Title: Coral calcification mechanisms facilitate adaptive responses to ocean acidification

Running head: Coral resistance to ocean acidification

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Supplementary Materials

Supplementary Methods

Experimental protocol

Carbon dioxide gas and CO_2-free air were mixed using a custom system to achieve the desired pH_T levels, whereas temperature was controlled using heaters and chillers (±0.5°C). Lighting was provided via metal halide lights on a 12 hour light cycle with maximum irradiances of 400-500 μmol photons m^{-2} s^{-1}, resulting in a daily integral light flux of 8-10 mol photons m^{-2} d^{-1}. Aquarium pumps provided water motion and mixing in each tank, resulting in flow speeds of ~5-15 cm s^{-1} near the corals. Corals were fed weekly using ~1 g dry weight of newly hatched *Artemia* nauplii or Reef-Roids coral food [1]. At the end of the experiment, corals were frozen at -80°C and subsequently stored at -20°C until processing for geochemical analyses.

Environmental monitoring

Temperature was measured in each aquarium about 5-6 days per week at various times of day using a thermometer accurate to ±0.05°C. Salinity was measured 1-2 times per week, while pH was measured in the morning and evening on a given day, typically 3 days per week. Salinity was measured with a YSI conductivity meter, and pH was determined spectrophotometrically using m-cresol purple [2]. Total alkalinity (TA) was measured ~weekly using a modified Gran titration, and the accuracy of titrations was verified with Certified Reference Materials obtained from Andrew Dickson [2]. TA and salinity sampling were uneven across the experiment, so means and errors were temporally weighted. To better characterise the diel cycle of chemistry variation, pH and temperature were also measured in

each aquarium every 3 hr for a 24 hr period (TA and salinity were measured every 6 hr) about midway through the experiment. This 24 hr sampling showed that diel pH variation was quasi-sinusoidal and that the morning and evening pH sampling scheme described above provided robust estimates of daily minimum, mean, and maximum pH. The daily pH range averaged 0.22±0.01 units, with the variation about evenly distributed around the mean. TA tended to increase by ~10-40 μeq kg^{-1} at night in the aquariums as compared to daytime values, likely due to lower night time calcification rates by the corals. TA averaged 2212±6 μeq kg^{-1} in the incoming seawater. Median daytime values for pCO_2 and Ω_{arag} were calculated using CO2SYS [3].

Geochemical analyses

Prior to shipping the corals from HIMB to the University of Western Australia (UWA), corals were soaked in a 1:1 solution of household bleach and deionized (DI) water for 48 hours to remove the tissue and organics within the skeleton. Skeletons were then rinsed extensively with DI water and air-dried. At UWA, the rinsing process was repeated using a sonic bath filled with DI water. The uppermost layer of the dried branch tips was then gently shaved with a diamond-tipped Dremel tool [4, 5]. Only corals that met all of the following strict criteria were sampled: (1) no partial mortality, infestation by the *Porites*-eating nudibranch *Phestilla* spp. or visible signs of bleaching occurred at any point during the experiment, (2) the stain line was clearly visible, (3) new growth beyond the stain line occurred primarily along the branch tips, as boron isotopes can differ between branch tips and other locations [6], and (4) net growth during the second temperature phase was substantially more than the material needed for boron isotope and trace element analyses (i.e., 10 mg). The only treatment that did not fulfil all four criteria was the 25.3°C/pH 7.71 treatment for *P. compressa* corals from Waimānalo Bay, which did not have net positive growth during either

the first or second temperature phase of the experiment. However, they showed extensive apical growth beyond the stain line (5-10 mm), suggesting that growth along the tips occurred throughout the experiment, while other parts of the coral experienced dissolution. These corals were therefore nevertheless sampled for geochemical analyses.

