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Interactions among chronic and acute impacts on coral recruits: the importance of size-escape thresholds

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Abstract. Newly settled recruits typically suffer high mortality from disturbances, but rapid growth reduces their mortality once size-escape thresholds are attained. Ocean acidification (OA) reduces the growth of recruiting benthic invertebrates, yet no direct effects on survivorship have been demonstrated. We tested whether the reduced growth of coral recruits caused by OA would increase their mortality by prolonging their vulnerability to an acute disturbance: fish herbivory on surrounding algal turf. After two months' growth in ambient or elevated CO₂ levels, the linear extension and calcification of coral (*Acropora millepora*) recruits decreased as CO₂ partial pressure (*p*CO₂) increased. When recruits were subjected to incidental fish grazing, their mortality was inversely size dependent. However, we also found an additive effect of *p*CO₂ such that recruit mortality was higher under elevated *p*CO₂ irrespective of size. Compared to ambient conditions, coral recruits needed to double their size at the highest *p*CO₂ to escape incidental grazing mortality. This general trend was observed with three groups of predators (blenny, surgeonfish, and parrotfish), although the magnitude of the fish treatment varied among species. Our study demonstrates the importance of size-escape thresholds in early recruit survival and how OA can shift these thresholds, potentially intensifying population bottlenecks in benthic invertebrate recruitment.

Key words: climate change; coral; herbivory; ocean acidification; recruitment; size-escape threshold.

INTRODUCTION

The supply of new recruits is critical to the maintenance and recovery of invertebrate populations (Connell and Keough 1985). Yet, following settlement from the plankton to the benthos, 50–90% of benthic invertebrate recruits do not survive beyond three months (Gosselin and Qian 1997). Owing to their small size, recruits generally have much higher mortality relative to their adult counterparts, as small individuals are either completely unharmed or killed outright by disturbances, whereas larger individuals may experience partial mortality (Hughes and Jackson 1985, Babcock 1991). A prominent cause of mortality for recruiting benthic organisms is direct or incidental predation by grazers. However, size-escape thresholds have been reported beyond which recruit vulnerability to predation

is reduced across many taxa, including barnacles (Connell 1985, Navarrete 1996), mussels (Paine 1976, Wootton 1993), corals (Raymundo and Maypa 2004, Box and Mumby 2007), and fish (Rice et al. 1993). Even within very small size classes, size-escape can be an important mechanism of predator avoidance for newly settled recruits (Brock 1979, Christiansen et al. 2009), making rapid early growth vital to survivorship.

Any process that reduces the growth rate of invertebrate recruits will likely prolong the exposure to sources of mortality (pre-size-escape) and therefore elevate mortality rates. One of the most insidious disturbances to influence the growth rate of aquatic invertebrates is ocean acidification (OA). OA is a chronic disturbance that is caused by the uptake of atmospheric carbon dioxide (CO₂), which decreases seawater pH and carbonate saturation. OA can alter community structure in both tropical (Fabricius et al. 2011) and temperate ecosystems (Hall-Spencer et al. 2008), and most calcifying organisms exhibit reduced growth from elevated CO₂ partial pressure (*p*CO₂; Kroeker et al.

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2010; but see Ries et al. 2009). Due to the importance of recruitment, many recent investigations have evaluated the effects of OA on the early life-history of organisms, and have demonstrated that it can decrease the fertilization, settlement, and growth of recruits from a broad range of benthic invertebrates (Kurihara 2008, Byrne 2011). This includes the post-settlement growth of abalone (Byrne et al. 2011), oyster (Parker et al. 2009) and mussel shells (Gaylord et al. 2011), urchin spines (Byrne et al. 2011), and the linear growth (Cohen et al. 2009, Albright et al. 2010) and calcification (Cohen et al. 2009, de Putron et al. 2011) of coral recruits. However, no direct effects of OA on post-settlement survivorship of invertebrate recruits have been recorded to date.

A potentially important source of sudden mortality in newly settled invertebrates, and corals in particular, is the action of fish that graze the substrate frequently, principally to harvest epilithic algal turfs (Steneck 1988, Rotjan and Lewis 2008). Whether grazing fish actively prey upon small invertebrates or merely cause incidental damage during the process of grazing is unclear for most species, but coral mortality has been documented for various fish groups (Brock 1979, Christiansen et al. 2009). Here, we aimed to determine whether the chronic effect of OA would alter the survivorship of newly settled recruits (<5 mm) following the common, acute disturbance of herbivorous fish grazing on algal turfs. We hypothesized that OA would decrease coral recruit growth and increase their mortality following grazing by herbivorous fish, with the rate of mortality dependent on the type of fish feeding method (scraping, tearing, and combing).

METHODS

General protocol

All experiments were conducted at Heron Island Research Station (HIRS) from 29 November 2010 to 18 February 2011. Coral larvae were settled on tiles in ambient seawater and grown in a flow-through aquarium system with three CO₂ treatments. The treatments represented ambient (pH 8.04, *p*CO₂ 389 μatm [1 atmosphere = 101.3 kPa], aragonite saturation state Ω_{arag} 3.6) and two elevated (pH 7.81, *p*CO₂ 753 μatm, Ω_{arag} 2.3; pH 7.60, *p*CO₂ 1267 μatm, Ω_{arag} 1.6) concentrations of CO₂ (Appendix A). After two months, the recruits were mapped and measured to evaluate the effects of OA on post-settlement growth. The tiles were then subjected to individual grazing trials with herbivorous fish. Following the grazing trials, the coral recruits on the tiles were remapped to assess recruit mortality in each treatment.

