE corals found no disruption of the orderly crystal structure in ENCORE corals (Takabayashi, 1996; Bucher unpub. data) but significant increases in micro-density (Fig. 16) suggest that some changes had occurred at the scale of crystal architecture and/or chemical composition.

TABLE 10

Numbers of unhealed wounds on *Acropora longicyathus* nubbins after six months of the high nutrient loading phase of ENCORE.

	Treatment			
	Control	+N	+P	+N+P
Number of *nubbins* with unhealed wounds (max. 45 per treatment)	5	15	4	5
Number of *colonies* with unhealed wounds (max. 15 per treatment)	2	8	3	3
Number of *reefs* containing unhealed nubbins (max. 3 per treatment)	2	3	2	2

Coral photophysiology. Photosynthetic performance of the nubbins of two species of corals, *P. damicornis* and *S. pistillata*, subjected to the ENCORE treatments showed no significant difference from control corals during the initial, low-loading phase of ENCORE (Hoegh-Guldberg and Moreno, unpub. data). Nutrient effects were observed in corals during the second, high-loading phase. Takabayashi (1996) measured the maximum gross photosynthetic rate (p_c g max), respiratory rate (r_c), maximum net photosynthetic rate (p_c n max),

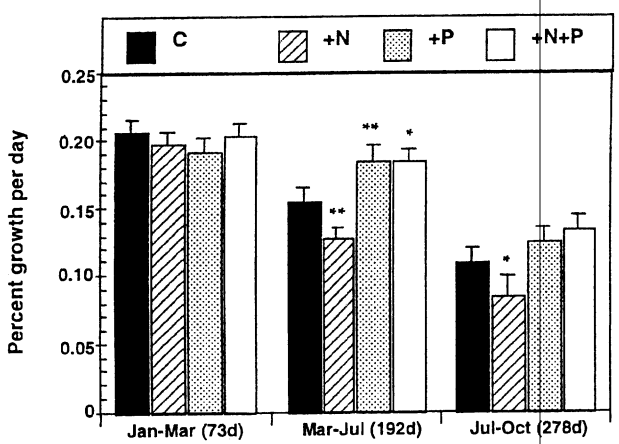

Fig. 12 Adjusted mean percent daily growth (% day^{-1}) of *A. palifera* nubbins grouped by nutrient treatment over three time periods. Errorbars are mean standard errors. Asterisks indicate treatment means (adapted from Steven, 1999); significantly different from controls: * $p < 0.1$, ** $p < 0.05$.

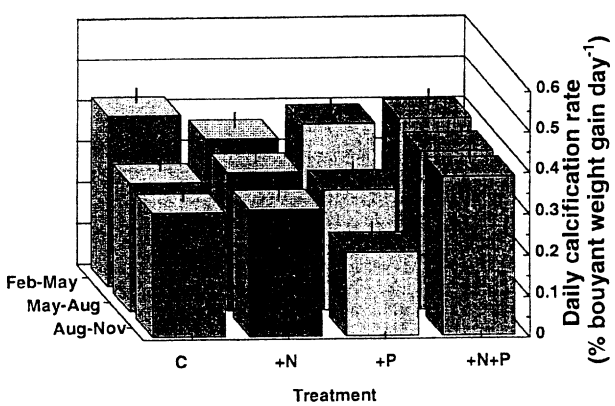

Fig. 13 Daily calcification rate (mean ± SE) of the nubbins of *S. pistillata* during (a) the first three-month period ($n = 15$), (b) the second three-month period ($n = 10$), and (c) the third three-month period ($n = 5$) of the ENCORE nutrient treatment exposure (adapted from Takabayashi, 1996).

