**Supplementary material：**

Detailed calculation formulas for the potential DRs, DNRARs and NFRs.

(1) The formula for calculating the potential DR is as follows:

|  |  |
| --- | --- |
|  | (1) |
|  | (2) |

where DR (μmol N kg-1 h-1) is the potential rate of denitrification; D29 (μmol N kg-1 h-1) is the rate of 29N2 generation during denitrification; P30 (μmol N kg-1 h-1) is the total production rate of 30N2 measured in the slurry cultivation experiment; and Fn (%) represents 15N in NO3-, calculated by the concentration of 15NO3-added at the beginning of the slurry experiment and the concentration of residual NO3-measured at the end of the experiment.

(2) The calculation formula of the potential DNRAR is as follows:

|  |  |
| --- | --- |
|  | (3) |

where DNRAR (µmol N kg-1 h-1) is the potential rate of DNRA; Mi and Mf (μmol NL-1) are the initial and final concentrations of 15NH4+-N in the sample bottles, respectively; V (L) is the volume of the slurry; W (g) is the dry weight of the sediment in the sample bottle; and T (h) is the culture time.

(3) The formula for calculating the potential NFR is as follows:

|  |  |
| --- | --- |
|  | (3) |

where NFR (μmol N kg-1 h-1) is the nitrogen fixation rate; Ci and Cf are the initial and final concentrations, respectively, of 15N-labeled substances in the sample bottles (μmol NL-1); V (L) is the volume of cultivation slurry; W (g) is the dry weight of the sediment in the sample bottle; and T (h) is the culture time.

**Table S1.** Physical and chemical properties of water around roots.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Type of ditch** | **T-W (℃)** | **DO-W (mg/L)** | **pH-W** | **ORP-W (mV)** | **TN-W(mg/L)** | **NH4+-W(mg/L)** | **NO3--W(mg/L)** |
| A-Phr | 29.13±0.66ab | 5.88±0.68b | 7.57±0.51a | 206.36±11.25a | 1.58±0.06b | 0.69±0.06c | 0.66±0.13a |
| A-Typ | 27.80±1.08b | 7.50±0.51a | 7.53±0.37a | 207.95±9.24a | 1.66±0.08b | 0.76±0.07bc | 0.47±0.05b |
| N-Phr | 30.80±1.19a | 2.58±0.25c | 7.35±0.15a | 107.45±9.63c | 1.86±0.12a | 0.93±0.08b | 0.74±0.13a |
| N-Typ | 27.49±1.54b | 6.51±0.59ab | 7.50±0.34a | 152.97±18.04b | 2.03±0.14a | 1.11±0.16a | 0.63±0.05ab |

A-Phr, artificially cultivated ditch with *P. australis* rhizosphere sediment; A-Typ, artificially cultivated ditch with *T. orientalis* rhizosphere sediment; N-Phr, natural ditch with *P. australis* rhizosphere sediment; N-Typ, natural ditch with *T. orientalis* rhizosphere sediment.

T-W is the temperature in water; DO-W is the dissolved oxygen in water; pH-W is the pH value in water; ORP-W is the oxidation‒reduction potential in water; TN-W is the total nitrogen in water; NH4+-W is the ammonium nitrogen in water; and NO3-W is the nitrate nitrogen in water.

The different lowercase letters indicate significant differences among the different rhizosphere sediment samples (*p*<0.05) according to one-way ANOVA and LSD. The values are the means ± SDs (n=3).

**Table S2.** Physical and chemical properties of all sediment samples (n = 3).

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Type of ditch** | **EC****(****μs/cm)** | **TOC****(g/kg)** | **DOC****(g/kg)** | **TN****(g/kg)** | **NH4+-N****(mg/kg)** | **NO3--N****(mg/kg)** | **C/N** | **DOC/NO3---N** | **Sand****(wt, %)** | **Clay****(wt, %)** |
| A-Phr | 428±39b | 7.77±0.58a | 0.035±0.004a | 0.76±0.09b | 2.90±0.08a | 0.54±0.16b | 10.27±0.84a | 0.067±0.016a | 37±8ab | 28±13ab |
| A-Typ | 424±35b | 8.20±0.88a | 0.027±0.002b | 1.01±0.12a | 1.97±0.26b | 0.93±0.12a | 8.13±0.16b | 0.029±0.004b | 29±16b | 32±12a |
| N-Phr | 270±37c | 3.84±0.26b | 0.036±0.002a | 0.43±0.06c | 2.09±0.04b | 0.70±0.03b | 9.02±1.13ab | 0.052±0.004a | 58±14a | 11±4b |
| N-Typ | 518±19a | 8.08±0.64a | 0.033±0.002a | 0.95±0.08a | 1.35±0.18c | 1.03±0.13a | 8.57±1.34ab | 0.033±0.006b | 33±07b | 31±5a |