Boron isotopes and trace elements were analysed following the method of McCulloch et al. [7]. Sample weighing, chemical dissolution and boron extraction procedures were undertaken in the metal-free hepa-filtered (ISO 7) clean room complex at the Advanced Geochemical Facility for Indian Ocean Research (AGFIOR) at UWA. To ensure complete removal of organic matter, powders were cleaned with 6.25% NaClO as described previously [5]. This cleaning procedure does not influence boron isotopes or trace elements [8]. After dissolution in 0.51 N HNO_3, an aliquot of the resulting solution diluted to 100 ppm Ca was used for analysis of B/Ca. The remaining undiluted solution was used for boron extraction using combined cation-anion exchange columns. The extracted boron was then analysed using a NU Plasma II Multi-Collector Inductively Coupled Plasma Mass Spectrometer (MC-ICP-MS; NU Instruments) at UWA, with sample measurements bracketed by an in-house standard (~19.8‰) and blank measurements. The boron isotopic composition of the skeleton ($\delta^{11}B$) is reported as the per mil deviation of the stable isotopes ^{11}B:^{10}B relative to NIST SRM-951 boric acid standard. All samples were analysed in duplicate. The overall precision of this method is ~ ±0.3‰ (2sd). B/Ca was analysed on an X-Series 2 Quadrupole Inductively Coupled Plasma Mass Spectrometer (Q-ICPMS; Thermo Fisher Scientific) at UWA using the standard Xt interface and the plasma screen fitted.

Carbonate chemistry of the calcifying fluid

Boron exists in seawater as two different species, boric acid ($B(OH)_3$) and borate ($B(OH)_4^-$). Importantly, the two species have a distinct isotopic composition and their

relative abundance in seawater is pH-dependent. Since only borate is thought to be incorporated into coral aragonite [9, 10], coral skeletal $\delta^{11}B$ ($\delta^{11}B_{carb}$) reflects the internal pH inside the calcifying fluid (pH_{cf}) rather than seawater pH [11, 12]. pH_{cf} was calculated using the following equation [13]:

$$pH_{cf} = pK_B - \log\left[\frac{(\delta^{11}B_{sw} - \delta^{11}B_{carb})}{\left(\alpha_{(B3-B4)}\,\delta^{11}B_{carb} - \delta^{11}B_{sw} + 1000\left(\alpha_{(B3-B4)} - 1\right)\right)}\right] \quad (1)$$

where pK_B is the dissociation constant of boric acid, adjusted to the respective treatment seawater salinity and temperature [14], $\delta^{11}B_{sw}$ is the boron isotopic value of seawater, with a value of 39.61‰ [15], and $\alpha_{(B3-B4)}$ is the fractionation factor, with a value of 1.0272 [16]. It was assumed that $\delta^{11}B$ of the calcifying fluid has the same isotopic composition as seawater, since seawater is the ultimate source of boron at the site of calcification.

Recently, it has also been shown that the coral skeletal B/Ca concentration can be used to constrain the dissolved inorganic carbon (DIC) concentration of the coral calcifying fluid (DIC_{cf}) [17, 18]. This is because the partitioning of borate versus carbonate into aragonite appears to be sensitive to pH_{cf} [12, 17, 19]. We therefore estimated the carbonate ion concentration within the calcifying fluid ($[CO_3^{2-}]_{cf}$) from measurements of both pH_{cf} and coral skeletal B/Ca using the following equation [18]:

$$\left[CO_3^{2-}\right]_{cf} = K_D \times \left[B(OH)_4^-\right]_{cf} \Big/ \left(B/Ca\right)_{CaCO_3} \quad (2)$$

where

$$K_D = 0.00297 \exp(-0.0202 [H^+]_{cf}) \quad (3)$$

Finally, the DIC_{cf} and the aragonite saturation state within the calcifying fluid (Ω_{cf}) were calculated from the estimated values for pH_{cf} and $[CO_3^{2-}]_{cf}$, and the percent fraction of DIC in the calcifying fluid that is present as carbonate. Biological DIC-upregulation within the calcifying fluid was calculated as the ratio of DIC_{cf} and seawater DIC (DIC_{cf}/DIC_{sw}).

For typical calcifying fluid pH_{cf} values of ~8.3 to~ 8.4, K_D ~0.0027, an order of magnitude higher than the previous estimate of Allison et al. [19]. This difference in K_D values is important because our calculated K_D, based on the experimental determination of Holcomb et al. [17], is now compatible with direct substitution of CO_3^{2-} ions with $B(OH)_4^-$ ions during the formation of aragonite, obviating the need for ad hoc scenarios of HCO_3^{2-} substitution [19]. We also assumed that the total boron and calcium concentrations of the calcifying fluid are equal to their concentrations in seawater and solely a function of seawater salinity.

Supplementary Tables

Table S1. Sample sizes per treatment for each response variable. cf = calcifying fluid, ΔpH = biological pH-upregulation, DIC = dissolved inorganic carbon, sw = seawater, [carb] = carbonate ion concentration, Ω = aragonite saturation state, calc. geochem corals = calcification rates of all corals used for geochemical analyses, calc. all corals = calcification rate of all corals in the experiment.