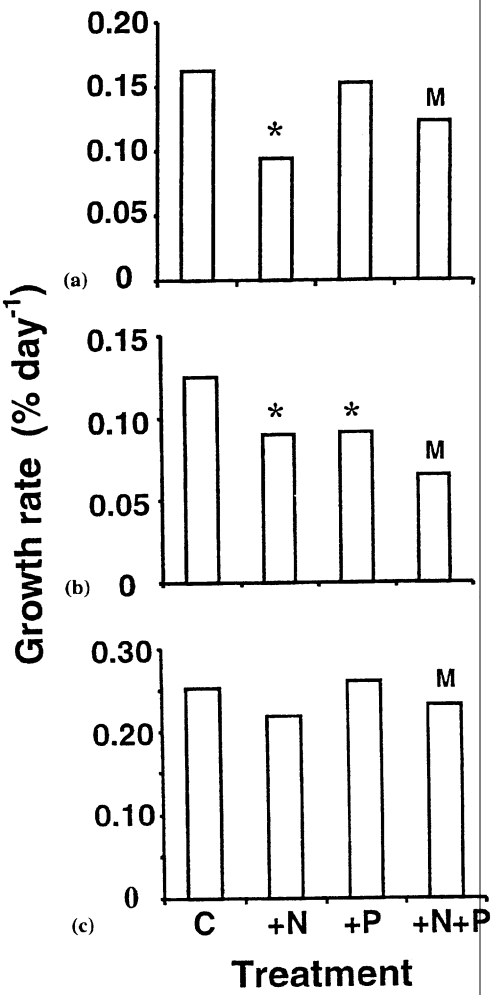

Fig. 14 Growth rates of (a): *P. damicornis* (brown morphotype), (b): *P. damicornis* (pink morphotype) and (c): *A. longicyathus* after nine months in different treatments of the ENCORE project at One Tree Island reef. Shown are means and 95% confidence intervals. Asterisks indicated differences significant at $p = 0.05$. M indicates the fact that the mean is shown for comparison for the N + P treatments but that the loss of nubbins through mortality prevented the data from this treatment being included in the associated ANOVA. The means were calculated from $n = 22$ (*P. damicornis*, brown morphotype), $n = 21$ (*P. damicornis*, pink morphotype) and $n = 21$ (*A. longicyathus*). (Hoegh-Guldberg, unpub. data).

and photosynthetic efficiency (α) in nubbins of *S. pistillata* 3 and 9 months after the start of the high-loading phase. In this study, elevated concentrations of phosphate increased the photosynthetic production and respiration of corals after 3 months of exposure to the high load. The addition of ammonium did not affect these parameters. After 9 months, however, the apparent stimulation of production and consumption by phosphate were replaced by an almost twofold increase in production and consumption per surface area in corals exposed to ammonium (Table 11). This was due to an increase in the number of zooxanthellae (and hence chlorophyll *a*) per surface area, as indicated by a dampening of this difference when rates were standardized to chlorophyll (Table 11).

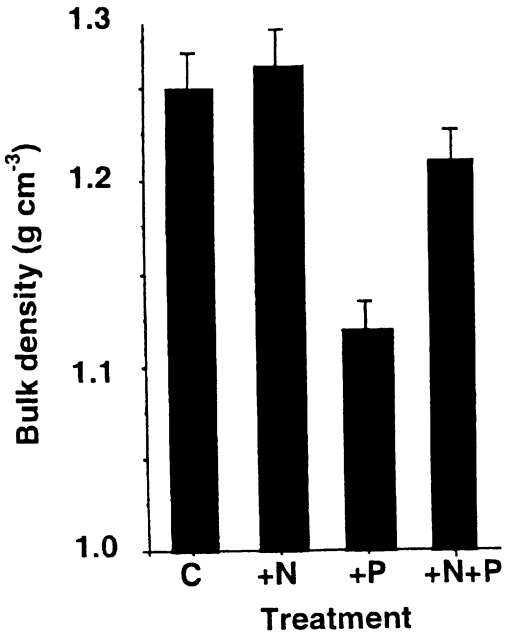

Fig. 15 Mean bulk density of branch tips from *A. longicyathus* grown during the high-loading phase of the ENCORE study. Orthogonal analyses of variance showed significantly lower bulk density (i.e. greater porosity) ($p < 0.001$) in the presence of phosphate and significantly higher bulk density ($p = 0.005$) in the presence of ammonium in *A. longicyathus* branch tips ($n = 45$ branches per treatment, error bars are standard errors).

