

**Table S1.** Apparent activation energy ( $E_a$ , kJ mol<sup>-1</sup>) and temperature coefficient ( $Q_{10}$ ) of the sediment–water fluxes of  $\text{NO}_2^-$ ,  $\text{NH}_4^+$ ,  $\text{N}_2\text{O}$  and  $\text{CH}_4$  on each experimental day (1–4).  $E_a$  was calculated from Eq. 2 and  $Q_{10}$  from Eq. 3, referenced to 18 °C; SE, standard error. The temperature range used for each Arrhenius fit is indicated (°C).

|                                   | <i>Day</i> | <i>E<sub>a</sub></i> | <i>E<sub>a</sub> SE</i> | <i>Q<sub>10</sub></i> | <i>Q<sub>10</sub> SE</i> | <i>T range (°C)</i> |
|-----------------------------------|------------|----------------------|-------------------------|-----------------------|--------------------------|---------------------|
| <i>NO<sub>2</sub><sup>-</sup></i> | <b>1</b>   | 25,46                | 3,76                    | 1,39                  | 1,05                     | 10-34               |
|                                   | <b>2</b>   | 34,50                | 8,09                    | 1,61                  | 1,11                     | 10-26               |
|                                   | <b>3</b>   | 25,30                | 4,05                    | 1,41                  | 1,05                     | 10-26               |
|                                   | <b>4</b>   | 6,83                 | 7,94                    | 1,10                  | 1,11                     | 10-34               |
| <i>NH<sub>4</sub><sup>+</sup></i> | <b>1</b>   | 31,04                | 7,37                    | 1,53                  | 1,10                     | 10-34               |
|                                   | <b>2</b>   | 38,54                | 7,54                    | 1,70                  | 1,10                     | 10-34               |
|                                   | <b>3</b>   | 17,34                | 10,63                   | 1,27                  | 1,15                     | 10-34               |
|                                   | <b>4</b>   | 82,00                | 31,44                   | 3,08                  | 1,51                     | 10-34               |
| <i>N<sub>2</sub>O</i>             | <b>1</b>   | 13,16                | 5,45                    | 1,19                  | 1,07                     | 10-42               |
|                                   | <b>2</b>   | 12,27                | 4,09                    | 1,18                  | 1,05                     | 10-34               |
|                                   | <b>3</b>   | 4,00                 | 6,94                    | 1,06                  | 1,09                     | 10-42               |
|                                   | <b>4</b>   | 3,20                 | 2,58                    | 1,04                  | 1,03                     | 10-42               |
| <i>CH<sub>4</sub></i>             | <b>1</b>   | 73,57                | 15,39                   | 2,74                  | 1,22                     | 10-42               |
|                                   | <b>2</b>   | 99,87                | 15,22                   | 3,94                  | 1,22                     | 10-42               |
|                                   | <b>3</b>   | 92,01                | 31,28                   | 3,53                  | 1,50                     | 10-42               |
|                                   | <b>4</b>   | 76,46                | 11,72                   | 2,85                  | 1,16                     | 10-42               |

**Table S2.** Primer sets, quantitative PCR (qPCR) thermal-cycling conditions and standard-curve characteristics (amplification efficiency and R<sup>2</sup>) for the nitrogen-cycle functional genes targeted in this study.

| Process |  | Target<br>gene | Primer<br>set | Sequence  | q-PCR<br>conditions used | Reference   | Efficiency<br>(%) | Slope  | R <sup>2</sup> |
|---------|--|----------------|---------------|-----------|--------------------------|-------------|-------------------|--------|----------------|
|         |  | 16S rDNA       | Bact13        | CGGTGAATA | 95°C-15min;              | Suzuki et   | 92.4              | -3.518 | 0.998          |
|         |  |                | 69F           | CGTTCYCGG | 40x (95°C-               | al., (2000) |                   |        |                |
|         |  |                | Prok14        | GGWTACCTT | 10sec; 59°C-             |             |                   |        |                |
|         |  |                | 92R           | GTTAC     | 15sec; 72°C-             |             |                   |        |                |
|         |  |                |               | GACTT     | 20sec)                   |             |                   |        |                |

|                 |                                      |             |                                                           |                                                       |                                                                      |                                           |      |        |       |
|-----------------|--------------------------------------|-------------|-----------------------------------------------------------|-------------------------------------------------------|----------------------------------------------------------------------|-------------------------------------------|------|--------|-------|
| Denitrification | Copper containing nitrite reductases | <b>nirK</b> | <b>nirK</b><br><b>FlaCu</b><br><b>nirK</b><br><b>R3Cu</b> | ATCATGGTSC<br>TGCCGCG<br>GCCTCGATC<br>AGRTTGTGGT<br>T | 95°C-15min;<br>40x (95°C-<br>15sec; 57°C-<br>20sec; 72°C-<br>1min)   | Hallin and<br>Lindgren,<br>(1999)         | 92.0 | -3.530 | 0.997 |
|                 | Haem containing nitrite reductases   | <b>nirS</b> | <b>nirS1F</b><br><b>nirS</b><br><b>3R</b>                 | CCTAYTGGC<br>CGGCR CART<br>GCCGCCGTC<br>RTGVAGGAA     | 95°C-15min;<br>40x (95°C-<br>15sec; 57°C-<br>20sec; 72°C-<br>1min)   | Levy-<br>Booth and<br>Winder,<br>(2010)   | 99.3 | -3.338 | 0.997 |
|                 | nitrous oxide reductase              | <b>nosZ</b> | <b>nosZ-</b><br><b>1527F-</b><br><b>q</b><br><b>nosZ-</b> | CGCTGTTCHT<br>CGACAGYCA<br>ATRTCGATCA<br>RCTGBTCGTT   | 95°C-15 min;<br>40x (95°C-15<br>seg; 56°C-15<br>seg; 72°C-30<br>seg) | Modified<br>Scala &<br>Kerkhoff<br>(1998) | 83.4 | -3.796 | 0.996 |