Site	**Kāneohe Bay**						**Waimānalo Bay**					
Temp.	**25.3°C**			**26.8°C**			**25.3°C**			**26.8°C**		
pH_T	**8.04**	**7.88**	**7.71**	**8.04**	**7.88**	**7.71**	**8.04**	**7.88**	**7.71**	**8.04**	**7.88**	**7.71**
Montipora capitata												
$\delta^{11}B$	9	8	6	5	5	6	5	6	5	4	7	6
pH_{cf}	9	8	6	5	5	6	5	6	5	4	7	6
ΔpH	9	8	6	5	5	6	5	6	5	4	7	6
B/Ca	9	8	6	5	5	6	5	6	5	3	6	6
DIC_{cf}	9	8	6	5	5	6	5	6	5	3	6	6
DIC_{cf}/DIC_{sw}	9	8	6	5	5	6	5	6	5	3	6	6
$[carb]_{cf}$	9	8	6	5	5	6	5	6	5	3	6	6
Ω_{cf}	9	8	6	5	5	6	5	6	5	3	6	6
Calc. geochem. corals	9	8	6	5	5	6	5	6	5	4	7	6
Calc. all corals	11	11	12	11	11	11	10	12	12	7	10	7
Porites compressa												
$\delta^{11}B$	5	5	6	4	6	5	3	3	2	3	4	3
pH_{cf}	5	5	6	4	6	5	3	3	2	3	4	3
ΔpH	5	5	6	4	6	5	3	3	2	3	4	3
B/Ca	5	5	6	4	6	5	4	3	2	3	4	3
DIC_{cf}	5	5	6	4	6	5	4	3	2	3	4	3
DIC_{cf}/DIC_{sw}	5	5	6	4	6	5	4	3	2	3	4	3
$[carb]_{cf}$	5	5	6	4	6	5	4	3	2	3	4	3
Ω_{cf}	5	5	6	4	6	5	4	3	2	3	4	3
Calc. geochem. corals	5	5	6	4	6	5	3	3	2	3	4	3
Calc. all corals	11	12	12	12	11	5	11	11	11	10	11	4

Table S2. Environmental conditions during the second, 9-week temperature phase of the experiment. Treatment (treatm.), salinity (sal; psu), temperature (temp; °C), pH_T, total alkalinity (TA; µeq kg^{-1}), carbonate ion concentration (CO_3^{2-}; µmol kg^{-1}), dissolved inorganic carbon concentration (DIC; µmol kg^{-1}) and aragonite saturation state (Ω_{arag}). Measured values reported as overall mean values ±SEM. Sample size (n) as indicated.

Treatm.	**Tank**	**Sal.** (13)	**Temp.** (38)	**pH_T** (14)	**TA** (8-9)	**CO_3^{2-}**	**DIC**	**Ω_{arag}**
pH 8.04,	4	34.6±0.2	25.3±0.0	8.04±0.01	2158±6	198	1871	3.15
25.3°C	13	34.6±0.3	25.2±0.0	8.02±0.01	2146±6	189	1872	3.01
pH 7.88,	6	34.7±0.2	25.3±0.1	7.90±0.01	2171±6	154	1956	2.45
25.3°C	14	34.6±0.3	25.2±0.0	7.89±0.01	2131±10	148	1924	2.35
pH 7.71,	18	34.6±0.3	25.1±0.1	7.76±0.01	2152±9	115	2002	1.84
25.3°C	24	34.6±0.2	25.2±0.0	7.74±0.01	2181±4	113	2037	1.79
pH 8.04,	1	34.6±0.2	26.7±0.0	8.02±0.02	2153±10	198	1866	3.17
26.8°C	10	34.7±0.2	26.8±0.1	8.04±0.01	2164±14	207	1863	3.32
pH 7.88,	5	34.7±0.2	26.7±0.0	7.89±0.01	2154±10	156	1935	2.51
26.8°C	16	34.7±0.2	26.8±0.1	7.87±0.01	2125±9	149	1917	2.39
pH 7.71,	15	34.6±0.2	26.9±0.0	7.70±0.01	2211±4	111	2072	1.79
26.8°C	23	34.6±0.2	26.7±0.1	7.72±0.01	2164±4	113	2020	1.81

Table S3. Results from generalized linear mixed models to test for the effects of species (sp.), site, pH and temperature (temp.) on boron isotopes ($\delta^{11}B$), calcifying fluid pH (pH_{cf}) and biological pH-upregulation (ΔpH) of *Montipora capitata* (MC) and *Porites compressa* (PC). Post hoc Tukey tests results are given when main effects (but no interaction terms) were significant. Effects with *p*-values ≤0.05 are highlighted in bold. Num df = numerator degrees of freedom, den df = denominator degrees of freedom. Var. = variable, KB = Kāneʻohe Bay, Wai = Waimānalo Bay.

