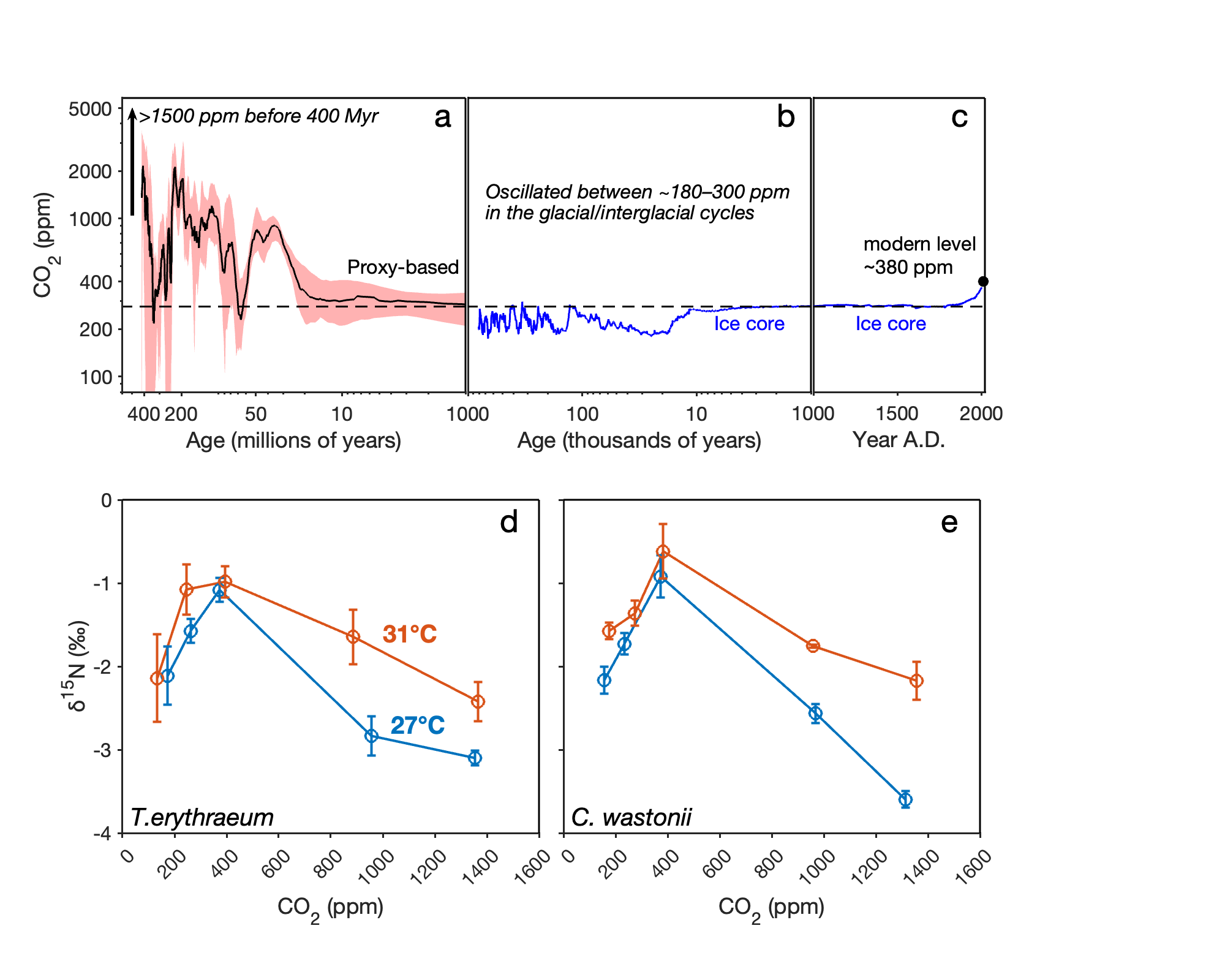
*Supplement of*

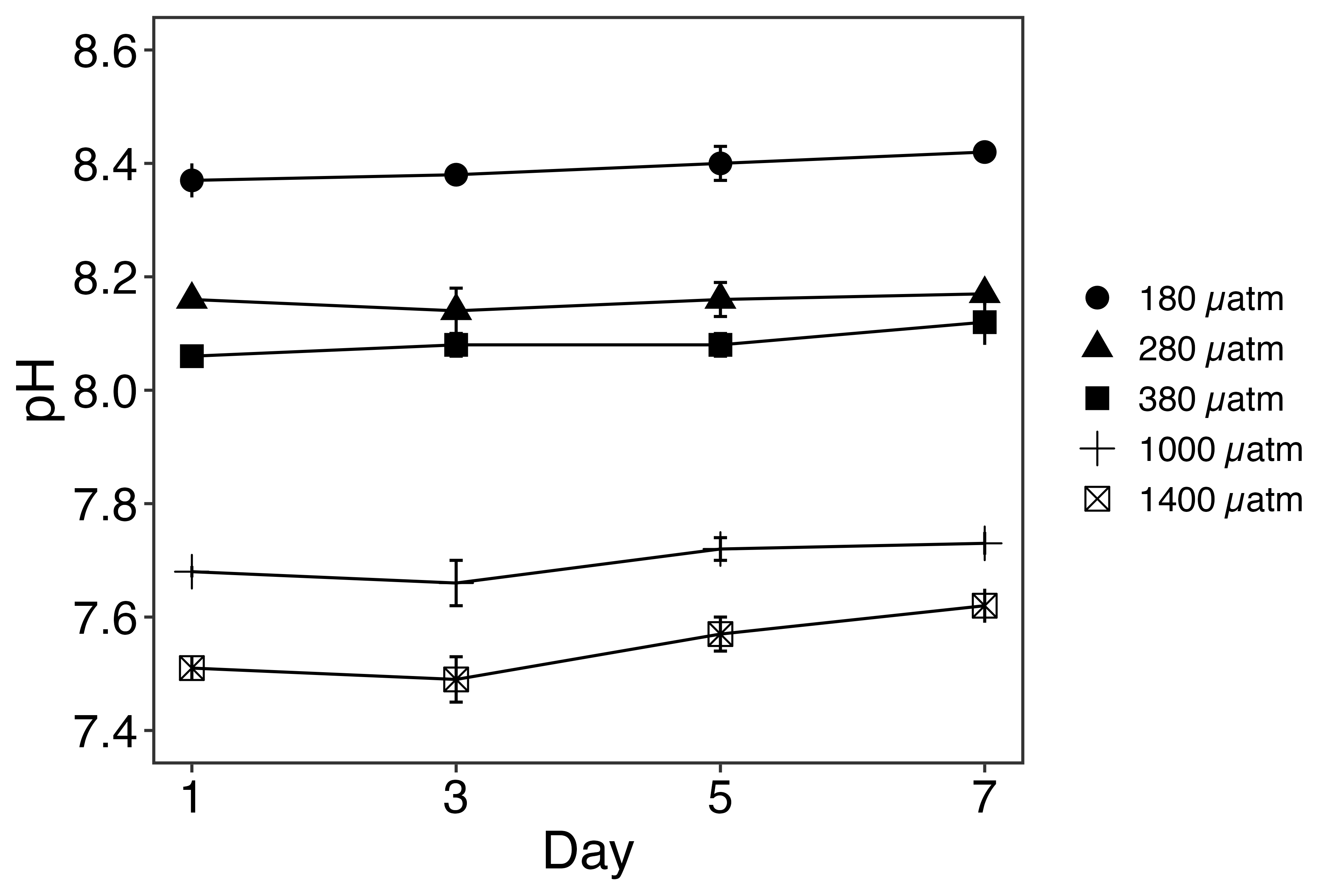