[illegible]

|                                                                |                                                                      |             |                                                                    |                                                            |                                                                    |                          |      |        |       |
|----------------------------------------------------------------|----------------------------------------------------------------------|-------------|--------------------------------------------------------------------|------------------------------------------------------------|--------------------------------------------------------------------|--------------------------|------|--------|-------|
| <b>Dissimilatory<br/>nitrate<br/>reduction to<br/>ammonium</b> | dissimilatory<br>periplasmic<br>cytochrome<br>c nitrite<br>reductase | <b>nrfA</b> | <b>nrfAF</b><br><br><b>2aw</b><br><br><b>nrfAR</b><br><br><b>1</b> | CARTGYCAY<br><br>GTBGARTA<br><br>TWNGGCATR<br><br>TGRCAATC | 95°C-15min;<br>40x (94°C-<br>30sec; 48°C-<br>30sec; 72°C-<br>1min) | Welsh et<br>al., (2014)  | 87.2 | -3.671 | 0.994 |
| <b>Ammonia<br/>oxidation<br/>(Bacteria)</b>                    | ammonia<br>monooxygenase                                             | <b>AmoA</b> | <b>BacamoA-1F</b><br><br><b>BacamoA-2R</b>                         | GGGGHTTYT<br>ACTGGTGGT<br>CCCCTCBGSA<br>AAVCCCTTCTT<br>C   | 95°C-15min;<br>40x (95°C-30<br>sec, 47°C-30<br>sec, 72°C-1<br>min) | Hornek et<br>al., (2006) | 90.7 | -3.566 | 1.000 |

|                                   |                                    |      |                       |                                       |                                                                    |                           |       |        |       |
|-----------------------------------|------------------------------------|------|-----------------------|---------------------------------------|--------------------------------------------------------------------|---------------------------|-------|--------|-------|
| Ammonia<br>oxidation<br>(Archaea) |                                    | amoA | Arch-<br>amoW<br>AF   | CTGAYTGCG<br>CYTGGACAT<br>C           | 95°C-15min;<br>40x (95°C-<br>30sec; 58°C-<br>40sec, 72°C-<br>1min) | Wuchter et<br>al., (2006) | 105.7 | -3.192 | 0.997 |
|                                   |                                    |      | Arch-<br>amoW<br>AR   | TTCTTCTTTG<br>TTGCCCAGTA              |                                                                    |                           |       |        |       |
|                                   | Ammonia<br>oxidation<br>(Comammox) |      | Ntsp-<br>amoA<br>162F | GGATTCTCGG<br>NTSGATTGGA<br>WAGTTNGAC | 95°C-15min;<br>40x (94°C-<br>30sec; 48°C-<br>30sec; 72°C-<br>1min) | Fowler et<br>al., (2018)  | 87.2  | -3.672 | 0.997 |
|                                   |                                    |      | Ntspa<br>moA<br>359R  | CACCASTACC<br>A                       |                                                                    |                           |       |        |       |
|                                   |                                    |      |                       |                                       |                                                                    |                           |       |        |       |

**Table S3.** Likelihood-ratio test (LRT) results for the effects of temperature, exposure time and their interaction on sediment–water fluxes and oxygen uptake. Main effects were evaluated using additive models and interpreted primarily when the interaction was not significant.

| Response                     | Effect                      | $\chi^2$ | df | p        | Interpretation                   |
|------------------------------|-----------------------------|----------|----|----------|----------------------------------|
| DOU                          | Temperature                 | 34.27    | 4  | < 0.0001 | Main-effect LRT (additive model) |
|                              | Exposure time               | 1.04     | 3  | 0.7926   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | 0.0002   |                                  |
|                              |                             | 37.03    | 12 |          | Interaction LRT                  |
| TOU                          | Temperature                 | 8.20     | 4  | 0.0845   | Main-effect LRT (additive model) |
|                              | Exposure time               | 10.83    | 3  | 0.0127   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | < 0.0001 |                                  |
|                              |                             | 45.47    | 12 |          | Interaction LRT                  |
| N <sub>2</sub> O             | Temperature                 | 5.77     | 4  | 0.2172   | Main-effect LRT (additive model) |
|                              | Exposure time               | 0.96     | 3  | 0.8112   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | 0.0010   |                                  |
|                              |                             | 32.92    | 12 |          | Interaction LRT                  |
| CH <sub>4</sub>              | Temperature                 | 43.43    | 4  | < 0.0001 | Main-effect LRT (additive model) |
|                              | Exposure time               | 6.10     | 3  | 0.1069   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | 0.1683   |                                  |
|                              |                             | 16.53    | 12 |          | Interaction LRT                  |
| NO <sub>2</sub> <sup>-</sup> | Temperature                 | 34.08    | 4  | < 0.0001 | Main-effect LRT (additive model) |
|                              | Exposure time               | 4.71     | 3  | 0.1940   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | 0.0096   |                                  |
|                              |                             | 26.34    | 12 |          | Interaction LRT                  |
| NO <sub>3</sub> <sup>-</sup> | Temperature                 | 1.22     | 4  | 0.8749   | Main-effect LRT (additive model) |
|                              | Exposure time               | 13.97    | 3  | 0.0029   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | < 0.0001 |                                  |
|                              |                             | 58.43    | 12 |          | Interaction LRT                  |
| NH <sub>4</sub> <sup>+</sup> | Temperature                 | 25.90    | 4  | < 0.0001 | Main-effect LRT (additive model) |
|                              | Exposure time               | 41.57    | 3  | < 0.0001 | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | 0.0085   |                                  |
|                              |                             | 26.71    | 12 |          | Interaction LRT                  |
| DIC                          | Temperature                 | 20.62    | 4  | 0.0004   | Main-effect LRT (additive model) |
|                              | Exposure time               | 18.92    | 3  | 0.0003   | Main-effect LRT (additive model) |
|                              | Temperature × exposure time |          |    | < 0.0001 |                                  |
|                              |                             | 63.17    | 12 |          | Interaction LRT                  |

**Table S4.** Significant Tukey-adjusted pairwise comparisons among temperatures within each exposure time for variables showing a significant temperature  $\times$  exposure-time interaction. Degrees of freedom were estimated using the Satterthwaite method.

