Supplementary Material

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **pH****treatment** | **pH****observed** | **TA****(μmol/L)** | ***p*CO2****(μatm)** | **CO2****(μmol/kg-SW)** | **HCO3–****(μmol/kg-SW)** | **CO32–****(μmol/kg-SW)** | ***Ω*arag** | **Temperature****(℃)** | **Salinity****(ppt)** | **PO43+-P****(μmol/L)** | **NO2–-N****(μ mol/L)** | **NO3–-N****(μmol/L)** | **SiO32–-Si****(μmol/L)** | **NH4+-N****(μmol/L)** |
| **pH 8.1** | **8.11 ± 0.01** | 2610.25 ± 45.75 a | 497.42± 9.05 a | 13.44 ± 0.24 a | 2030.49 ± 36.95 a | 241.69 ± 4.39 c | 5.84 ± 0.01 c | 27 ± 0.3a | 34 ± 0.1a | 0.03 ± 0.01 a | 0.45 ± 0.43 a | < 0.05 | 0.18± 0.18 a | 0.35± 0.35 a |
| **pH 7.7** | **7.69 ± 0.01** | 2608.00 ± 41.98 a | 1115.37± 18.30 b | 30.13 ± 0.49 b | 2282.09 ± 37.51 b | 136.14 ± 2.23 b | 3.29 ± 0.05 b | 27 ± 0.3a | 34 ± 0.1a | 0.03 ± 0.01 a | 0.49 ± 0.33 a | < 0.05 | 0.36± 0.18 a | 0.51± 0.33 a |
| **pH 7.4** | **7.39 ± 0.02** | 2657.50 ± 42.52 a | 2419.20± 39.12 c | 65.36 ± 1.05 c | 2480.71 ± 40.12 c | 74.17 ± 1.19 a | 1.79 ± 0.02 a | 27 ± 0.3a | 34 ± 0.1a | 0.05 ± 0.01 a | 0.58 ± 0.32 a | < 0.05 | 0.51 ± 0.26 a | 0.84± 0.45a |

**Table S1.** Carbonate chemistry and water quality parameters in the three experimental pH treatments. Values are expressed as the mean ± SE of six determinations (n = 6). Different superscript letters represent statistically significant differences among the pH treatments for each parameter measured (*p* < 0.05). The carbonate seawater chemistry parameters were calculated from pH, TA, temperature, and salinity using the free-access CO2SYS.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Dependent variable | SS | *df* | MS | *F* | *p* |
| TA | 6250.5 | 2 | 3125.2 | 1.655 | 0.244 |
| *p*CO2 | 7700054 | 2 | 3850027 | 5930.5 | **0.000** |
| CO2 | 5621.4 | 2 | 2810.7 | 5945.9 | **0.000** |
| HCO3– | 407275 | 2 | 203637 | 139.386 | **0.000** |
| CO32 | 57392.0 | 2 | 28696.02 | 3337.6 | **0.000** |
| *Ω*arag | 33.510 | 2 | 16.755 | 3352.868 | **0.000** |
| PO43+-P | 0.001 | 2 | 0.001 | 1.546 | 0.287 |
| NO2–-N | 0.03 | 2 | 0.015 | 0.037 | 0.964 |
| NO3–-N | 0.000 | 2 | 0.000 |  |  |
| SiO32–-Si | 0.167 | 2 | 0.083 | 0.633 | 0.563 |
| NH4+-N | 0.378 | 2 | 0.189 | 0.437 | 0.665 |

**Table S2.** One-way ANOVA comparing carbonate chemistry and water quality parameters in the three experimental pH treatments. Significant differences (*p* < 0.05) are highlighted in bold. SS: type III sum of squares; *df*: degree of freedom; MS: mean square; *F*: *F*-ratio; *p*: *p*-value.

**Table S3.** Effect of ocean acidification on photochemical efficiency, and PFC and DDN assimilation in *G. fascicularis.* Values are expressed as mean ± SE. Different superscript letters represent statistically significant differences among the pH treatments for each parameter measured (*p* < 0.05).

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Duration | pH treatment  | Δ*F*/*Fm*′ | *Fv/Fm* | Calcification rate % week–1 | PFC assimilation(nmol Ctracer µmol Ctissue–1 d–1) | DDN assimilation(nmol Ntracer µmol Ntissue–1 d–1) | δ13C (‰) | δ15N (‰) |
| 0 days | 8.1 | 0.57 ± 0.01 a | 0.66 ± 0.01 a | \_ | 6.80 ± 0.83 a | 0.17 ± 0.03 b | -27.97 ± 2.30 a | 4.59 ± 0.13 a |
|  | 7.7 | 0.55 ± 0.02 a | 0.65 ± 0.01 a | \_ | 6.79 ± 0.82 a | 0.16 ± 0.04 b | -28.41 ± 0.02 a | 4.70 ± 1.43 a |
|  | 7.4 | 0.57 ± 0.02 a | 0.66 ± 0.01 a | \_ | 6.55 ± 0.77 a | 0.17 ± 0.03 b | -29.06 ± 0.35 a | 4.69 ± 0.38 a |
| 14 days | 8.1 | 0.58 ± 0.02 a | 0.67 ± 0.01 a | 0.22 ± 0.05 b | \_ | \_ | \_ | \_ |
|  | 7.7 | 0.56 ± 0.01 a | 0.66 ± 0.01 a | 0.09 ± 0.05 b | \_ | \_ | \_ | \_ |
|  | 7.4 | 0.56 ± 0.01 a | 0.65 ± 0.01 a | -0.01 ± 0.03 a | \_ | \_ | \_ | \_ |
| 28 days | 8.1 | 0.58 ± 0.01 b | 0.67 ± 0.01 a | 0.18 ± 0.05 b | 9.24 ± 0.99 a | 0.16 ± 0.02 b | -27.78 ± 0.40 a | 5.64 ± 0.26 b |
|  | 7.7 | 0.55 ± 0.02 ab | 0.63 ± 0.01 a | -0.04 ± 0.03 ab | 6.84 ± 1.04 a | 0.13 ± 0.03 ab | -28.75 ± 0.58 a | 4.80 ± 0.41 ab |
|  | 7.4 | 0.49 ± 0.02 a | 0.63 ± 0.02 a | -0.14 ± 0.13 a | 7.11 ± 1.95 a | 0.03 ± 0.02 a | -30.56 ± 0.40 a | 4.45 ± 0.06 a |



**Fig. S1.** Fluctuations in the seawater pH over the course of the experiment. The real-time pH values of seawater during the period of (A) 24 days and (B) 24 hours for each pH treatment were recorded using CO2SYS ZB-LS 3.0. Values are expressed as mean ± SE.