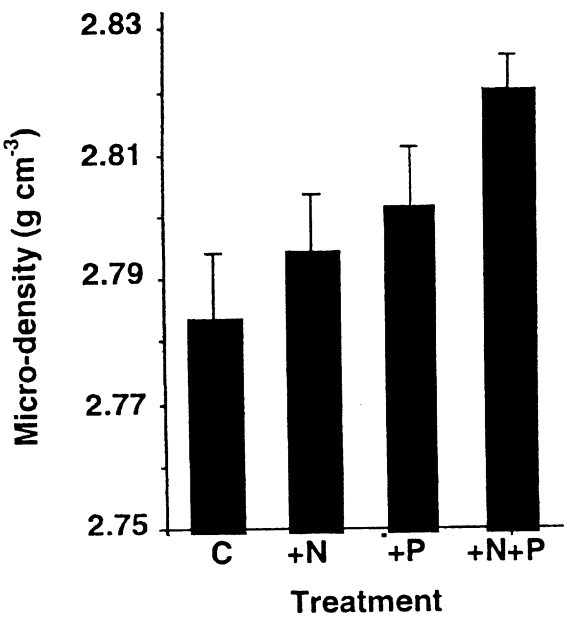

Fig. 16 Mean micro-density of branch tips from *A. longicyathus* grown during the high-loading phase of the ENCORE study. Orthogonal analyses of variance showed significantly greater micro-density ($p < 0.001$) in *A. longicyathus* branch tips in both the ammonium and phosphate treatments ($n = 45$ branches per treatment, error bars are standard errors).

The addition of ammonium also increased the compensation irradiance (I_c) and the intercept irradiance (I_k) after 3 months (Table 12). After 12 months, the

TABLE 11

Maximum gross photosynthetic rate ($p_{c\ g\ max}$), respiratory rate (r_c), maximum net photosynthetic rate ($p_{c\ n\ max}$), and initial slope of $p_{c\ g\ max}$ (α) measured after nine-month incubation in ENCORE treatments during the high-loading phase of the study.[a]

Treatment	$p_{c\ g\ max}$ (μmol O_2 h^{-1} x)	r_c (μmol O_2 h^{-1} x)	$p_{c\ n\ max}$ (μmol O_2 h^{-1} x)	$\alpha(10^{-2}$ μmol O_2 m^2 s μE^{-1} h^{-1} x)
Area				
C	0.95 ± 0.19	0.24 ± 0.05	0.71 ± 0.14	1.26 ± 0.39
+N	1.80 ± 0.92	0.58 ± 0.24	1.23 ± 0.69	3.78 ± 1.44
+P	0.77 ± 0.17	0.24 ± 0.07	0.53 ± 0.11	3.08 ± 1.20
+N+P	0.82 ± 0.24	0.22 ± 0.06	0.60 ± 0.18	0.64 ± 0.13
Chl a				
C	2.37 ± 0.73	0.59 ± 0.02	1.78 ± 0.57	2.84 ± 0.77
+N	2.28 ± 0.99	0.74 ± 0.03	1.54 ± 0.76	7.50 ± 0.41
+P	1.60 ± 0.27	0.47 ± 0.09	1.13 ± 0.20	5.95 ± 0.20
+N+P	1.75 ± 0.78	0.46 ± 0.02	1.29 ± 0.57	1.06 ± 0.25

[a] The figures are expressed as means ± S.E. and are adapted from Takabayashi (1996). Each treatment is calculated per area (cm^{-2}) and also standardized to chlorophyll *a*; thus 'x' in the units is for 'cm^{-2}' in the 'area' table and 'chl a^{-1}' in the 'chl *a*' table.

TABLE 12

The irradiance at which the initial slope of the gross photosynthesis intercepts the horizontal asymptote (I_k), the compensation irradiance (I_c), and the ratio between $p_{c\ g\ max}$ and r_c measured in coral subcolonies after three-month incubation in the ENCORE treatments. The figures are expressed as mean ± S.E. ($n = 2$).

Treatment	I_c (μE m^{-2} s^{-1})	I_k (μE m^{-2} s^{-1})	$p_{c\ g\ max}/r_c$
C	58.9 ± 7.71	141.7 ± 16.0	3.55 ± 0.146
+N	73.0 ± 10.6	206.6 ± 61.7	3.58 ± 0.600
+P	48.1 ± 3.06	121.8 ± 14.0	3.55 ± 0.300
+N+P	71.5 ± 5.95	221.2 ± 25.4	4.07 ± 0.225

stimulation by ammonium had disappeared and phosphate caused a dramatic decrease in I_c and I_k but this did not change the ratio of gross photosynthesis to respiration (Fig. 17). Nutrient treatment significantly ($p > 0.05$) affected I_c and the initial slope (α) only. The Student–Newman–Keuls test revealed that I_c from the +N+P treatment was significantly greater ($p < 0.05$) than from the +P treatment. Also, the α in the +P treatment was significantly greater ($p < 0.05$) than that in the +N+P treatment.

Coral reproduction. Many aspects of sexual reproduction in acroporid species of corals were affected by nutrients; most were inhibited but some were enhanced. Effects were dependent on time, species and the nutrient in question.