| Response                     | Exposure time (h) | Contrast | Estimate | SE   | df   | t-ratio | Adjusted p |
|------------------------------|-------------------|----------|----------|------|------|---------|------------|
| DOU                          | 12                | 10 - 42  | 0.55     | 0.19 | 60.0 | 2.84    | 0.0464     |
| DOU                          | 36                | 10 - 26  | 0.75     | 0.19 | 60.0 | 3.84    | 0.0026     |
| DOU                          | 36                | 10 - 34  | 1.04     | 0.19 | 60.0 | 5.34    | < 0.0001   |
| DOU                          | 36                | 10 - 42  | 1.30     | 0.19 | 60.0 | 6.70    | < 0.0001   |
| DOU                          | 36                | 18 - 26  | 0.56     | 0.19 | 60.0 | 2.91    | 0.0392     |
| DOU                          | 36                | 18 - 34  | 0.85     | 0.19 | 60.0 | 4.41    | 0.0004     |
| DOU                          | 36                | 18 - 42  | 1.12     | 0.19 | 60.0 | 5.76    | < 0.0001   |
| DOU                          | 36                | 26 - 42  | 0.55     | 0.19 | 60.0 | 2.85    | 0.0453     |
| DOU                          | 60                | 10 - 26  | 0.69     | 0.19 | 60.0 | 3.57    | 0.0061     |
| DOU                          | 60                | 10 - 34  | 1.12     | 0.19 | 60.0 | 5.76    | < 0.0001   |
| DOU                          | 60                | 10 - 42  | 1.31     | 0.19 | 60.0 | 6.75    | < 0.0001   |
| DOU                          | 60                | 18 - 34  | 0.59     | 0.19 | 60.0 | 3.02    | 0.0297     |
| DOU                          | 60                | 18 - 42  | 0.78     | 0.19 | 60.0 | 4.01    | 0.0016     |
| DOU                          | 60                | 26 - 42  | 0.62     | 0.19 | 60.0 | 3.18    | 0.0189     |
| DOU                          | 84                | 10 - 26  | 1.36     | 0.19 | 60.0 | 7.00    | < 0.0001   |
| DOU                          | 84                | 10 - 34  | 0.80     | 0.19 | 60.0 | 4.14    | 0.0010     |
| DOU                          | 84                | 10 - 42  | 0.62     | 0.19 | 60.0 | 3.20    | 0.0178     |
| DOU                          | 84                | 18 - 26  | 0.85     | 0.19 | 60.0 | 4.40    | 0.0004     |
| DOU                          | 84                | 26 - 34  | -0.56    | 0.19 | 60.0 | -2.87   | 0.0438     |
| DOU                          | 84                | 26 - 42  | -0.74    | 0.19 | 60.0 | -3.80   | 0.0030     |
| TOU                          | 12                | 10 - 26  | 1.74     | 0.50 | 20.0 | 3.48    | 0.0179     |
| TOU                          | 12                | 10 - 34  | 1.77     | 0.50 | 20.0 | 3.54    | 0.0155     |
| TOU                          | 36                | 10 - 42  | 1.66     | 0.50 | 20.0 | 3.32    | 0.0250     |
| N <sub>2</sub> O             | 12                | 10 - 42  | -0.22    | 0.05 | 57.2 | -4.60   | 0.0002     |
| N <sub>2</sub> O             | 12                | 18 - 42  | -0.21    | 0.05 | 57.2 | -4.20   | 0.0009     |
| N <sub>2</sub> O             | 12                | 26 - 42  | -0.23    | 0.05 | 57.2 | -4.67   | 0.0002     |
| N <sub>2</sub> O             | 12                | 34 - 42  | -0.16    | 0.05 | 57.2 | -3.21   | 0.0179     |
| NO <sub>2</sub> <sup>-</sup> | 12                | 10 - 26  | -18.69   | 4.87 | 60.0 | -3.84   | 0.0027     |
| NO <sub>2</sub> <sup>-</sup> | 12                | 10 - 34  | -24.19   | 4.87 | 60.0 | -4.97   | 0.0001     |
| NO <sub>2</sub> <sup>-</sup> | 12                | 26 - 42  | 18.03    | 4.87 | 60.0 | 3.71    | 0.0041     |
| NO <sub>2</sub> <sup>-</sup> | 12                | 34 - 42  | 23.53    | 4.87 | 60.0 | 4.83    | 0.0001     |