Experimental CO₂ aquarium system

The three seawater treatments were controlled by CO₂ dosing to adjust the set pH of the seawater in 200-L sumps, following standard protocols for OA research (Gattuso et al. 2010). The seawater pH was continually measured with temperature compensated electrodes

(InPro4501VP; Mettler-Toledo, Melbourne, Victoria, Australia) that were monitored daily for calibration validity, and recalibrated to 0.01 pH units when necessary. Targeted pH levels were maintained with a control unit (Aquatronica; AEB technologies, Cavriago, Reggio Emilia, Italy), which opened solenoid valves that slowly injected CO₂ into the sumps when the pH exceeded the desired threshold. For the control treatment, CO₂-scrubbed air was bubbled into the sump when the target pH fell below the desired threshold. The alkalinity of seawater samples was measured every six hours for a period of 48 hours from each treatment over both a spring (2.8 m) and a neap (1.5 m) tidal cycle. Alkalinity replicates within a sample were measured using Gran titration until a maximum 1% error was met, using a T50 titrator (Mettler-Toledo). The carbonate chemistry of the seawater was calculated with CO2SYS (Lewis and Wallace 2006) using pH, total alkalinity, salinity (35.4‰ ± 0.2‰ [mean ± SE]; *n* = 8), and temperature as the inputs (Appendix A).

The treatment seawater was fed continually from the sumps to seven replicate tanks per treatment at a flow rate of 1 L/min. The pH of the tanks was regularly verified with a portable SG2 SevenGo pH meter (Mettler-Toledo). Tank walls were regularly cleaned to remove any algae and contained small power heads for extra seawater circulation. Replicate tanks were randomized on the table and placed under thin shade cloth and neutral density filter (Lee 298 ND 0.15; LEE Filters Limited, Andover, UK) to accommodate the heterogeneity in light, which averaged 143.5 ± 1.8 [mean ± SE] μmol·m⁻²·s⁻¹ between 06:00 and 18:00.

Coral recruit culturing

To culture the coral recruits, five gravid colonies of *Acropora millepora* were collected from Heron reef flat, placed in 60-L flow-through aquaria with ambient seawater, and their gametes collected upon spawning (29 November 2010). The gametes were fertilized and reared for five days at 25°C with ambient seawater using techniques described in Doropoulos et al. (2012). Mature planulae were settled during three days in ambient seawater onto 5 × 5 cm unglazed terracotta tiles. The tiles were pre-conditioned on the reef flat for ≥6 months to develop a microbial and encrusting algal community important to planulae attachment and metamorphosis. This short settlement period minimized any recruit calcification before the tiles were scored and randomly assigned among the replicate tanks in the experimental system. Settled recruits were grown for 60 days in the three CO₂ treatments and, during this time, the tiles were cleaned every three days by gently brushing them with a soft toothbrush to remove algal turfs. At the end of this culturing period, the number of recruits per tile, polyps per recruit, and size of each recruit were mapped on the underside of each tile with a dissecting microscope (Appendix B).

Grazing experiments and fish collection and handling

At the end of the coral growth period, an algal turf community was allowed to develop on the tiles with the recruits for five days in the three CO₂ treatments. Twelve-hour grazing trials (06:00 to 18:00) were then conducted on those tiles, with blennies (*Salarius fasciatus*), surgeonfish (*Acanthurus nigrofuscus*), and initial-phase (IP) parrotfish (*Scarus* spp.). The parrotfish appeared to be *S. rivulatus* but we could not be certain of their identification at this early initial phase. Each trial consisted of a single tile placed with a single fish in an aquarium with flowing ambient seawater and PVC fish shelter. Aquaria without a fish acted as the no-fish control. There were seven tile replicates for each CO₂ treatment × fish type combination and five tile replicates for each CO₂ treatment × no-fish control.

The blennies were caught on the reef flat (~0.3–2.5 m) with a hand net and solution of clove oil, ethanol, and seawater (1:1:9; Christiansen et al. 2009). They ranged between 75 and 120 mm in total length (TL; 93.6 ± 8.3 mm [mean ± SD]). Surgeonfish and parrotfish were caught on a shallow reef slope (~1–4 m) with a barrier net, and ranged between 135 and 180 mm (163.4 ± 13.0 mm) and 130 and 175 mm (150.9 ± 14.2 mm) TL, respectively. All fish were brought back to HIRS and placed in large aquaria with flowing seawater for ≥1 week prior to the experimental trials. Fouled tiles, live rock, and PVC structures were placed with the fish during the acclimation period. On the evening prior to each grazing trial, fish were placed in individual aquaria with shelter and flowing seawater. A tile with the coral recruits was then placed in the aquarium the following morning, minimizing handling stress to the animal. All fish were returned to the wild following the trials.