Var.	Effect	Num df	Den df	*F*-statistic	*p*-value	Tukey
$\delta^{11}B$	**sp**	1	36	24.33	**<.0001**	MC>PC
	site	1	36	19.52	**<.0001**	KB>Wai
	sp*site	1	36	0.87	0.3576	
	pH	2	62	22.64	**<.0001**	8.04>7.88>7.71
	sp*pH	2	62	0.19	0.8265	
	site*pH	2	62	2.28	0.1107	
	sp*site*pH	2	62	0.52	0.5959	
	temp	1	62	2.64	0.1090	
	sp*temp	1	62	0.66	0.4206	
	site*temp	1	62	0.00	0.9526	
	sp*site*temp	1	62	0.76	0.3858	
	pH*temp	2	62	0.55	0.5793	
	sp*pH*temp	2	62	1.61	0.2077	
	site*pH*temp	2	62	0.49	0.6148	
	sp*site*pH*temp	2	62	0.64	0.5285	
pH_{cf}	**sp**	1	36	22.94	**<.0001**	MC>PC
	site	1	36	19.96	**<.0001**	KB>Wai
	sp*site	1	36	0.76	0.3880	
	pH	2	62	21.06	**<.0001**	8.04>7.88>7.71
	sp*pH	2	62	0.21	0.8139	
	site*pH	2	62	2.02	0.1418	
	sp*site*pH	2	62	0.53	0.5907	
	temp	1	62	0.03	0.8524	
	sp*temp	1	62	0.53	0.4685	
	site*temp	1	62	0.02	0.8830	
	sp*site*temp	1	62	0.8	0.3760	
	pH*temp	2	62	0.69	0.5030	
	sp*pH*temp	2	62	1.55	0.2203	
	site*pH*temp	2	62	0.46	0.6308	
	sp*site*pH*temp	2	62	0.65	0.5253	
ΔpH	**sp**	1	36	23.83	**<.0001**	MC>PC
	site	1	36	20.46	**<.0001**	KB>Wai
	sp*site	1	36	0.79	0.3786	

pH	2	62	161.91	**<.0001**	7.71>7.88>8.04
sp*pH	2	62	0.46	0.6332	
site*pH	2	62	1.84	0.1672	
sp*site*pH	2	62	0.54	0.5873	
temp	1	62	2.54	0.1158	
sp*temp	1	62	0.56	0.4578	
site*temp	1	62	0.14	0.7050	
sp*site*temp	1	62	0.75	0.3905	
pH*temp	2	62	0.59	0.5575	
sp*pH*temp	2	62	1.65	0.1999	
site*pH*temp	2	62	0.46	0.6312	
sp*site*pH*temp	2	62	0.65	0.5259	

Table S4. Results from generalized linear mixed models to test for the effects of species (sp.), site, pH and temperature (temp.) on B/Ca ratios, estimated calcifying fluid DIC (DIC_{cf}) and biological DIC-upregulation (DIC_{cf}/DIC_{sw}) of *Montipora capitata* (MC) and *Porites compressa* (PC). Post hoc Tukey tests results are given when main effects (but no interaction terms) were significant. Effects with *p*-values ≤0.05 are highlighted in bold. Num df = numerator degrees of freedom, den df = denominator degrees of freedom. Var. = variable, KB = Kāneʻohe Bay, Wai = Waimānalo Bay.