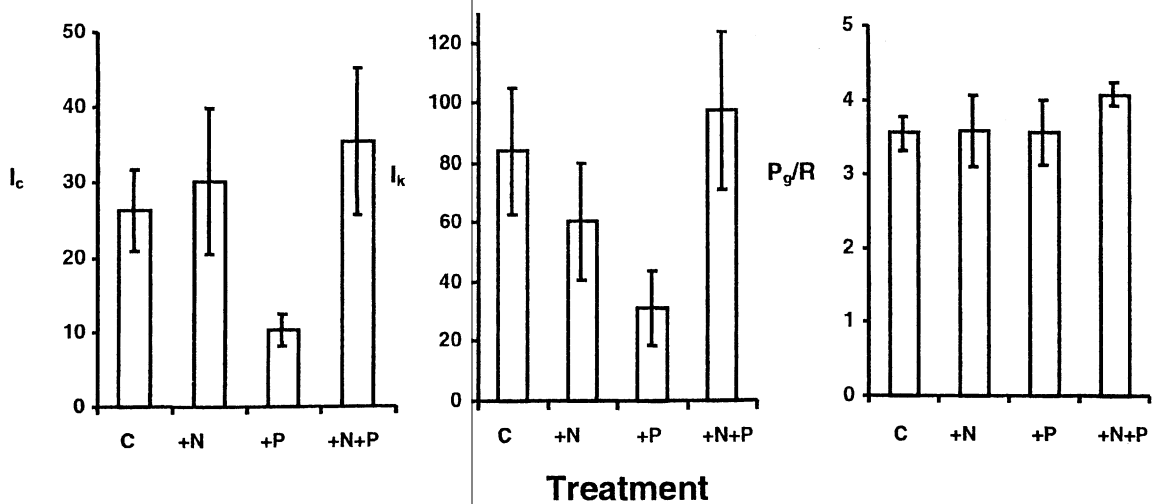

Fig. 17 The irradiance at which the initial slope of the gross photosynthesis intercepts the horizontal asymptote (I_k), the compensation irradiance (I_c), and the ratio between $p_c g$ max and r_c measured in the coral sub-colonies after the nine-month incubation in the second (high-loading) regime of the ENCORE study. The figures show mean ± SE ($n = 2$) and are adapted from Takabayashi (1996).

Ammonium

During the sampling period from 1993 to 1995 corals exposed to elevated nitrogen produced significantly smaller and fewer eggs and contained significantly less testes material than those not exposed to nitrogen (Ward and Harrison, unpub. data). Fertilization rates of *A. longicyathus* eggs were significantly reduced by low concentrations of nitrogen (down to 1M ammonium). Fertilized eggs showed a significant increase in the number of irregular embryos and of embryos that stopped development at the first cleavage stage (Harrison and Ward, unpub. data). Gametes exposed to +N+P in the laboratory had very low fertilization rates (Fig. 18). In similar trials using gametes of the brain coral *G. aspera*, the percentage fertilization was significantly reduced only following exposure to 50 μM + N + P, but there were significantly more deformed embryos developed following exposure of gametes to +N and +N+P treatments (Harrison and Ward, unpub. data).

In settlement trials using larvae of *A. longicyathus* in 1993, settlement rates were reduced by nitrogen treatments with very low settlement in the nitrogen plus phosphorus treatment (Ward and Harrison, 1997). Settlement tiles were mapped and rescored every three months until 1996 to monitor settlement, mortality and spat growth and on these tiles nitrogen reduced settlement of spat of both spawning and brooding species of corals (Ward and Harrison, unpub. data; Fig. 19).

Phosphate. Exposure to phosphorus enrichment affected a variety of reproductive activities in the coral species examined. Contrary to the pattern found in controls and other treatments, corals exposed to phosphorus alone did not have a reduction in the number of eggs per polyp as the gametogenic cycle progressed. The egg numbers prior to spawning were significantly higher than those of corals from controls and other treatments (Ward and Harrison, 2000). Egg size was reduced by phosphorus treatments and these patterns were consistent for both

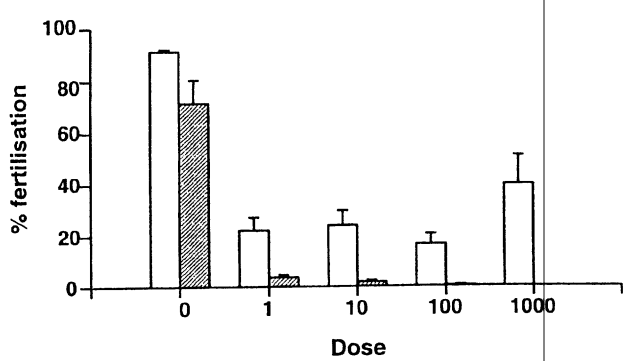

Fig. 18 The percentage fertilization recorded in fertilization trials using eggs and sperm from *A. longicyathus*, which had been exposed to added N and P in the laboratory. Doses were 0, 1, 10, 100 and 1000 M of N and P above background levels. The blank columns represent cross 1 and the shaded columns cross 2. Error bars are standard errors.