Response variables and statistical analyses

To determine the effect of OA on linear growth, the diameter of all recruits from each CO₂ treatment was measured after two months using an optical micrometer (100 μm). A total of 1392 recruits were measured and their size ranged between 0.3 and 4.7 mm in diameter (Appendix B). Each CO₂ treatment had 26 tile replicates, which were distributed among the seven replicate tanks per treatment. The effect of pCO₂ on recruit diameter was analyzed with a mixed effects ANOVA, using permutations as the data did not conform to normality and were fourth root transformed to meet the requirements of homogeneity. CO₂ treatment was a fixed factor with three levels (389, 753, 1267 μatm), with tanks random and nested in CO₂ treatment, and tiles were random and nested in tanks. Pairwise comparisons were conducted to investigate differences among CO₂ treatments.

To examine the effect of OA on recruit calcification, individual recruits were sampled after the grazing trials from the no-fish control tiles. Recruits could only be taken from the control tiles to avoid a bias associated with sampling from tiles that were subjected to grazing.

After the trials, the tiles were thoroughly rinsed in distilled water to remove any salts and oven dried at 60°C for three days. Following Cohen et al. (2009), the skeletons of four or five recruits from each tile (five tiles per CO₂ treatment) were photographed with a dissecting microscope camera, scraped from the tile using a scalpel and paintbrush, and individual recruits were weighed on a micro-balance (Mettler Toledo X5105) to 0.01 mg until a 1% error was met. The maximum diameter of each recruit was quantified using ImageJ (Abramoff et al. 2004) to standardize the mass of the recruit to mg/mm. The effect of pCO₂ on calcification was analyzed with a mixed-effects ANOVA on the raw data that conformed to normality and homogeneity, with pCO₂ as the fixed factor and tiles random and nested in pCO₂. Tukey's HSD was used to determine differences among CO₂ treatments.

The number of recruits killed by the herbivorous fish was quantified by measuring coral survivorship as dead or alive, as initial exploration of the data indicated that 97% of the recruits were either unharmed or entirely killed (i.e., there were few that suffered partial mortality). To analyze survivorship in the original model, we used fish type and pCO₂ as fixed factors, recruit diameter, number of recruits per tile, grazer length, and percent algae lost per tile as continuous predictors. The final model was determined with step-wise model simplification that identified the model with the lowest Akaike information criterion (AIC) from these possible explanatory variables and their interactions. The final model used a generalized mixed-effects model (GLMM) to analyze the binomial response of the number of recruits killed with fish type (no fish, blenny, surgeon, parrot) and pCO₂ (389, 753, 1267 μatm) as fixed factors, individual recruit diameter and number of recruits per tile as continuous predictors, and tile as a random factor.

To determine the size at which net recruit survivorship was positive (>0.5), negative (<0.5), or random (~0.5) following grazing by the different fish, we pooled pCO₂ and analyzed the mean survival of each recruit size class (in 0.1 mm units). The total number of *Acropora millepora* recruits in each size class (mm) used for the grazing trials are presented in Appendix C. Regression analyses were fitted with a logarithmic function on the mean survival for every recruit size among fish types. The GLMM was then utilized to model the survivorship of recruits up to 5.0 mm diameter following grazing by the different herbivorous fish at the three CO₂ levels.

To quantify algal turf cover on the tiles and the efficiency of turf removal by the herbivorous fish, photographs of the tiles were taken immediately before and after the grazing trials. The abundance of filamentous algal turfs was quantified by classifying whether turf was present under 100 points per tile (25 cm²) using CPCe (Kohler and Gill 2006). Turf cover was analyzed with a three-way ANOVA, using fish type (no fish, blenny, surgeonfish, parrotfish), pCO₂ (389, 753, 1267

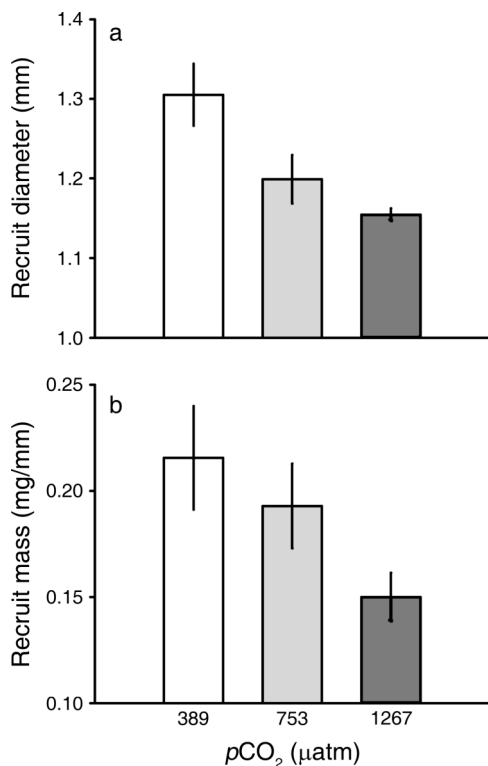


FIG. 1. The effect of ambient (389 μatm) and elevated (753 and 1267 μatm) $p\text{CO}_2$ on mean (a) linear extension ($n = 7$) and (b) calcification ($n = 5$) of *Acropora millepora* recruits after 60 days post-settlement growth. Note the different scales for the y-axes. SI conversion: 1 atm = 101.3 kPa; $p\text{CO}_2$ represents the partial pressure of CO_2 . Error bars indicate $\pm\text{SE}$.

μatm), and time (before and after) as fixed factors, and individual tiles as replicates. Permutation was used to generate P values as the raw or transformed data would not conform to normality, and pairwise comparisons were used to investigate significant terms.