Var.	Effect	Num df	Den df	*F*-statistic	*p*-value	Tukey
B/Ca	**sp**	1	35	431.51	**<.0001**	MC>PC
	site	1	35	39.98	**<.0001**	KB>Wai
	sp*site	1	35	1.69	0.2015	
	pH	2	61	11.29	**<.0001**	8.04>7.88=7.71
	sp*pH	2	61	0.86	0.4303	
	site*pH	2	61	0.67	0.5139	
	sp*site*pH	2	61	2.89	0.0633	
	temp	1	61	0.30	0.5834	
	sp*temp	1	61	1.26	0.2658	
	site*temp	1	61	0.09	0.7653	
	sp*site*temp	1	61	0.02	0.8772	
	pH*temp	2	61	0.92	0.4058	
	sp*pH*temp	2	61	0.17	0.8416	
	site*pH*temp	2	61	0.97	0.3846	
	sp*site*pH*temp	2	61	0.64	0.5282	
DIC_{cf}	**sp**	1	35	476.87	**<.0001**	
	site	1	35	61.62	**<.0001**	
	sp*site	1	35	9.79	**0.0035**	See text
	pH	2	61	31.1	**<.0001**	
	sp*pH	2	61	4.72	**0.0124**	See text
	site*pH	2	61	0.41	0.6634	
	sp*site*pH	2	61	2.40	0.0996	
	temp	1	61	6.86	**0.0111**	25.3>26.8
	sp*temp	1	61	1.38	0.2448	
	site*temp	1	61	0.53	0.4706	
	sp*site*temp	1	61	0.28	0.6008	
	pH*temp	2	61	0.88	0.4193	
	sp*pH*temp	2	61	0.02	0.9800	
	site*pH*temp	2	61	1.44	0.2455	
	sp*site*pH*temp	2	61	1.21	0.3055	
DIC_{cf}/ DIC_{sw}	**sp**	1	35	482.35	**<.0001**	
	site	1	35	56.8	**<.0001**	
	sp*site	1	35	8.10	**0.0074**	See text

pH	2	61	2.67	0.0774	
sp*pH	2	61	4.05	**0.0224**	See text
site*pH	2	61	1.20	0.3071	
sp*site*pH	2	61	1.50	0.2302	
temp	1	61	8.11	**0.0060**	25.3>26.8
sp*temp	1	61	0.36	0.5516	
site*temp	1	61	0.12	0.7348	
sp*site*temp	1	61	0.03	0.8705	
pH*temp	2	61	0.05	0.9514	
sp*pH*temp	2	61	0.33	0.7218	
site*pH*temp	2	61	0.84	0.4345	
sp*site*pH*temp	2	61	0.79	0.4575	

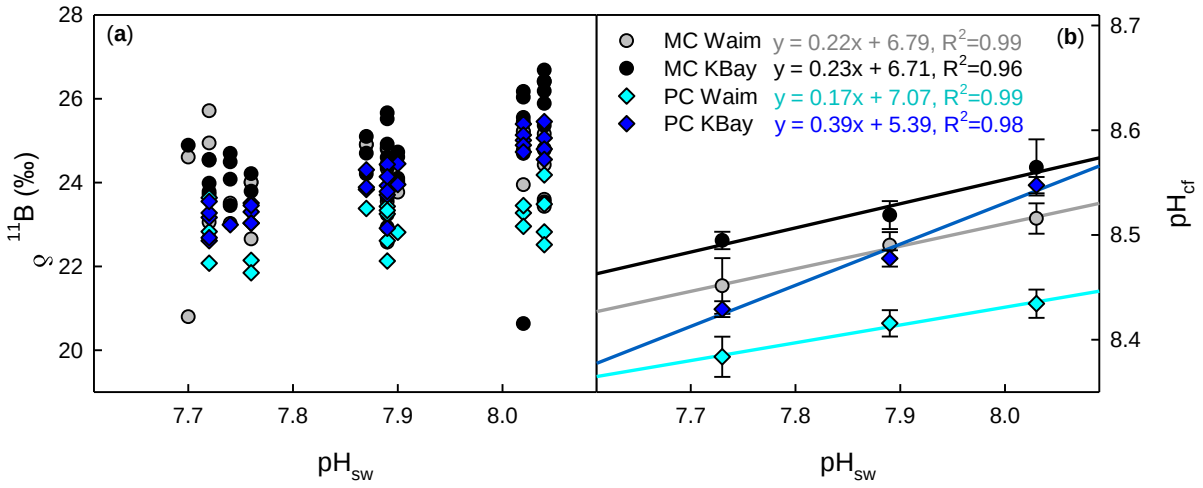
Table S5. Results from generalized linear mixed models to test for the effects of species (sp.), site, pH and temperature (temp.) on estimated calcifying fluid carbonate ion concentration ($carb_{cf}$), aragonite saturation state (Ω_{cf}) and calcification rate (calc.) of *Montipora capitata* (MC) and *Porites compressa* (PC). Post hoc Tukey tests results are given when main effects (but no interaction terms) were significant. Effects with *p*-values ≤0.05 are highlighted in bold. Num df = numerator degrees of freedom, den df = denominator degrees of freedom. Var. = variable, KB = Kāneʻohe Bay, Wai = Waimānalo Bay.