| Response                     | Exposure time (h) | Contrast | Estimate | SE     | df   | t-ratio | Adjusted p |
|------------------------------|-------------------|----------|----------|--------|------|---------|------------|
| NO <sub>2</sub> <sup>-</sup> | 36                | 10 - 26  | -27.61   | 4.87   | 60.0 | -5.67   | < 0.0001   |
| NO <sub>2</sub> <sup>-</sup> | 36                | 18 - 26  | -26.10   | 4.87   | 60.0 | -5.36   | < 0.0001   |
| NO <sub>2</sub> <sup>-</sup> | 36                | 26 - 34  | 19.84    | 4.87   | 60.0 | 4.08    | 0.0012     |
| NO <sub>2</sub> <sup>-</sup> | 36                | 26 - 42  | 29.65    | 4.87   | 60.0 | 6.09    | < 0.0001   |
| NO <sub>2</sub> <sup>-</sup> | 60                | 10 - 26  | -20.26   | 4.87   | 60.0 | -4.16   | 0.0009     |
| NO <sub>2</sub> <sup>-</sup> | 60                | 10 - 42  | 15.39    | 4.87   | 60.0 | 3.16    | 0.0200     |
| NO <sub>2</sub> <sup>-</sup> | 60                | 18 - 26  | -16.11   | 4.87   | 60.0 | -3.31   | 0.0132     |
| NO <sub>2</sub> <sup>-</sup> | 60                | 18 - 42  | 19.53    | 4.87   | 60.0 | 4.01    | 0.0015     |
| NO <sub>2</sub> <sup>-</sup> | 60                | 26 - 34  | 16.92    | 4.87   | 60.0 | 3.48    | 0.0081     |
| NO <sub>2</sub> <sup>-</sup> | 60                | 26 - 42  | 35.65    | 4.87   | 60.0 | 7.32    | < 0.0001   |
| NO <sub>2</sub> <sup>-</sup> | 60                | 34 - 42  | 18.72    | 4.87   | 60.0 | 3.85    | 0.0026     |
| NO <sub>2</sub> <sup>-</sup> | 84                | 10 - 42  | 15.22    | 4.87   | 60.0 | 3.13    | 0.0220     |
| NO <sub>2</sub> <sup>-</sup> | 84                | 18 - 42  | 17.95    | 4.87   | 60.0 | 3.69    | 0.0043     |
| NO <sub>2</sub> <sup>-</sup> | 84                | 26 - 42  | 23.98    | 4.87   | 60.0 | 4.93    | 0.0001     |
| NO <sub>2</sub> <sup>-</sup> | 84                | 34 - 42  | 21.82    | 4.87   | 60.0 | 4.48    | 0.0003     |
| NO <sub>3</sub> <sup>-</sup> | 36                | 10 - 26  | -68.57   | 22.22  | 57.1 | -3.09   | 0.0250     |
| NO <sub>3</sub> <sup>-</sup> | 36                | 26 - 34  | 96.13    | 22.22  | 57.1 | 4.33    | 0.0006     |
| NO <sub>3</sub> <sup>-</sup> | 36                | 26 - 42  | 79.45    | 22.22  | 57.1 | 3.58    | 0.0062     |
| NO <sub>3</sub> <sup>-</sup> | 60                | 10 - 26  | 147.01   | 22.22  | 57.1 | 6.62    | < 0.0001   |
| NO <sub>3</sub> <sup>-</sup> | 60                | 18 - 26  | 88.99    | 22.22  | 57.1 | 4.01    | 0.0016     |
| NO <sub>3</sub> <sup>-</sup> | 60                | 26 - 34  | -96.18   | 22.22  | 57.1 | -4.33   | 0.0006     |
| NO <sub>3</sub> <sup>-</sup> | 60                | 26 - 42  | -93.87   | 22.22  | 57.1 | -4.22   | 0.0008     |
| NO <sub>3</sub> <sup>-</sup> | 84                | 10 - 26  | -62.75   | 22.22  | 57.1 | -2.82   | 0.0491     |
| NO <sub>3</sub> <sup>-</sup> | 84                | 18 - 26  | -64.65   | 22.22  | 57.1 | -2.91   | 0.0396     |
| NO <sub>3</sub> <sup>-</sup> | 84                | 26 - 42  | 93.71    | 22.22  | 57.1 | 4.22    | 0.0008     |
| NO <sub>3</sub> <sup>-</sup> | 84                | 34 - 42  | 64.58    | 22.22  | 57.1 | 2.91    | 0.0399     |
| NH <sub>4</sub> <sup>+</sup> | 12                | 10 - 34  | 908.47   | 125.29 | 58.8 | 7.25    | < 0.0001   |
| NH <sub>4</sub> <sup>+</sup> | 12                | 10 - 42  | 611.52   | 125.29 | 58.8 | 4.88    | 0.0001     |
| NH <sub>4</sub> <sup>+</sup> | 12                | 18 - 34  | 591.48   | 125.29 | 58.8 | 4.72    | 0.0001     |
| NH <sub>4</sub> <sup>+</sup> | 12                | 26 - 34  | 733.01   | 125.29 | 58.8 | 5.85    | < 0.0001   |
| NH <sub>4</sub> <sup>+</sup> | 12                | 26 - 42  | 436.06   | 125.29 | 58.8 | 3.48    | 0.0081     |
| NH <sub>4</sub> <sup>+</sup> | 36                | 10 - 34  | 625.28   | 125.29 | 58.8 | 4.99    | 0.0001     |
| NH <sub>4</sub> <sup>+</sup> | 36                | 10 - 42  | 446.77   | 125.29 | 58.8 | 3.57    | 0.0063     |
| NH <sub>4</sub> <sup>+</sup> | 36                | 18 - 34  | 574.43   | 125.29 | 58.8 | 4.58    | 0.0002     |
| NH <sub>4</sub> <sup>+</sup> | 36                | 18 - 42  | 395.92   | 125.29 | 58.8 | 3.16    | 0.0203     |
| NH <sub>4</sub> <sup>+</sup> | 36                | 26 - 34  | 377.70   | 125.29 | 58.8 | 3.01    | 0.0299     |
| NH <sub>4</sub> <sup>+</sup> | 84                | 10 - 42  | 431.43   | 125.29 | 58.8 | 3.44    | 0.0091     |

| <b>Response</b> | <b>Exposure<br/>time (h)</b> | <b>Contrast</b> | <b>Estimate</b> | <b>SE</b> | <b>df</b> | <b>t-ratio</b> | <b>Adjusted p</b> |
|-----------------|------------------------------|-----------------|-----------------|-----------|-----------|----------------|-------------------|
| DIC             | 12                           | 10 - 18         | 7.29            | 2.42      | 60.0      | 3.02           | 0.0296            |
| DIC             | 12                           | 10 - 34         | -9.26           | 2.42      | 60.0      | -3.83          | 0.0027            |
| DIC             | 12                           | 18 - 34         | -16.56          | 2.42      | 60.0      | -6.85          | < 0.0001          |
| DIC             | 12                           | 18 - 42         | -12.67          | 2.42      | 60.0      | -5.24          | < 0.0001          |
| DIC             | 12                           | 26 - 34         | -15.20          | 2.42      | 60.0      | -6.29          | < 0.0001          |
| DIC             | 12                           | 26 - 42         | -11.31          | 2.42      | 60.0      | -4.68          | 0.0002            |
| DIC             | 36                           | 10 - 42         | -12.76          | 2.42      | 60.0      | -5.28          | < 0.0001          |
| DIC             | 36                           | 18 - 42         | -9.87           | 2.42      | 60.0      | -4.08          | 0.0012            |
| DIC             | 36                           | 26 - 42         | -15.86          | 2.42      | 60.0      | -6.56          | < 0.0001          |
| DIC             | 36                           | 34 - 42         | -11.37          | 2.42      | 60.0      | -4.70          | 0.0001            |
| DIC             | 60                           | 10 - 42         | -14.27          | 2.42      | 60.0      | -5.90          | < 0.0001          |
| DIC             | 60                           | 18 - 42         | -14.65          | 2.42      | 60.0      | -6.06          | < 0.0001          |
| DIC             | 60                           | 26 - 42         | -13.73          | 2.42      | 60.0      | -5.68          | < 0.0001          |
| DIC             | 60                           | 34 - 42         | -14.03          | 2.42      | 60.0      | -5.80          | < 0.0001          |
| DIC             | 84                           | 34 - 42         | 6.88            | 2.42      | 60.0      | 2.84           | 0.0462            |

**Table S5.** Significant Tukey-adjusted pairwise comparisons among exposure times within each temperature for variables showing a significant temperature  $\times$  exposure-time interaction.