Turf algal samples were collected from the no-fish control tiles to quantify the C:N ratios among $p\text{CO}_2$. The turf community was sampled under a dissecting microscope using forceps and scalpel to avoid any unwanted material (e.g., sand, coralline algae). The percentage of carbon and nitrogen was quantified with combustion by elemental analysis (ANCA-GSL; Europa, Crewe, UK) at Edith Cowan University, Western Australia. Data on C:N ratios conformed to assumptions of normality and homogeneity of variance and were analyzed using one-way ANOVA with $p\text{CO}_2$ as the fixed factor and individual tiles as replicates.

Permutational ANOVAs were conducted using Primer-E version 6 software (Clarke and Gorley 2006) with the PERMANOVA+ extension (Anderson et al. 2008). Statistica (version 10; StatSoft, Tulsa, Oklahoma, USA) was used for all other ANOVAs. AIC, GLMM, and survivorship models were conducted using R (version 2.12.0; R Development Core Team 2010) and regression

analyses with Sigmaplot (version 10; Systat Software, San Jose, California, USA).

RESULTS

OA effects on coral recruit growth

Coral recruits grown for 60 days in the elevated CO_2 treatments had significantly reduced growth and calcification compared to those grown in ambient conditions (Appendix D). The mean diameter of recruits was 8% (753 μatm $p\text{CO}_2$) and 12% (1267 μatm $p\text{CO}_2$) less than control recruits (Fig. 1a), and calcification was reduced by 11% and 30% at 753 and 1267 μatm $p\text{CO}_2$, respectively (Fig. 1b).

OA and grazing effects on coral recruit mortality

Recruits grown in elevated CO_2 levels suffered significantly higher levels of mortality than recruits grown in ambient conditions ($P = 0.002$), and survival was affected by the type of herbivorous fish grazing ($P < 0.001$; Fig. 2). Recruit mortality was highest in those grown at 1267 ($P < 0.001$), followed by 753 μatm $p\text{CO}_2$ ($P = 0.011$), compared to those grown in ambient seawater. There was no significant difference in mortality between recruits grown in 753 and 1267 μatm $p\text{CO}_2$. Parrotfish grazing caused the highest recruit mortality compared to any other fish type ($P < 0.001$), ranging from 25% in the ambient treatment, to 51% and 64% mortality of the recruits grown at 753 and 1267 μatm $p\text{CO}_2$ (Fig. 2). Surgeonfish grazing caused higher recruit mortality than controls ($P = 0.004$) but was equal to blenny grazing ($P = 0.378$), ranging from 6% mortality

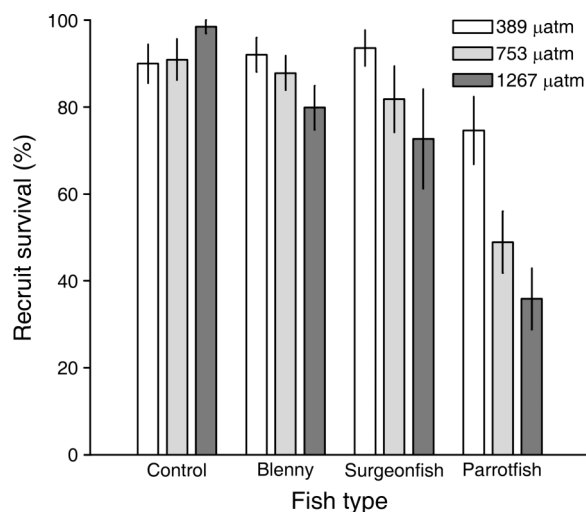


FIG. 2. Survival of coral recruits (mean \pm SE) following herbivorous fish grazing trials. Newly settled coral recruits (*A. millepora*) were grown for 60 days at ambient (389 μatm) and elevated (753 and 1267 μatm) $p\text{CO}_2$ on 25-cm² settlement tiles before the 12-h grazing trials, with one fish and one tile per trial. Treatments are: control, no fish ($n = 5$); blenny, *Salaria fasciatus*; surgeonfish, *Acanthurus nigrofasciatus*; parrotfish, initial-phase *Scarus* spp. ($n = 7$).