Var.	Effect	Num df	Den df	*F*-statistic	*p*-value	Tukey
$carb_{cf}$	**sp**	1	35	74.28	**<.0001**	
	site	1	35	0.37	0.5447	
	sp*site	1	35	0.01	0.9175	
	pH	2	61	9.67	**0.0002**	
	sp*pH	2	61	0.03	0.9741	
	site*pH	2	61	1.39	0.2567	
	sp*site*pH	2	61	3.24	**0.0461**	See text
	temp	1	61	1.38	0.2440	
	sp*temp	1	61	0.01	0.9207	
	site*temp	1	61	0.02	0.8817	
	sp*site*temp	1	61	0.77	0.3825	
	pH*temp	2	61	0.48	0.6234	
	sp*pH*temp	2	61	1.96	0.1491	
	site*pH*temp	2	61	0.10	0.9075	
	sp*site*pH*temp	2	61	0.12	0.8849	
Ω_{cf}	**sp**	1	35	74.38	**<.0001**	
	site	1	35	0.37	0.5462	
	sp*site	1	35	0.01	0.9111	
	pH	2	61	9.73	**0.0002**	
	sp*pH	2	61	0.03	0.9732	
	site*pH	2	61	1.39	0.2567	
	sp*site*pH	2	61	3.25	**0.0455**	See text
	temp	1	61	3.00	0.0885	
	sp*temp	1	61	0.02	0.8895	
	site*temp	1	61	0.02	0.8843	
	sp*site*temp	1	61	0.77	0.3838	
	pH*temp	2	61	0.49	0.6153	
	sp*pH*temp	2	61	1.97	0.1486	
	site*pH*temp	2	61	0.10	0.9067	
	sp*site*pH*temp	2	61	0.12	0.8866	
Calc	**sp**	1	36	19.27	**<.0001**	MC>PC
	site	1	36	9.80	**0.0034**	KB>Wai
	sp*site	1	36	0.06	0.8061	