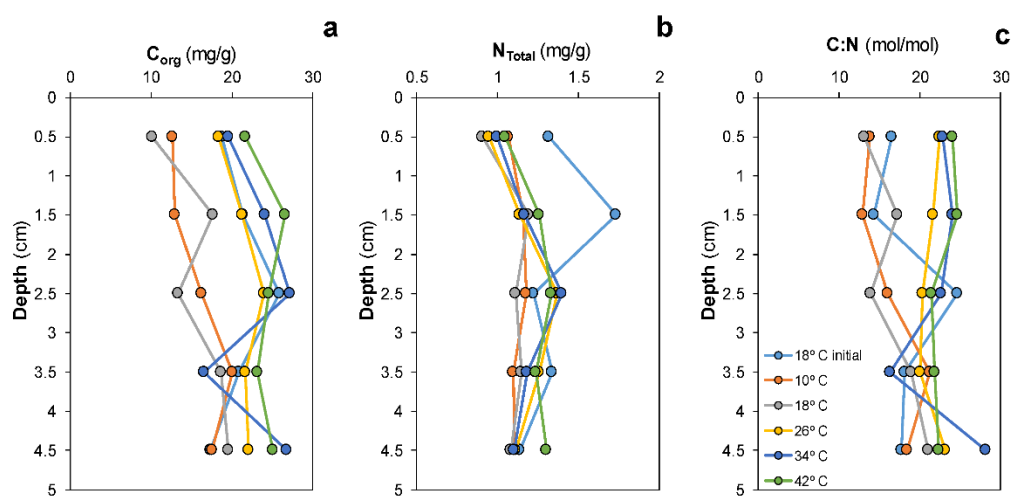
Degrees of freedom were estimated using the Satterthwaite method.

| Response                     | Temp. (°C) | Contrast | Estimate | SE    | df   | t-ratio | Adjusted p |
|------------------------------|------------|----------|----------|-------|------|---------|------------|
| DOU                          | 26         | 12 - 84  | 0.75     | 0.19  | 60.0 | 3.88    | 0.0015     |
| DOU                          | 26         | 60 - 84  | 0.68     | 0.19  | 60.0 | 3.51    | 0.0046     |
| DOU                          | 42         | 36 - 84  | -0.79    | 0.19  | 60.0 | -4.05   | 0.0008     |
| DOU                          | 42         | 60 - 84  | -0.67    | 0.19  | 60.0 | -3.47   | 0.0052     |
| TOU                          | 10         | 12 - 84  | 0.57     | 0.21  | 45.0 | 2.67    | 0.0492     |
| TOU                          | 10         | 36 - 84  | 0.83     | 0.21  | 45.0 | 3.88    | 0.0018     |
| TOU                          | 10         | 60 - 84  | 0.72     | 0.21  | 45.0 | 3.38    | 0.0079     |
| TOU                          | 18         | 12 - 60  | -0.65    | 0.21  | 45.0 | -3.02   | 0.0207     |
| TOU                          | 26         | 12 - 36  | -0.67    | 0.21  | 45.0 | -3.13   | 0.0157     |
| TOU                          | 26         | 12 - 60  | -0.85    | 0.21  | 45.0 | -3.96   | 0.0014     |
| TOU                          | 26         | 12 - 84  | -1.10    | 0.21  | 45.0 | -5.13   | < 0.0001   |
| TOU                          | 34         | 12 - 36  | -0.74    | 0.21  | 45.0 | -3.44   | 0.0066     |
| TOU                          | 34         | 12 - 60  | -1.01    | 0.21  | 45.0 | -4.72   | 0.0001     |
| TOU                          | 34         | 60 - 84  | 0.66     | 0.21  | 45.0 | 3.11    | 0.0166     |
| TOU                          | 42         | 12 - 36  | 0.95     | 0.21  | 45.0 | 4.44    | 0.0003     |
| TOU                          | 42         | 12 - 84  | 0.84     | 0.21  | 45.0 | 3.95    | 0.0015     |
| TOU                          | 42         | 36 - 60  | -0.75    | 0.21  | 45.0 | -3.52   | 0.0054     |
| TOU                          | 42         | 60 - 84  | 0.65     | 0.21  | 45.0 | 3.03    | 0.0205     |
| N <sub>2</sub> O             | 26         | 12 - 60  | -0.14    | 0.05  | 45.0 | -2.98   | 0.0234     |
| N <sub>2</sub> O             | 42         | 12 - 36  | 0.13     | 0.05  | 45.0 | 2.87    | 0.0306     |
| N <sub>2</sub> O             | 42         | 12 - 60  | 0.22     | 0.05  | 45.0 | 4.84    | 0.0001     |
| N <sub>2</sub> O             | 42         | 12 - 84  | 0.21     | 0.05  | 45.0 | 4.67    | 0.0002     |
| NO <sub>2</sub> <sup>-</sup> | 26         | 12 - 36  | -13.70   | 4.87  | 60.0 | -2.81   | 0.0325     |
| NO <sub>2</sub> <sup>-</sup> | 26         | 36 - 84  | 17.86    | 4.87  | 60.0 | 3.67    | 0.0028     |
| NO <sub>2</sub> <sup>-</sup> | 26         | 60 - 84  | 13.91    | 4.87  | 60.0 | 2.86    | 0.0291     |
| NO <sub>3</sub> <sup>-</sup> | 26         | 12 - 36  | -79.92   | 20.73 | 45.0 | -3.86   | 0.0020     |
| NO <sub>3</sub> <sup>-</sup> | 26         | 12 - 60  | 91.40    | 20.73 | 45.0 | 4.41    | 0.0004     |
| NO <sub>3</sub> <sup>-</sup> | 26         | 12 - 84  | -107.26  | 20.73 | 45.0 | -5.17   | < 0.0001   |
| NO <sub>3</sub> <sup>-</sup> | 26         | 36 - 60  | 171.32   | 20.73 | 45.0 | 8.27    | < 0.0001   |
| NO <sub>3</sub> <sup>-</sup> | 26         | 60 - 84  | -198.66  | 20.73 | 45.0 | -9.58   | < 0.0001   |