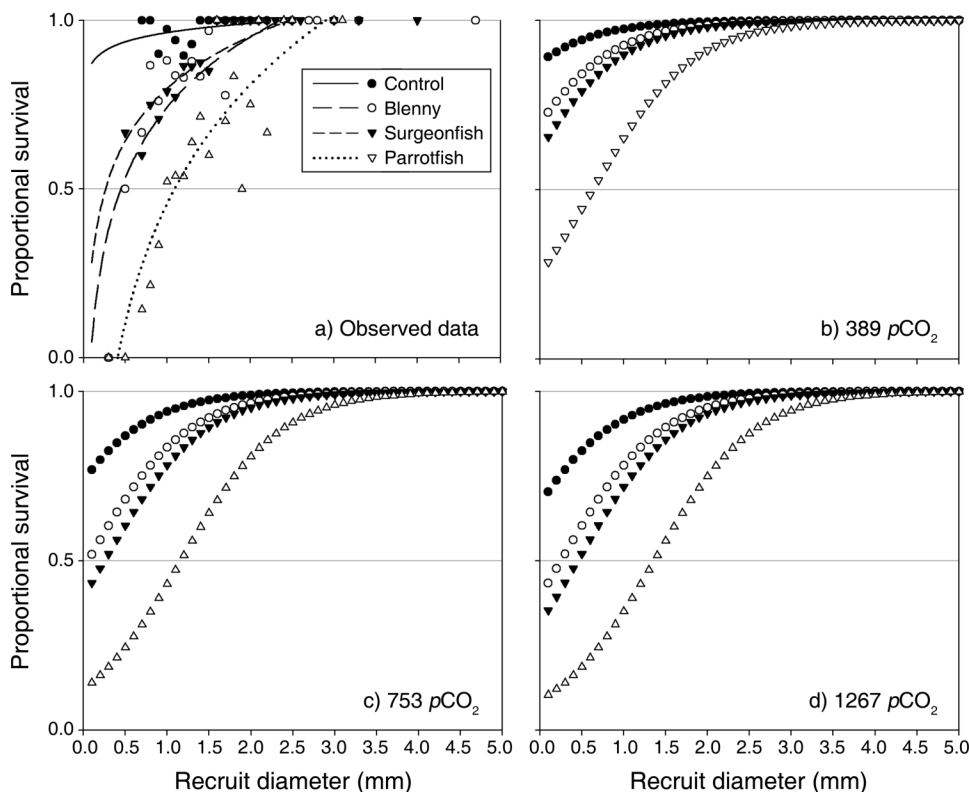


FIG. 3. The effect of size on coral recruit survivorship following 12-h grazing trials with three herbivorous fish and a no-fish control depending on: (a) the mean observed values and model predictions of those grown at (b) ambient, (c) intermediate, and (d) high $p\text{CO}_2$.

in the ambient CO_2 treatment to 27% mortality in the highest CO_2 treatment. Blenny grazing also caused significantly higher mortality than the control ($P = 0.020$), ranging from 8% mortality of the recruits grown in ambient seawater and 20% mortality of recruits grown at $1267 \mu\text{atm } p\text{CO}_2$.

There was a highly significant effect of recruit diameter ($P < 0.001$) on coral mortality, while the number of recruits per tile did not affect survival ($P = 0.086$). The proportion of recruits killed was inversely related to size, such that the smallest recruits suffered the highest mortality (Fig. 3a). This was consistent for each CO_2 treatment and fish type (except the no-fish control). Yet, the actual diameter necessary for net survival doubled from ~ 0.5 mm following blenny and surgeonfish grazing, to 1.0 mm following parrotfish grazing (Fig. 3a). The survivorship curves were best described by a logarithmic function that fitted significantly ($P < 0.001$) to the observed values for each fish type (R^2 for blenny = 0.72 , surgeonfish = 0.81 , parrotfish = 0.84), but there was no relationship between recruit size and mortality in the control ($P = 0.232$; $R^2 = 0.12$).

There were distinct differences between the survivorship of recruits ranging from 0.1 to 5.0 mm among the CO_2 treatments (Fig. 3b–d). At ambient levels (Fig. 3b), blenny and surgeonfish grazing did not affect net

survival, and only those smaller than 0.7 mm suffered net mortality following parrotfish grazing. When $p\text{CO}_2$ increased to $753 \mu\text{atm}$ (Fig. 3c), net survival still generally occurred on coral recruits following blenny and surgeonfish grazing, yet the size required for the net survivorship of recruits following parrotfish grazing almost doubled to >1.2 mm. At $1267 \mu\text{atm}$ (Fig. 3d), coral recruits needed to be >0.4 mm for net survival following blenny and surgeonfish grazing, and >1.4 mm to survive parrotfish grazing.

OA and grazing effects on algal turfs

Prior to grazing trials, the average turf cover on the settlement tiles was 86% after five days growth (Appendix E). This was consistent for tiles allocated among the different fish types or CO_2 treatments (Appendix F). Elevated $p\text{CO}_2$ during the algal turf growth did not affect the fish grazing rate (Appendix F), nor the turf C:N ratios (C:N average was 10.68 ± 0.44 [mean \pm SE]; $F_{2,12} = 0.267$, $P = 0.770$).

DISCUSSION

This series of experiments has demonstrated the complexities of OA effects on coral demography at multiple scales, from the physiology of individuals (i.e., skeletal growth), to ecological interactions (i.e., recruit-

herbivore dynamics). The chronic effect of OA decreased the post-settlement growth of coral recruits and subsequently increased their rate of mortality because mortality rates were higher in smaller size classes. However, OA had an additional effect on mortality, over and above that related to coral size: coral mortality was greater under elevated CO₂ for a given size. While evidence for a size escape in mortality was found, with the threshold varying among fish “predators,” all thresholds increased under elevated CO₂ levels, indicating that recruit survival may be compromised on reefs as OA continues to rise.

The first impact of OA on coral survivorship was mediated through its inhibitory affect on coral growth. Previous studies have also reported an inverse relationship between OA and coral recruit growth (Cohen et al. 2009, Albright et al. 2010, de Putron et al. 2011). Here, *A. millepora* recruits exhibited a nonlinear response in calcification to OA, similar to those of *Favia fragum* and *Porites astreoides* (de Putron et al. 2011), which only significantly declined in calcification below 2.33 Ω_{arag} (>753 μatm). Recruit diameter also responded nonlinearly, such that there was a significant reduction in linear growth below ambient Ω_{arag} (3.63), but not between the two lowered Ω_{arag} (2.3 and 1.56), a similar response to that found in recruits of *Acropora palmata* (Albright et al. 2010). Both of these types of reduced post-settlement growth of the recruits caused by OA decreased their survival following fish grazing.