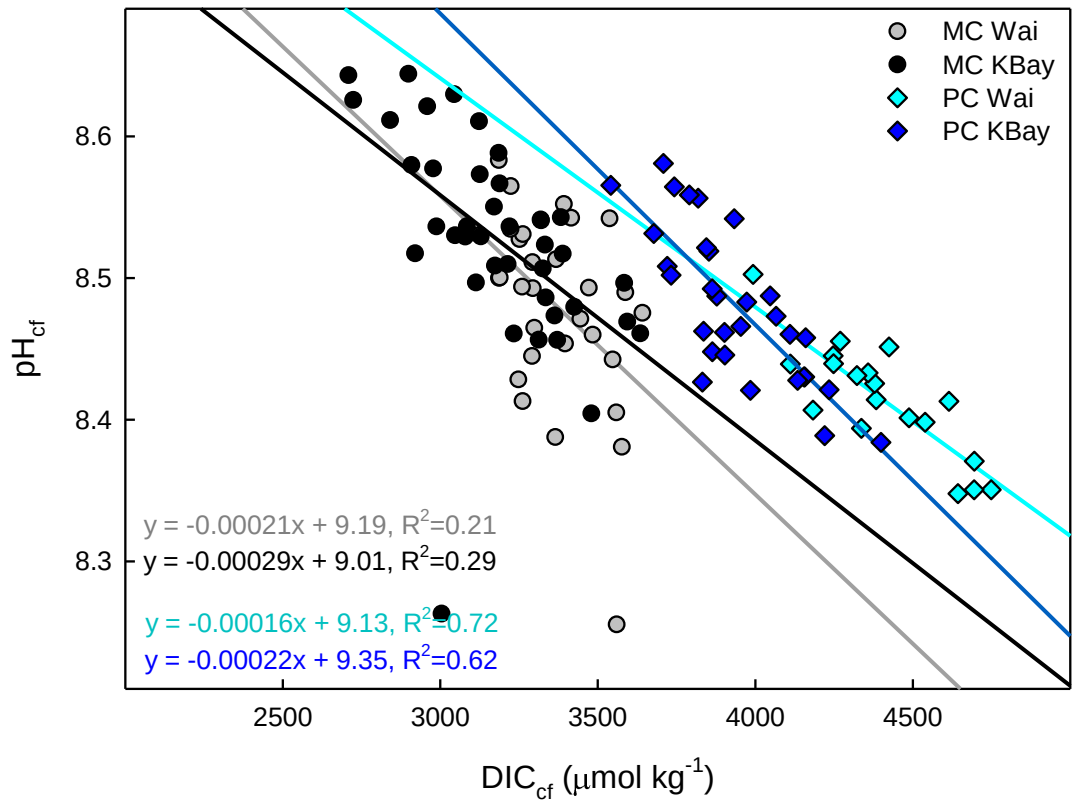
	pH	2	62	2.61	0.0819	
	sp*pH	2	62	0.68	0.5115	
	site*pH	2	62	1.35	0.2677	
	sp*site*pH	2	62	1.16	0.3189	
	temp	1	62	0.00	0.9599	
	sp*temp	1	62	0.29	0.5921	
	site*temp	1	62	0.02	0.8861	
	sp*site*temp	1	62	0.39	0.5347	
	pH*temp	2	62	0.92	0.4022	
	sp*pH*temp	2	62	0.89	0.4156	
	site*pH*temp	2	62	0.90	0.4105	
	sp*site*pH*temp	2	62	1.42	0.2488	
Calc*	**sp**	1	43	25.34	**<.0001**	MC>PC
	site	1	43	9.56	**0.0035**	KB>Wai
	sp*site	1	43	0.06	0.8043	
	pH	2	179	4.59	**0.0114**	
	sp*pH	2	179	0.42	0.6561	
	site*pH	2	179	0.16	0.8481	
	sp*site*pH	2	179	1.72	0.1826	
	temp	1	179	0.31	0.5812	
	sp*temp	1	179	3.05	0.0824	
	site*temp	1	179	0.15	0.6954	
	sp*site*temp	1	179	1.49	0.2240	
	pH*temp	2	179	4.66	**0.0106**	See text
	sp*pH*temp	2	179	0.03	0.9701	
	site*pH*temp	2	179	0.82	0.4425	
	sp*site*pH*temp	2	179	0.04	0.9618	

*Calcification rates of all corals in the experiment, not just the ones for which geochemical analyses were conducted.

Supplementary Figures



Supplementary Figure S1. Correlation between (a) skeletal boron isotopes ($\delta^{11}B$) and (b) calcifying fluid pH (pH_{cf}) and seawater pH (pH_{sw}), respectively, for *Montipora capitata* (MC) and *Porites compressa* (PC) from Kāneʻohe Bay (KBay) and Waimānalo Bay (Waim), Hawaiʻi. Results from linear regression analysis are indicated. In panel b, error bars (±1 SE) are given for pH_{cf} averages. For seawater pH, error bars were typically ±0.01 (SE) and are therefore not shown (see Table S2).



Supplementary Figure S2: Correlation between calcifying fluid pH (pH_{cf}) and dissolved inorganic carbon concentration (DIC_{cf}) in *Montipora capitata* (MC) and *Porites compressa* (PC) from Kāne'ohe Bay (KBay) and Waimānalo Bay (Wai), Hawai'i. Results from linear regression analysis are indicated. When the outliers are removed for MC Wai and MC KBay, the following two equations were obtained: $y = -0.0001x + 8.98$, $R^2 = 0.15$ (MC Wai) and $y = -0.0002x + 9.17$, $R^2 = 0.60$ (MC Kbay).