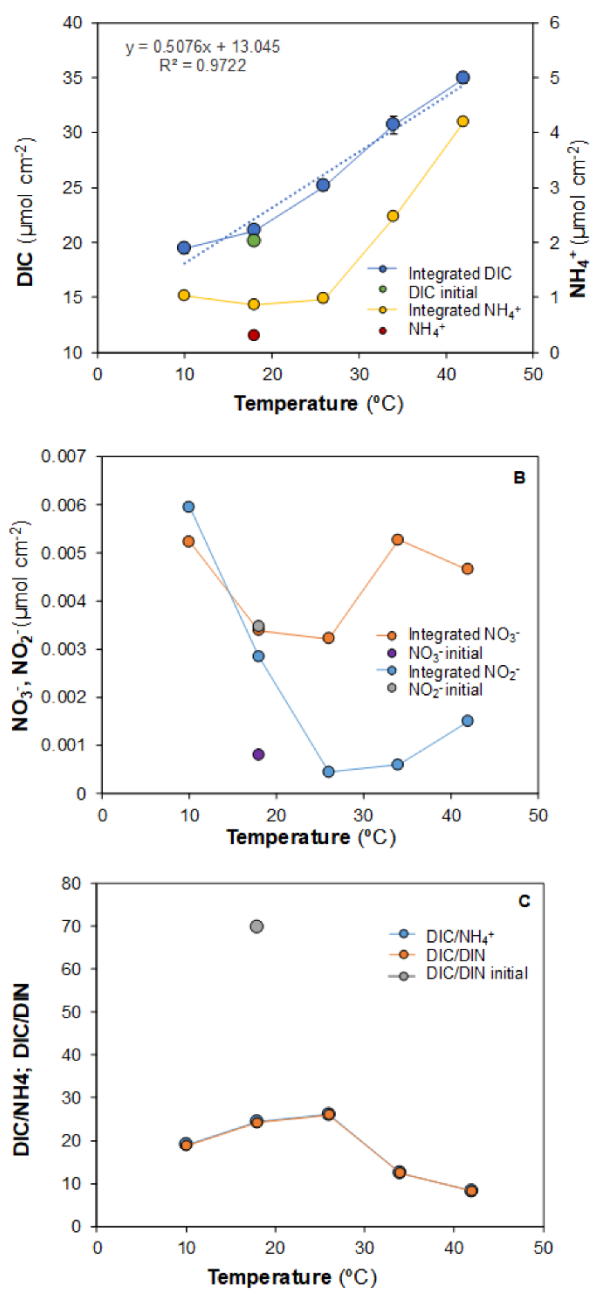
| Response                     | Temp. (°C) | Contrast | Estimate | SE     | df   | t-ratio | Adjusted p |
|------------------------------|------------|----------|----------|--------|------|---------|------------|
| NO <sub>3</sub> <sup>-</sup> | 34         | 12 - 84  | -59.54   | 20.73  | 45.0 | -2.87   | 0.0304     |
| NO <sub>3</sub> <sup>-</sup> | 34         | 36 - 84  | -94.33   | 20.73  | 45.0 | -4.55   | 0.0002     |
| NO <sub>3</sub> <sup>-</sup> | 34         | 60 - 84  | -73.35   | 20.73  | 45.0 | -3.54   | 0.0051     |
| NH <sub>4</sub> <sup>+</sup> | 18         | 12 - 36  | -401.06  | 120.07 | 45.0 | -3.34   | 0.0089     |
| NH <sub>4</sub> <sup>+</sup> | 18         | 12 - 84  | -529.76  | 120.07 | 45.0 | -4.41   | 0.0004     |
| NH <sub>4</sub> <sup>+</sup> | 34         | 12 - 36  | -418.11  | 120.07 | 45.0 | -3.48   | 0.0059     |
| NH <sub>4</sub> <sup>+</sup> | 34         | 12 - 60  | -822.44  | 120.07 | 45.0 | -6.85   | < 0.0001   |
| NH <sub>4</sub> <sup>+</sup> | 34         | 12 - 84  | -972.29  | 120.07 | 45.0 | -8.10   | < 0.0001   |
| NH <sub>4</sub> <sup>+</sup> | 34         | 36 - 60  | -404.33  | 120.07 | 45.0 | -3.37   | 0.0082     |
| NH <sub>4</sub> <sup>+</sup> | 34         | 36 - 84  | -554.18  | 120.07 | 45.0 | -4.62   | 0.0002     |
| NH <sub>4</sub> <sup>+</sup> | 42         | 12 - 60  | -492.42  | 120.07 | 45.0 | -4.10   | 0.0009     |
| NH <sub>4</sub> <sup>+</sup> | 42         | 12 - 84  | -475.56  | 120.07 | 45.0 | -3.96   | 0.0015     |
| DIC                          | 18         | 12 - 36  | -7.85    | 2.42   | 60.0 | -3.25   | 0.0100     |
| DIC                          | 18         | 36 - 84  | 7.90     | 2.42   | 60.0 | 3.27    | 0.0095     |
| DIC                          | 34         | 12 - 36  | 10.21    | 2.42   | 60.0 | 4.22    | 0.0005     |
| DIC                          | 34         | 12 - 60  | 11.24    | 2.42   | 60.0 | 4.65    | 0.0001     |
| DIC                          | 34         | 12 - 84  | 13.43    | 2.42   | 60.0 | 5.56    | < 0.0001   |
| DIC                          | 42         | 12 - 60  | -6.68    | 2.42   | 60.0 | -2.76   | 0.0373     |
| DIC                          | 42         | 12 - 84  | 16.42    | 2.42   | 60.0 | 6.79    | < 0.0001   |
| DIC                          | 42         | 36 - 84  | 21.47    | 2.42   | 60.0 | 8.88    | < 0.0001   |
| DIC                          | 42         | 60 - 84  | 23.10    | 2.42   | 60.0 | 9.55    | < 0.0001   |

**Table S6.** Tukey-adjusted pairwise comparisons among temperatures for CH<sub>4</sub> fluxes, averaged across exposure times, based on the additive linear mixed-effects model. CH<sub>4</sub> fluxes were transformed as  $\ln(\text{CH}_4 + 0.32)$  before analysis; therefore, estimates and standard errors are expressed on the transformed scale. Degrees of freedom were estimated using the Satterthwaite method.