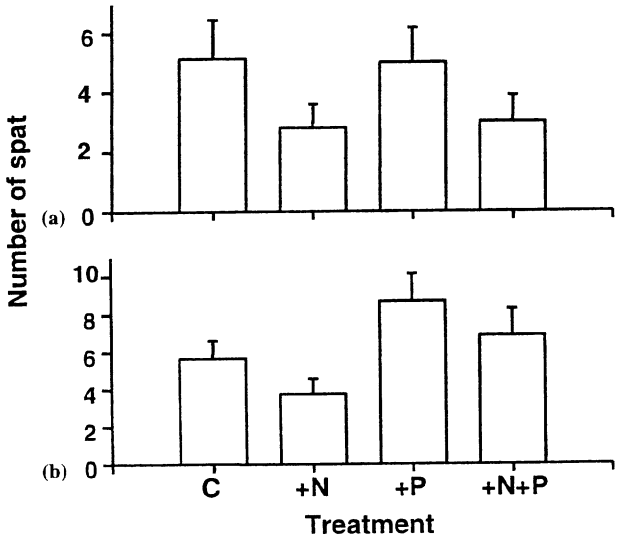

Fig. 19 The number of spat of spawning (a) and brooding coral species, which settled on pairs of terracotta tiles in different nutrient treatments between November 1994 and January 1996. These data do not take account of which spat survived; they are of settlement only. Error bars are standard errors.

A. longicyathus and *A. aspera*. Just prior to spawning, eggs from all colonies exposed to phosphorus alone were very bright red in contrast to eggs in corals from other treatments, which ranged from cream to red with no consistent pattern (Ward, 1997). Phosphorus dramatically reduced fertilization rates of *A. longicyathus* and significantly increased the incidence of irregular embryos and embryos that stopped developing at the first cleavage stage (Harrison and Ward, unpub. data). In fertilization trials with gametes of *G. aspera*, there was a significant increase in the percentage of irregular embryos formed after they were exposed to slightly elevated levels of phosphorus (> 0.5–1 μM) (Harrison and Ward, unpub. data). During the 1993 settlement trials, phosphorus significantly reduced settlement rates and this pattern continued when the tiles were rescored during 1994. When tiles were rescored from November 1994 to January 1996, the phosphorus treatments enhanced the settlement of spat of brooding coral species, but did not affect the settlement of spat of broadcast spawning coral species (Ward and Harrison, unpub. data).

Lipid levels in corals. Lipid levels were monitored in the corals studied for reproduction (*A. longicyathus* and *A. aspera*; see section above). Exposure to elevated nitrogen reduced the amount of lipid in the tissues of the corals, while exposure to phosphorus increased the amount of lipid present at various times during the ENCORE experiment. Reproductive material of corals is rich in lipids and our results followed the general patterns found for the measures of fecundity in these species under similar treatments. Samples were also taken in February 1995 before the gametogenic cycles