Even at such a small size (<5 mm), the diameter of each recruit was highly important to their survival. That is, the mortality of the coral recruits was inversely related to size such that smaller individuals suffered higher mortality within each $p\text{CO}_2$, following principles of size-escape theory (e.g., Paine 1976, Gosselin and Qian 1997). In addition to prolonging the period during which recruits are vulnerable to grazer-induced mortality, OA appeared to have an additional deleterious impact on mortality. The most likely cause of this OA influence is a change in skeletal density brought about by reduced calcification, even in corals of identical size. Conceivably, corals with a weaker skeleton from elevated CO₂ may incur greater damage during fish feeding activities than those with denser skeletons. Such density-based phenomena are seen in adult corals where those species with denser skeletons lose less skeletal material when fed upon by parrotfish (Bruggemann et al. 1994). The models illustrated that rising OA incrementally increased the critical size-escape thresholds for net survival of the two month old coral recruits, resulting from their lowered calcification.

In our study, coral recruit survivorship was affected by the type of herbivore that grazed on the surrounding algal turfs. To survive grazing by parrotfish, recruits had to be double the size they needed to be to survive blenny and surgeonfish grazing because of the different feeding methods of the fish. Parrotfish scrape the substrate and remove everything growing on the surface (Bellwood

and Choat 1990), whereas blennies “brush” algal turfs with their comb-like dentition (Wilson et al. 2003) and grazing surgeonfish rapidly nip at algal turfs, tearing filaments off with a sideward flick of the head (Purcell and Bellwood 1993). Therefore, the rates of post-settlement mortality of benthic invertebrate recruits are likely to increase under future levels of elevated CO₂ because of interactions with herbivorous fish, even at very small size classes following settlement. Future work should investigate the wider impact of herbivores on recruit survivorship throughout coral ontogeny, given that positive associations between recruit density and grazing have been widely reported (Birkeland 1977, Mumby 2009) and cryptic settlement may allow recruits to escape incidental mortality by herbivory (Raimondi and Morse 2000).

There are two limitations of this study that warrant further work. In accordance with other studies in aquatic systems (Ledger and Hildrew 2005, Witt et al. 2011), we did not observe any obvious effects of OA on turf cover, palatability or grazing, after five days algal growth. Yet, the composition of the algal communities in these systems can be altered by reduced pH (Ledger and Hildrew 2005, Hall-Spencer et al. 2008, Witt et al. 2011), as can the algae-herbivore dynamics of specialized, but not generalist grazers (Ledger and Hildrew 2005). Future experiments should exclusively test these relationships over longer periods of time in marine ecosystems. Secondly, although these results and other recent work have demonstrated that elevated $p\text{CO}_2$ alters trophic (e.g., Munday et al. 2010, Ferrari et al. 2011) and non-trophic (e.g., Diaz-Pulido et al. 2011, Doropoulos et al. 2012) interactions, caution must be applied to the predictive value of these studies, as adaptation over multiple generations may serve an important role in the evolution of species and their interactions on reefs as they adapt to progressive OA.

Ocean acidification is a chronic press disturbance on calcifying taxa (Anthony et al. 2011) and our results suggest that it has the potential to reduce coral recruitment through at least two mechanisms. The outcome of such processes might generate new recruitment bottlenecks or intensify existing ones (Mumby et al. 2007). Bottlenecks in recruitment may be compounded by pre-settlement reductions to invertebrate fertilization and metamorphosis, caused by elevated CO₂ (Kurihara 2008, Byrne 2011). This combination of effects suggests that the recovery of calcifying invertebrates could be constrained by multiple processes as atmospheric CO₂ continues to rise.