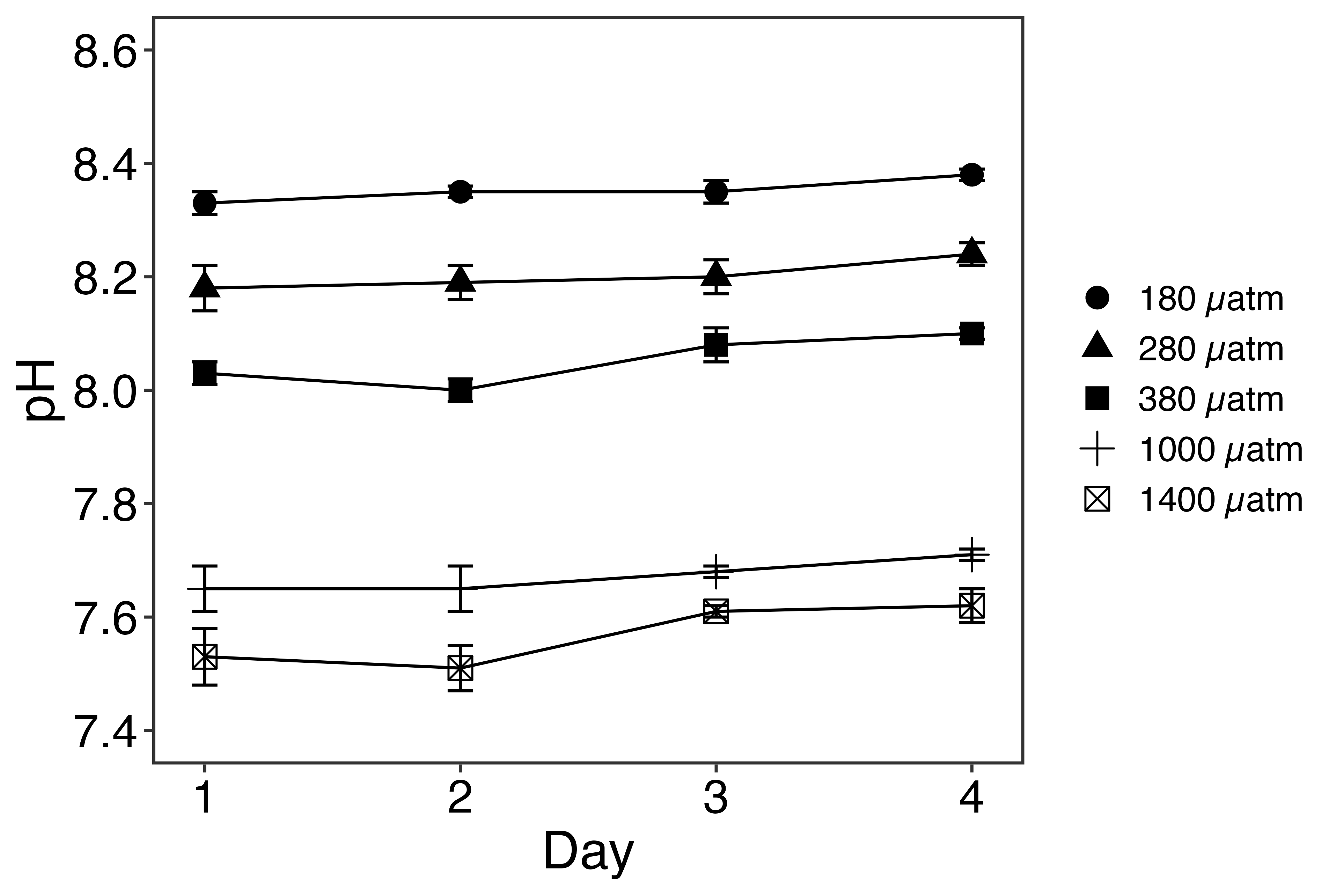
**Effects of CO2 on the nitrogen isotopic composition of *Trichodesmium* and *Crocosphaera***