| <b>Response</b> | <b>Contrast</b> | <b>Estimate</b> | <b>SE</b> | <b>df</b> | <b>t-ratio</b> | <b>Adjusted p</b> |
|-----------------|-----------------|-----------------|-----------|-----------|----------------|-------------------|
| CH <sub>4</sub> | 10 - 18         | 0.06            | 0.38      | 13.3      | 0.16           | 0.9998            |
| CH <sub>4</sub> | 10 - 26         | -0.09           | 0.38      | 13.3      | -0.24          | 0.9992            |
| CH <sub>4</sub> | 10 - 34         | -2.15           | 0.38      | 13.3      | -5.58          | 0.0007            |
| CH <sub>4</sub> | 10 - 42         | -3.57           | 0.38      | 13.3      | -9.28          | < 0.0001          |
| CH <sub>4</sub> | 18 - 26         | -0.15           | 0.38      | 13.3      | -0.40          | 0.9940            |
| CH <sub>4</sub> | 18 - 34         | -2.21           | 0.38      | 13.3      | -5.74          | 0.0005            |
| CH <sub>4</sub> | 18 - 42         | -3.63           | 0.38      | 13.3      | -9.44          | < 0.0001          |
| CH <sub>4</sub> | 26 - 34         | -2.06           | 0.38      | 13.3      | -5.34          | 0.0010            |
| CH <sub>4</sub> | 26 - 42         | -3.48           | 0.38      | 13.3      | -9.04          | < 0.0001          |
| CH <sub>4</sub> | 34 - 42         | -1.43           | 0.38      | 13.3      | -3.70          | 0.0180            |



**Figure S1.** Vertical profiles of sediment organic carbon ( $C_{org}$ ) content, total nitrogen ( $N_{Total}$ ) content and the molar carbon-to-nitrogen ratio (C:N) in the upper 5 cm, on the first (initial) and last day of the experiment at each temperature tested (10–42 °C).



**Fig. S2.** Depth-integrated porewater concentrations of dissolved inorganic carbon (DIC) and ammonium ( $\text{NH}_4^+$ ) in the sediment (upper 5 cm) as a function of temperature. For integration to

a common depth, missing 4–5 cm values were replaced by the corresponding 3–4 cm value: DIC at 42 °C, and  $\text{NH}_4^+$  at 34 and 42 °C (samples lost).

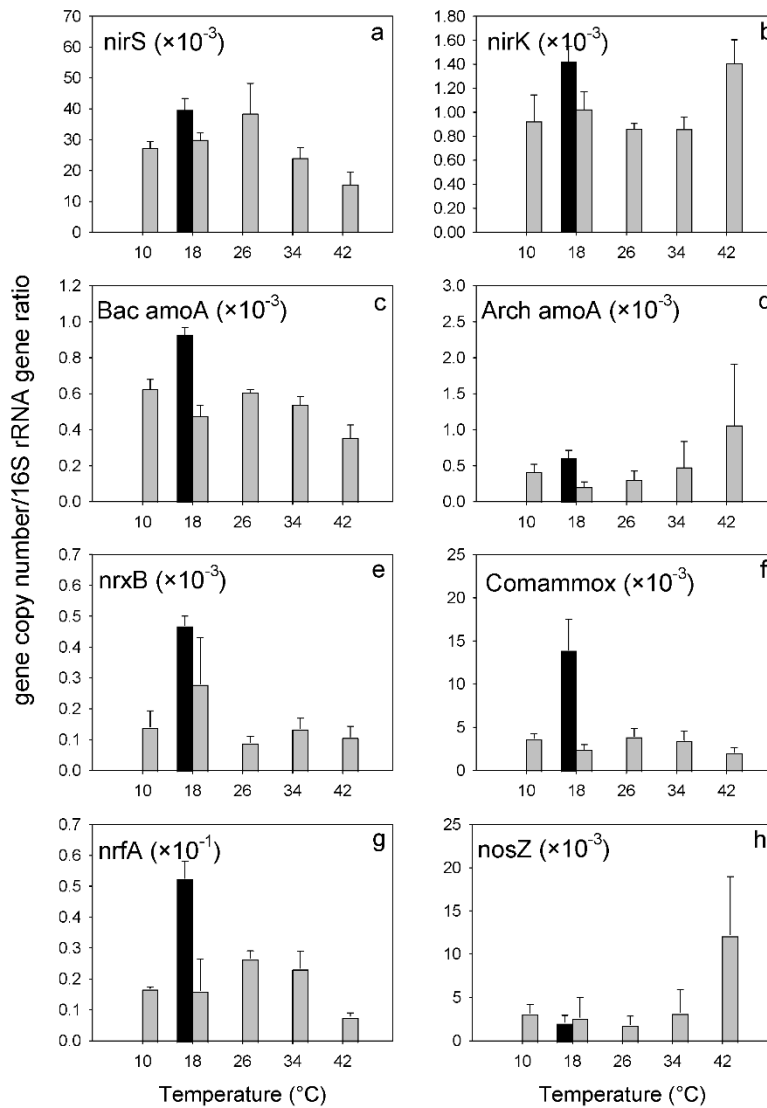
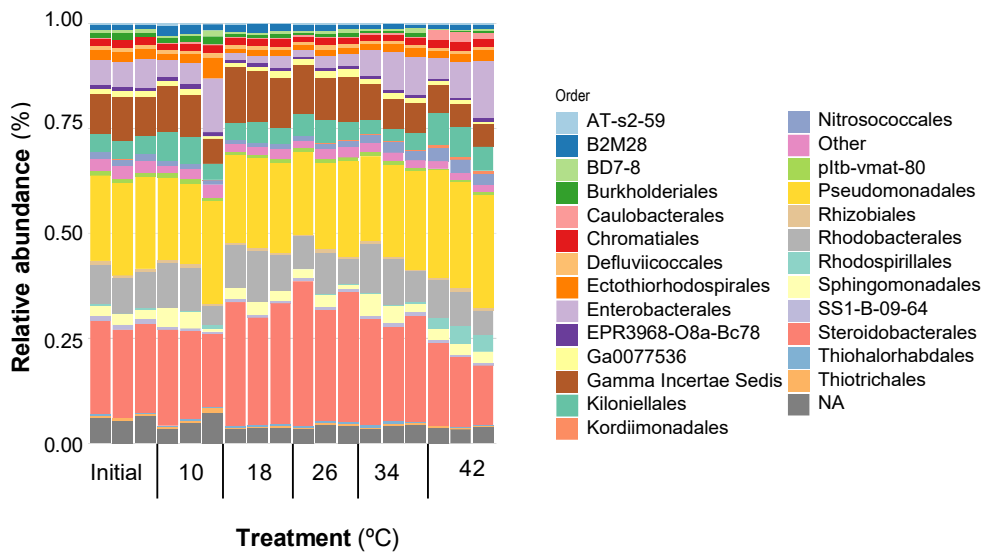
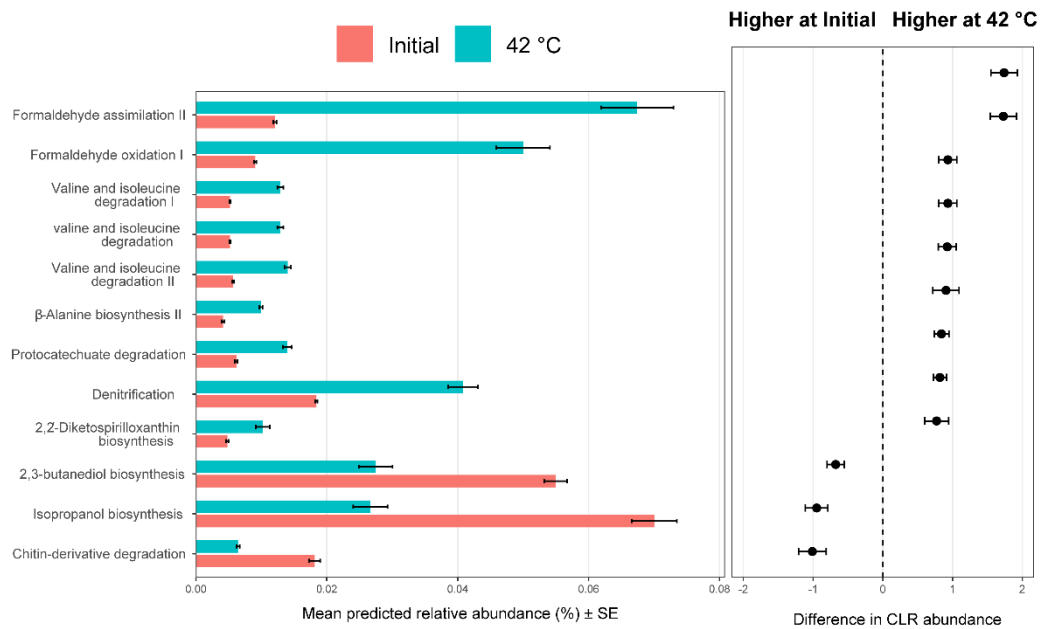