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LITERATURE CITED

- Abramoff, M. D., P. J. Magelhaes, and S. J. Ram. 2004. Image processing with ImageJ. *Biophotonics International* 11:36–42.
- Albright, R., B. Mason, M. Miller, and C. Langdon. 2010. Ocean acidification compromises recruitment success of the threatened Caribbean coral *Acropora palmata*. *Proceedings of the National Academy of Sciences USA* 107:20400–20404.
- Anderson, M. J., R. N. Gorley, and K. R. Clarke. 2008. PERMANOVA+ for PRIMER: guide to software and statistical methods. PRIMER-E, Plymouth, UK.
- Anthony, K. R. N., J. A. Maynard, G. Diaz-Pulido, P. J. Mumby, P. A. Marshall, L. Cao, and O. V. E. Hoegh-Guldberg. 2011. Ocean acidification and warming will lower coral reef resilience. *Global Change Biology* 17:1798–1808.
- Babcock, R. C. 1991. Comparative demography of three species of scleractinian corals using age-dependent and size-dependent classifications. *Ecological Monographs* 61:225–244.
- Bellwood, D. R., and J. H. Choat. 1990. A functional-analysis of grazing parrotfishes (Family Scaridae)—the ecological implications. *Environmental Biology of Fishes* 28:189–214.
- Birkeland, C. 1977. The importance of rate of biomass accumulation in early successional stages of benthic communities to the survival of coral recruits. Pages 15–21 in *Proceedings of the Third International Coral Reef Symposium*. Rosenstiel School of Marine and Atmospheric Science, University of Miami, Miami, Florida, USA.
- Box, S. J., and P. J. Mumby. 2007. Effect of macroalgal competition on growth and survival of juvenile Caribbean corals. *Marine Ecology Progress Series* 342:139–149.
- Brock, R. E. 1979. Experimental study on the effects of grazing by parrotfishes and role of refuges in benthic community structure. *Marine Biology* 51:381–388.
- Bruggemann, J. H., M. J. H. Vanoppen, and A. M. Breeman. 1994. Foraging by the stoplight parrotfish *Sparisoma viride*. 1. Food selection in different socially determined habitats. *Marine Ecology Progress Series* 106:41–55.
- Byrne, M. 2011. Impact of ocean warming and ocean acidification on marine invertebrate life history stages: vulnerabilities and potential for persistence in a changing ocean. *Oceanography and Marine Biology* 49:1–42.
- Byrne, M., M. Ho, E. Wong, N. A. Soars, P. Selvakumaraswamy, H. Shepard-Brennan, S. A. Dworjanyan, and A. R. Davis. 2011. Unshelled abalone and corrupted urchins: development of marine calcifiers in a changing ocean. *Proceedings of the Royal Society B* 278:2376–2383.
- Christiansen, N. A., S. Ward, S. Harii, and I. R. Tibbetts. 2009. Grazing by a small fish affects the early stages of a post-settlement stony coral. *Coral Reefs* 28:47–51.
- Clarke, K. R., and R. N. Gorley. 2006. PRIMER v6. Plymouth Marine Laboratory, Plymouth, UK.
- Cohen, A. L., D. C. McCorkle, S. de Putron, G. A. Gaetani, and K. A. Rose. 2009. Morphological and compositional changes in the skeletons of new coral recruits reared in acidified seawater: insights into the biomineralization response to ocean acidification. *Geochemistry Geophysics Geosystems* 10:1–12.
- Connell, J. H. 1985. The consequences of variation in initial settlement vs postsettlement mortality in rocky intertidal communities. *Journal of Experimental Marine Biology and Ecology* 93:11–45.
- Connell, J. H., and M. J. Keough. 1985. Disturbance and patch dynamics of subtidal marine animals on hard substrata. Pages 125–152 in S. T. A. Pickett and P. S. White, editors. *The ecology of natural disturbance and patch dynamics*. Academic Press, Orlando, Florida, USA.
- de Putron, S. J., D. C. McCorkle, A. L. Cohen, and A. B. Dillon. 2011. The impact of seawater saturation state and bicarbonate ion concentration on calcification by new recruits of two Atlantic corals. *Coral Reefs* 30:321–328.
- Diaz-Pulido, G., M. Gouezo, B. Tilbrook, S. Dove, and K. R. N. Anthony. 2011. High CO₂ enhances the competitive strength of seaweeds over corals. *Ecology Letters* 14:156–162.
- Doropoulos, C., S. Ward, G. Diaz-Pulido, O. Hoegh-Guldberg, and P. J. Mumby. 2012. Ocean acidification reduces coral recruitment by disrupting intimate larval-algal settlement interactions. *Ecology Letters* 15:338–346.
- Fabricius, K. E., C. Langdon, S. Uthicke, C. Humphrey, S. Noonan, G. De'ath, R. Okazaki, N. Muehllehner, M. S. Glas, and J. M. Lough. 2011. Losers and winners in coral reefs acclimatized to elevated carbon dioxide concentrations. *Nature Climate Change* 1:165–169.
- Ferrari, M. C. O., M. I. McCormick, P. L. Munday, M. G. Meekan, D. L. Dixon, O. Lonnstedt, and D. P. Chivers. 2011. Putting prey and predator into the CO₂ equation—qualitative and quantitative effects of ocean acidification on predator–prey interactions. *Ecology Letters* 14:1143–1148.
- Gattuso, J. P., G. Kunsham, K. Lee, B. Rost, and K. G. Schulz. 2010. Approaches and tools to manipulate the carbonate chemistry. Pages 41–52 in U. Riebesell, V. J. Fabry, L. Hansson, and J. P. Gattuso, editors. *Guide to best practices for ocean acidification research and data reporting*. Publications Office of the European Union, Luxembourg.
- Gaylord, B., T. M. Hill, E. Sanford, E. A. Lenz, L. A. Jacobs, K. N. Sato, A. D. Russell, and A. Hettinger. 2011. Functional impacts of ocean acidification in an ecologically critical foundation species. *Journal of Experimental Biology* 214:2586–2594.
- Gosselin, L. A., and P. Y. Qian. 1997. Juvenile mortality in benthic marine invertebrates. *Marine Ecology Progress Series* 146:265–282.
- Hall-Spencer, J. M., R. Rodolfo-Metalpa, S. Martin, E. Ransome, M. Fine, S. M. Turner, S. J. Rowley, D. Tedesco, and M. C. Buia. 2008. Volcanic carbon dioxide vents show ecosystem effects of ocean acidification. *Nature* 454:96–99.
- Hughes, T. P., and J. B. C. Jackson. 1985. Population dynamics and life histories of foliaceous corals. *Ecological Monographs* 55:141–166.
- Kohler, K. E., and S. M. Gill. 2006. Coral Point Count with Excel extensions (CPCe): a visual basic program for the determination of coral and substrate coverage using random point count methodology. *Computers and Geosciences* 32:1259–1269.
- Kroeker, K. J., R. L. Kordas, R. N. Crim, and G. G. Singh. 2010. Meta-analysis reveals negative yet variable effects of ocean acidification on marine organisms. *Ecology Letters* 13:1419–1434.
- Kurihara, H. 2008. Effects of CO₂-driven ocean acidification on the early developmental stages of invertebrates. *Marine Ecology Progress Series* 373:275–284.
- Ledger, M. E., and A. G. Hildrew. 2005. The ecology of acidification and recovery: changes in herbivore-algal food web linkages across a stream pH gradient. *Environmental Pollution* 137:103–118.
- Lewis, P. D. E., and D. W. R. Wallace. 2006. MS Excel program developed for CO₂ system calculations. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, USA.
- Mumby, P. 2009. Herbivory versus corallivory: are parrotfish good or bad for Caribbean coral reefs? *Coral Reefs* 28:683–690.
- Mumby, P. J., A. Hastings, and H. J. Edwards. 2007. Thresholds and the resilience of Caribbean coral reefs. *Nature* 450:98–101.