Zuozhu Wen et al.

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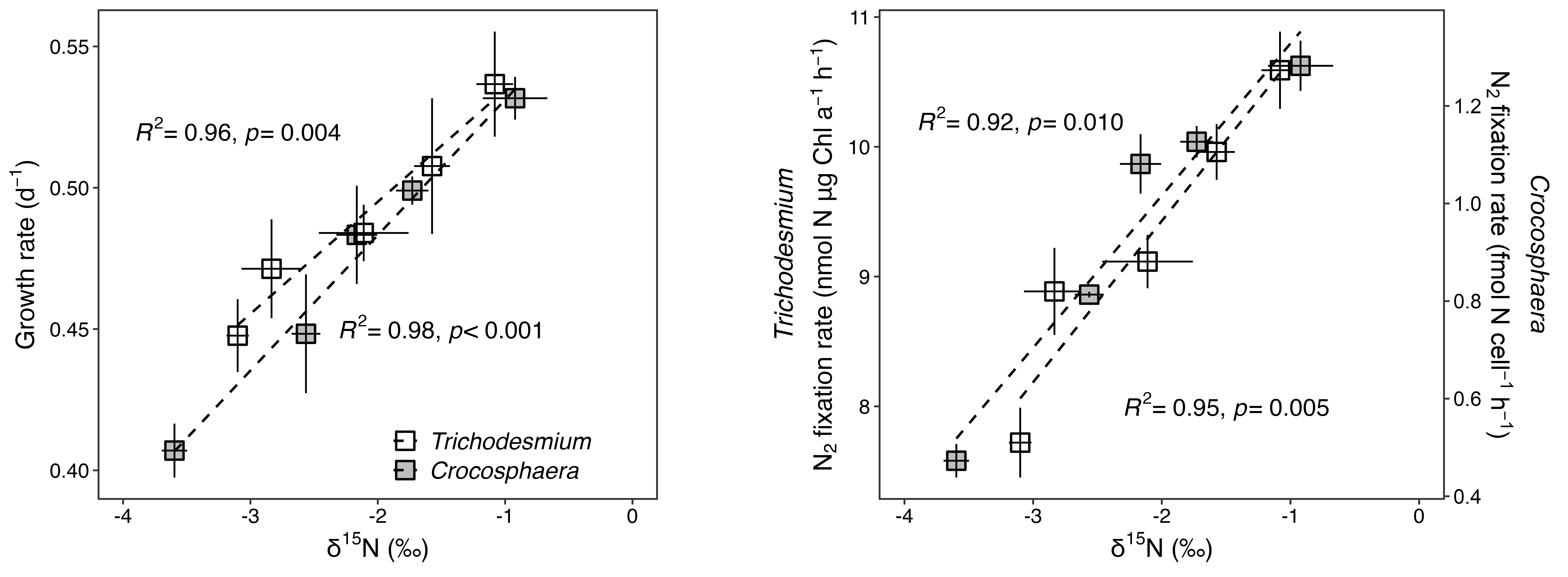
**Figure S1. Atmospheric CO2 fluctuations over the last 420 million years.** Proxy-based atmospheric CO2 on a log timescale and associated uncertainty envelope (red shadow) (Foster et al., 2017). (b and c) ﻿Ice core atmospheric CO2 from Bereiter et al. (2015). The dashed lines in (a), (b) and (c) are pre-industrial CO2 (278 ppm). Black dot in (c) is the modern CO2 level (~380 ppm).