A-Phr, artificially cultivated ditch with *P. australis* rhizosphere sediment; A-Typ, artificially cultivated ditch with *T. orientalis* rhizosphere sediment; N-Phr, natural ditch with *P. australis* rhizosphere sediment; N-Typ, natural ditch with *T. orientalis* rhizosphere sediment.

EC is the electrical conductivity of the sediments, TOC is the total organic carbon in the sediments, DOC is the dissolved organic carbon in the sediments, TN is the total nitrogen in the sediments, NH4+-N is the ammonium nitrogen in the sediments, NO3--N is the nitrate nitrogen in the sediments, C/N is the ratio of organic carbon to total nitrogen in the sediments, DOC/NO3--N is the ratio of dissolved organic carbon to nitrate nitrogen in the sediments, Sand is the sand content (the sediment particle size fraction is 2~0.02 mm) in the sediments, and Clay is the clay content (the sediment particle size fraction is <0.002 mm) in the sediments.

**Table S3.** RDA single environmental factor difference significance (envfit test).

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Environmental factor** | **R2** | ***p*-value** |  | **Environmental factor** | **R2** | ***p*-value** |
| Alpha diversity index (Figure 5a) |  | The nitrogen cycling pathways (Figure 5d) |
| ORP-W | 0.734 | 0.007 |  | NH4+-N | 0.9199 | 0.001 |
| NH4+-N | 0.554 | 0.032 |  | DOC/NO3--N | 0.8692 | 0.001 |
| TOC | 0.544 | 0.033 |  | NO3--N | 0.7734 | 0.002 |
| EC | 0.487 | 0.051 |  | DOC | 0.744 | 0.003 |
| The phylum level of bacteria (Figure 5b) |  | ORP-W | 0.7308 | 0.008 |
| DOC | 0.6708 | 0.007 |  | DO-W | 0.5305 | 0.034 |
| TN | 0.6238 | 0.019 |  | Nitrogen conversion rates (Figure 5e) |
| TOC | 0.6206 | 0.012 |  | NO3--N | 0.8683 | 0.002 |
| Sand | 0.6018 | 0.011 |  | DOC/NO3--N | 0.7775 | 0.001 |
| DO-W | 0.5629 | 0.033 |  | nrfC | 0.7635 | 0.005 |
| EC | 0.5305 | 0.035 |  | nrfA | 0.7007 | 0.007 |
| Clay | 0.5291 | 0.037 |  | DOC | 0.6924 | 0.005 |
| The genus level of bacteria (Figure 5c) |  | NH4+-N | 0.6567 | 0.009 |
| NH4+-N | 0.953 | 0.001 |  | nosZ | 0.5993 | 0.018 |
| DOC | 0.7771 | 0.001 |  | norB | 0.5796 | 0.027 |
| DOC/NO3--N | 0.7724 | 0.001 |  | TN | 0.572 | 0.021 |
| NO3--N | 0.759 | 0.003 |  | nirK | 0.5613 | 0.031 |
| ORP-W | 0.6915 | 0.01 |  | TOC | 0.5364 | 0.026 |
| DO-W | 0.5958 | 0.019 |  |  |  |  |
| TN | 0.5779 | 0.022 |  |  |  |  |

**Table S4.** Pearson’s correlation coefficients between the relative abundances of major functional genes involved in nitrogen cycling pathways and environmental factors.