- Munday, P. L., D. L. Dixon, M. I. McCormick, M. Meekan, M. C. O. Ferrari, and D. P. Chivers. 2010. Replenishment of fish populations is threatened by ocean acidification. *Proceedings of the National Academy of Sciences USA* 107:12930–12934.
- Navarrete, S. A. 1996. Variable predation: effects of whelks on a mid-intertidal successional community. *Ecological Monographs* 66:301–321.
- Paine, R. T. 1976. Size-limited predation: observational and experimental approach with *Mytilus–Pisaster* interaction. *Ecology* 57:858–873.
- Parker, L. M., P. M. Ross, and W. A. O'Connor. 2009. The effect of ocean acidification and temperature on the fertilization and embryonic development of the Sydney rock oyster *Saccostrea glomerata* (Gould 1850). *Global Change Biology* 15:2123–2136.
- Purcell, S. W., and D. R. Bellwood. 1993. A functional analysis of food procurement in 2 surgeonfish species, *Acanthurus nigrofuscus* and *Ctenochaetus striatus* (Acanthuridae). *Environmental Biology of Fishes* 37:139–159.
- R Development Core Team. 2010. R version 2.12.0. R Project for Statistical Computing. Vienna, Austria. www.r-project.com.
- Raimondi, P. T., and A. N. C. Morse. 2000. The consequences of complex larval behavior in a coral. *Ecology* 81:3193–3211.
- Raymundo, L. J., and A. P. Maypa. 2004. Getting bigger faster: Mediation of size-specific mortality via fusion in juvenile coral transplants. *Ecological Applications* 14:281–295.
- Rice, J. A., L. B. Crowder, and K. A. Rose. 1993. Interactions between size-structured predator and prey populations: experimental test and model comparison. *Transactions of the American Fisheries Society* 122:481–491.
- Ries, J. B., A. L. Cohen, and D. C. McCorkle. 2009. Marine calcifiers exhibit mixed responses to CO₂-induced ocean acidification. *Geology* 37:1131–1134.
- Rotjan, R. D., and S. M. Lewis. 2008. Impact of coral predators on tropical reefs. *Marine Ecology Progress Series* 367:73–91.
- Steneck, R. S. 1988. Herbivory on coral reefs: a synthesis. Pages 37–49 in J. H. Choat, et al., editors. *Proceedings of the Sixth International Coral Reef Symposium*. Executive Committee, Townsville, Australia.
- Wilson, S. K., D. R. Bellwood, J. H. Choat, and M. J. Furnas. 2003. Detritus in the epilithic algal matrix and its use by coral reef fishes. *Oceanography and Marine Biology* 41:279–309.
- Witt, V., C. Wild, K. R. N. Anthony, G. Diaz-Pulido, and S. Uthicke. 2011. Effects of ocean acidification on microbial community composition of, and oxygen fluxes through, biofilms from the Great Barrier Reef. *Environmental Microbiology* 13:2976–2989.
- Wootton, J. T. 1993. Size-dependent competition: effects on the dynamics vs. the end point of mussel bed succession. *Ecology* 74:195–206.

SUPPLEMENTAL MATERIAL

Appendix A

Summary of the physical and chemical seawater values for the CO₂ treatment levels (*Ecological Archives* E093-202-A1).

Appendix B

Ranges of *Acropora millepora* recruit sizes, number of polyps, number per tile, total numbers, and the tile replication, after two months growth at ambient and elevated *p*CO₂ that were used in each grazing trial (*Ecological Archives* E093-202-A2).

Appendix C

Total number of *Acropora millepora* recruits in each size class used for the grazing trials (*Ecological Archives* E093-202-A3).

Appendix D

ANOVA results comparing differences between the linear extension and calcification of *Acropora millepora* recruits grown at ambient and elevated CO₂ partial pressure (*p*CO₂) (*Ecological Archives* E093-202-A4).

Appendix E

Mean turf algae cover on coral settlement tiles before and after 12-h grazing trials with no-fish controls, blennies, surgeonfish, or parrotfish (*Ecological Archives* E093-202-A5).

Appendix F

ANOVA results comparing differences between the percent turf cover before and after the 12-h grazing trials with a no-fish control, blennies, surgeonfish, or parrotfish (*Ecological Archives* E093-202-A6).