a

b

**Figure S2. pH values throughout the experimental period.** (a) *Trichodesmium* and (b) *Crocosphaera*. Symbols represent the target CO2 levels manipulated. The values are presented as mean ± SD (n = 3).



a

b

**Figure S3. Relationships of biomass δ15N against growth and N2 fixation rates.** (a) Growth rate and (b) N2 fixation rate. Open squares, *Trichodesmium*; Grey squares, *Crocosphaera*. Note that N2 fixation rates of *Trichodesmium* and *Crocosphaera* are measured in different units. The values are presented as mean ± SD (n = 3)

**Table S1. Summary of data reported in this study.** Data includes carbonate chemistry (*p*CO2, µatm; pH; DIC, μmol kg-1), biomass δ15N (‰), growth rate (d-1), N2 fixation rate (*Trichodesmium*, nmol N µg Chla-1 h-1; *Crocosphaera*, fmol N cell-1 h-1), *NifH* protein abundance (pmol μg protein-1), and Nitrogenase efficiency (mol N mol *NifH*-1 min-1). Values are presented as mean ± SD (n=3).

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Species** | ***p*CO2** | **pH** | **DIC** | **Biomass δ15N** | **Growth rate** | **N2 fixation rate** | ***NifH* abundance** | **Nitrogenase efficiency** |
| ***T. erythraeum* IMS101** | 174±12 | 8.33±0.02 | ﻿1891±19 | -2.1±0.4 | 0.48±0.01 | 9.12±0.21 | 0.44±0.08 | 41.4±5.8 |
| 262±30 | 8.18±0.04 | ﻿1889±16 | -1.6±0.1 | 0.51±0.02 | 9.96±0.21 | 0.35±0.05 | 57.6±9.4 |
| 373±11 | 8.03±0.02 | ﻿1899±11 | -1.1±0.1 | 0.54±0.02 | 10.59±0.30 | 0.34±0.06 | 63.2±9.5 |
| 956±22 | 7.65±0.04 | ﻿1873±7 | -2.8±0.2 | 0.47±0.02 | 8.89±0.34 | 0.65±0.10 | 27.5±4.0 |
| 1355±21 | 7.53±0.05 | ﻿1869±18 | -3.1±0.1 | 0.45±0.01 | 7.72±0.27 | 0.71±0.17 | 22.2±4.9 |
| ***C. watsonii* WH8501** | ﻿﻿157±13 | 8.37±0.03 | ﻿1884±8 | -2.2±0.2 | 0.48±0.02 | 1.08±0.06 | 0.78±0.17 | 22.1±5.4 |
| ﻿234±12 | ﻿8.16±0.01 | ﻿1890±5 | -1.7±0.1 | 0.50±0.01 | 1.13±0.03 | 0.60±0.11 | 29.9±5.8 |
| ﻿374±6 | ﻿8.06±0.01 | ﻿1892±7 | -0.9±0.3 | 0.53±0.01 | 1.28±0.05 | 0.47±0.11 | 43.4±7.7 |
| ﻿969±22 | ﻿7.68±0.01 | ﻿1872±3 | -2.6±0.1 | 0.45±0.02 | 0.81±0.01 | 1.18±0.28 | 11.1±3.0 |
| ﻿1313±16 | ﻿7.51±0.02 | ﻿1867±10 | -3.6±0.1 | 0.41±0.01 | 0.47±0.03 | 1.24±0.29 | 6.2±2.0 |

**Reference:**

Bereiter, B., Eggleston, S., Schmitt, J., Nehrbass-Ahles, C., Stocker, T. F., Fischer, H., Kipfstuhl, S., and Chappellaz, J.: Revision of the EPICA Dome C CO2 record from 800 to 600 kyr before present, Geophys. Res. Lett., 42, 542-549, <https://doi.org/10.1002/2014GL061957>, 2015.

Foster, G. L., Royer, D. L., and Lunt, D. J.: Future climate forcing potentially without precedent in the last 420 million years, Nat. Commun., 8, 14845, <https://doi.org/10.1038/ncomms14845>, 2017.