References

1. Forsman ZH, Kimokeo B, Bird CE, Hunter CL, Toonen RJ. 2012 Coral farming: species-specific effects of light, water motion and artificial foods. *Journal of the Marine Biological Association of the UK* **92**, 721-729. (doi:10.1017/S0025315411001500)
2. DOE. 1994 *Handbook of methods for the analysis of the various parameters of the carbon dioxide system in sea water, version 2*, ORNL/CDIAC-74.
3. Lewis E, Wallace D. 1998 Program developed for CO_2 system calculations. (pp. ORNL/CDIAC-105, Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy.
4. Grottoli AG, Rodrigues LJ, Juarez C. 2004 Lipids and stable carbon isotopes in two species of Hawaiian corals, *Porites compressa* and *Montipora verrucosa*, following a bleaching event. *Mar. Biol.* **145**, 621-631. (doi:10.1007/s00227-004-1337-3)
5. Schoepf V, McCulloch MT, Warner ME, Levas SJ, Matsui Y, Aschaffenburg M, Grottoli AG. 2014 Short-term coral bleaching is not recorded by skeletal boron isotopes. *PLoS One* **9**, e112011. (doi:10.1371/journal.pone.0112011)
6. Holcomb M, Venn AA, Tambutte E, Tambutte S, Allemand D, Trotter J, McCulloch M. 2014 Coral calcifying fluid pH dictates response to ocean acidification. *Scientific Reports* **4**, 5207. (doi:10.1038/srep05207)
7. McCulloch MT, Holcomb M, Rankenburg K, Trotter J. 2014 Rapid, high-precision measurements of boron isotopic compositions in marine carbonates. *Rapid Communications in Mass Spectrometry* **28**, 1-9. (doi:10.1002/rcm.7065)
8. Holcomb M, DeCarlo TM, Schoepf V, Dissard D, Tanaka K, McCulloch M. 2015 Cleaning and pre-treatment procedures for biogenic and synthetic calcium carbonate powders for determination of elemental and boron isotopic compositions. *Chem. Geol.* **398**, 11-21. (doi:10.1016/j.chemgeo.2015.01.019)
9. Hemming NG, Hanson GN. 1992 Boron isotopic composition and concentration in modern marine carbonates. *Geochim. Cosmochim. Acta* **56**, 537-543. (doi:10.1016/0016-7037(92)90151-8)
10. Mavromatis V, Montouillout V, Noireaux J, Gaillardet J, Schott J. 2014 Characterization of boron incorporation and speciation in calcite and aragonite from co-precipitation experiments under controlled pH, temperature and precipitation rate. *Geochim. Cosmochim. Acta* **150**, 299–313. (doi:10.1016/j.gca.2014.10.024)
11. McCulloch MT, Falter J, Trotter J, Montagna P. 2012 Coral resilience to ocean acidification and global warming through pH up-regulation. *Nat. Clim. Change* **2**, 623-627. (doi:10.1038/nclimate1473)
12. Trotter J, Montagna P, McCulloch MT, Silenzi S, Reynaud S, Mortimer G, Martin S, Ferrier-Pages C, Gattuso J-P, Rodolfo-Metalpa R. 2011 Quantifying the pH 'vital effect' in the temperate zooxanthellate coral *Cladocora caespitosa*: Validation of the boron seawater pH proxy. *Earth Planet. Sci. Lett.* **303**, 163-173. (doi:10.1016/j.epsl.2011.01.030)
13. Zeebe R, Wolf-Gladrow DA. 2001 *CO_2 in Seawater: Equilibrium, Kinetics, Isotopes*. Amsterdam, Elsevier.
14. Dickson AG. 1990 Thermodynamics of the dissociation of boric acid in synthetic seawater from 273.15 to 318.15K. *Deep Sea Research Part A. Oceanographic Research Papers* **37**, 755-766. (doi:10.1016/0198-0149(90)90004-F)
15. Foster GL, Pogge von Strandmann PAE, Rae JWBCQ. 2010 Boron and magnesium isotopic composition of seawater. *G-cubed* **11**, Q08015. (doi:10.1029/2010gc003201)

16. Klochko K, Kaufman AJ, Yao W, Byrne RH, Tossell JA. 2006 Experimental measurement of boron isotope fractionation in seawater. *Earth Planet. Sci. Lett.* **248**, 276-285. (doi:10.1016/j.epsl.2006.05.034)
17. Holcomb M, DeCarlo TM, Gaetani GA, McCulloch M. 2016 Factors affecting B/Ca ratios in synthetic aragonite. *Chem. Geol.* **437**, 67-76. (doi:10.1016/j.chemgeo.2016.05.007)
18. McCulloch M, D'Olivo Cordero JP, Falter J, Holcomb M, Trotter J. 2017 Coral calcification in a changing world and the interactive dynamics of pH and DIC up-regulation. *Nat. Comm.* **8: 15686**. (doi:10.1038/ncomms15686)
19. Allison N, Cohen I, Finch AA, Erez J, Tudhope AW. 2014 Corals concentrate dissolved inorganic carbon to facilitate calcification. *Nat. Comm.* **5**, 5741. (doi:10.1038/ncomms6741)