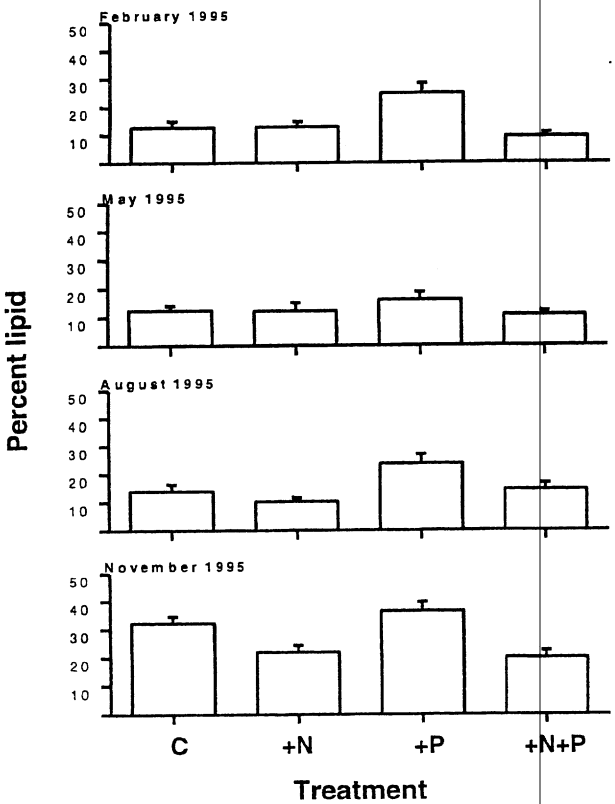

Fig. 20 The percentage of lipid in the tissue of *A. longicyathus* transplanted into experimental ENCORE patch reefs at One Tree Island reef in samples taken in February, May, August and November 1995. Error bars are standard errors.

commenced and the same patterns were observed (Ward, 1997). These results show that even very slight increases in levels of nitrogen and phosphorus can have large effects on lipid levels in coral tissues (Fig. 20).

Soft corals

Major physiological/biochemical indicators were measured in *Sarcophyton ehrenbergi*, a soft coral common on the Great Barrier Reef. In addition, a number of effects not directly related to nutrient enrichment were investigated, e.g. effects of transplantation and competition with a hard coral species, *P. damicornis*. These studies are reported elsewhere (Tentori *et al.*, 1997).

None of the nutrient treatments showed any effects on: (1) concentrations of sarcophytoxide (a terpene active in defence and competition), (2) levels of fatty esters (the primary lipid energy storage and membrane component of these corals), and (3) the ratio of terpene to lipid (an indicator of physiological change used to indicate stress in soft corals) in *S. ehrenbergi*. This study has shown that soft corals are not sensitive indicators of nutrient-induced stress in coral reefs (Fleury *et al.*, 2000). *Sarcophyton* species are common on inshore reefs, and so it is not surprising that they are able to accommodate a wide range of nutrient conditions without adverse effects.

Giant clams

One Tree Island is outside the geographic limit of all clam species with the exception of *T. maxima*. This species is present in significant numbers in the lagoon, in the patch reefs and on the reef front. For the ENCORE studies clams in two size classes (65–100 and 200–220 mm) from the One Tree Island reef crest were randomly transplanted into the experimental patch reefs (Ambariyanto and Hoegh-Guldberg, 1997; Belda-Baillie *et al.*, 1998) and then used for a range of biochemical, physiological and ecological measurements.

Clam growth. The dependence of tridacnids on the photosynthetic capacity of their symbiotic zooxanthellae population for much of their energy requirements could be expected to influence the biomass parameters of the whole animal. The nutrients ammonium and phosphate are essential to growth in photosynthetic autotrophs and an increase in availability in what is generally regarded as a low nutrient environment might be expected to cause an increase in biomass of the clam. Simple biomass parameters were measured to determine if any gross changes occurred during the course of the enrichment. A number of growth parameters were measured in the course of the first and second enrichment phases.

Growth of clams measured as changes in shell length and buoyant weight was relatively linear over the period of this study. The percentage daily change in length and buoyant weight of the clams was influenced by season (Ambariyanto and Hoegh-Guldberg, 1997). The highest growth and calcification rates were found during summer and autumn months. These rates were almost double those measured during the winter and spring months (Ambariyanto, 1996; Ambariyanto and Hoegh-Guldberg, 1997; Ambariyanto and Hoegh-Guldberg, 1999a).

There was no effect of nutrient enrichment on the growth (% change in shell length per day, Ambariyanto and Hoegh-Guldberg, 1997) of clams during the initial, low-loading phase of the experiment. In the second phase, however, differences were found after 12 months of nutrient enrichment. +N and +N+P enriched clams exhibited significantly greater growth in shell length than the control and +P treatments (Ambariyanto and Hoegh-Guldberg, 1997). With the exception of the three-month measurement in the first phase there was no significant difference in the % change of the clams' buoyant weight per day. In addition, neither the tissue wet weight, the protein content per gram of clam mantle nor the C:N ratio of the mantle tissue was significantly affected by nutrient enrichment in any of the nutrient treatments. However, changes in the N:P ratio were observed in the larger clams. Addition of P or N, but not N+P, caused corresponding changes in the N:P ratio (Ambariyanto and Hoegh-Guldberg, 1999b).