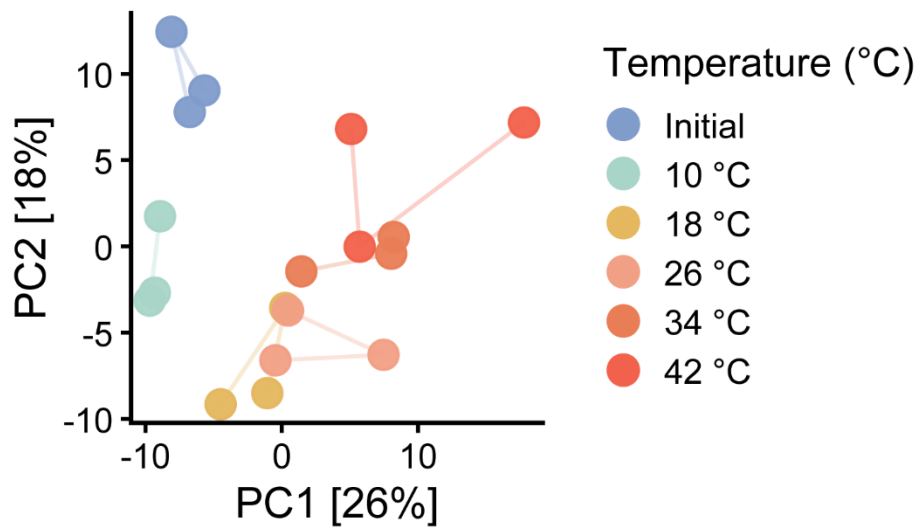
Fig S3. Abundance of nitrogen-cycling functional genes normalised to the 16S rRNA gene in the 0–1 cm sediment layer across the five temperatures (10, 18, 26, 34 and 42 °C): (a) nirS and (b) nirK (nitrite reductases; denitrification), (c) bacterial and (d) archaeal amoA (ammonia monooxygenase; ammonia oxidation), (e) nxrB (nitrite oxidoreductase; nitrite oxidation), (f) comammox amoA (complete ammonia oxidisers), (g) nrfA (marker of dissimilatory nitrate reduction to ammonium, DNRA) and (h) nosZ (nitrous oxide reductase). Black bars, initial conditions (0 h); grey bars, final conditions (84 h). Data are means  $\pm$  SE ( $n = 3$ ).



**Fig. S4.** Relative abundance of Pseudomonadota at the order level at the sediment surface across temperature treatments (initial, 10, 18, 26, 34 and 42 °C), under initial conditions and after 84 h. Stacked bars show mean proportions; only orders accounting for  $\geq 1\%$  of the community in at least one sample are shown individually, with the remainder pooled as 'NA'.



**Fig S5.** Differentially abundant predicted metabolic pathways (PICRUSt2) between initial (blue) and 42 °C (red) sediment microbial communities. Left: mean predicted pathway abundances; right: log<sub>2</sub> fold-changes with 90 % confidence intervals (dashed line at zero), with pathway identifiers and associated p-values. Bar and point colour indicates the condition in which each pathway was enriched.



**Figure S6.** Principal coordinates analysis (PCoA) of microbial community predicted MetaCyc metabolic pathways (PICRUST2) diversity at the sediment surface across temperature treatments. Each point is a replicate sample (n=3 per treatment) coloured by temperature — the initial community and the communities after 84 h of exposure at 10, 18, 26, 34 and 42 °C — and thin lines connect replicates to their group centroid. Axes PCo1 and PCo2 explain 26 % and 18 % of the total community variation, respectively.