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Nitrogen cycling pathways** | **Gene name** | **DO-W** | **ORP-W** | **EC** | **TOC** | **DOC** | **TN** | **NH4+-N** | **NO3--N** | **DOC/NO3--N** |
| Denitrification | napA | 0.180 | 0.430 | 0.170 | 0.150 | 0.400 | -0.050 | 0.490 | -0.520 | 0.628\* |
| norB | -0.460 | 0.070 | -0.688\* | -0.480 | 0.250 | -0.597\* | 0.832\*\* | -0.823\*\* | 0.711\*\* |
| nirK | -0.760\*\* | -0.956\*\* | -0.400 | -0.708\*\* | 0.588\* | -0.626\* | -0.410 | 0.070 | 0.040 |
| nosZ | -0.858\*\* | -0.897\*\* | -0.560 | -0.831\*\* | 0.720\*\* | -0.781\*\* | -0.170 | -0.180 | 0.240 |
| nirS | -0.867\*\* | -0.784\*\* | -0.814\*\* | -0.919\*\* | 0.550 | -0.847\*\* | -0.080 | -0.230 | 0.150 |
| Dissimilatory nitrate reduction | nrfC | -0.440 | 0.080 | -0.440 | -0.320 | 0.380 | -0.520 | 0.857\*\* | -0.885\*\* | 0.876\*\* |
| nrfA | -0.550 | -0.100 | -0.360 | -0.350 | 0.662\* | -0.634\* | 0.782\*\* | -0.831\*\* | 0.886\*\* |
| Nitrogen fixation | nifD | 0.638\* | 0.825\*\* | 0.140 | 0.510 | -0.705\* | 0.520 | 0.350 | -0.030 | -0.150 |
| nifK | 0.637\* | 0.824\*\* | 0.140 | 0.510 | -0.713\*\* | 0.520 | 0.330 | 0.000 | -0.180 |
| nifH | 0.638\* | 0.816\*\* | 0.130 | 0.500 | -0.714\*\* | 0.510 | 0.320  | 0.000  | -0.180 |
| Nitrification | hao | -0.050 | 0.380 | -0.040 | 0.060 | 0.350 | -0.210 | 0.867\*\* | -0.790\*\* | 0.847\*\* |
| Anammox | hzsA | 0.080 | 0.490 | 0.050 | 0.240 | -0.110 | 0.030 | 0.858\*\* | -0.767\*\* | 0.753\*\* |
| hzsC | 0.080 | 0.500 | 0.130 | 0.250 | 0.240 | -0.050 | 0.836\*\* | -0.729\*\* | 0.789\*\* |

**Table S5.** Pearson’s correlation coefficient between critical nitrogen conversion rates, environmental factors and the relative abundance of genes involved in nitrogen cycling pathways.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Factor** | **Gene name** | **DR** | **DNRAR** | **NFR** |
| Denitrification pathways | 0.718\*\* | 0.380  | -0.705\* |
| Denitrification | napA | 0.540  | 0.691\* | -0.140  |
| norB | 0.743\*\* | 0.693\* | -0.050  |
| nirK | 0.320  | -0.020  | -0.725\*\* |
| nosZ | 0.570  | 0.240  | -0.725\*\* |
| nirS | 0.560  | 0.240  | -0.500  |
| Dissimilatory nitrate reductionpathways | 0.800\*\* | 0.751\*\* | -0.150  |
| Dissimilatory nitrate reduction | nrfC | 0.840\*\* | 0.824\*\* | -0.110  |
| nrfA | 0.862\*\* | 0.755\*\* | -0.200  |
| Nitrogen fixationpathways | -0.380  | -0.110 | 0.697\* |
| Nitrogen fixation | nifD | -0.370 | -0.090 | 0.701\* |
| nifK | -0.380 | -0.110 | 0.701\* |
| nifH | -0.380 | -0.120 | 0.683\* |
| DO-W |  | -0.693\* | -0.350 | 0.480 |
| ORP-W |  | -0.280 | 0.080 | 0.690\* |
| NO3--W |  | 0.729\*\* | 0.410 | -0.550 |
| EC |  | -0.610\* | -0.330 | 0.360 |
| TOC |  | -0.640\* | -0.340 | 0.631\* |
| DOC |  | 0.716\*\* | 0.510 | -0.694\* |
| TN |  | -0.792\*\* | -0.460 | 0.530 |
| NH4+-N |  | 0.589\* | 0.658\* | 0.290 |
| NO3--N |  | -0.813\*\* | -0.888\*\* | -0.020 |
| DOC/NO3--N |  | 0.809\*\* | 0.871\*\* | -0.140 |
| Sand |  | 0.644\* | 0.520 | -0.570 |
| Clay |  | -0.646\* | -0.500 | 0.460 |

****

**Figure S1.** Number of high-quality sequences (Effective-CCS (**a**)), the number of features (OUT Num (**b**)) and the number of reads corresponding to the feature (Seq num (**c**)) obtained through 16S rRNA sequencing.