Supplement Material for:

Suspended Particular Matter drives the spatial segregation of nitrogen turnover along the hyper-turbid Ems estuary

S1: Oxygen vertical profiles

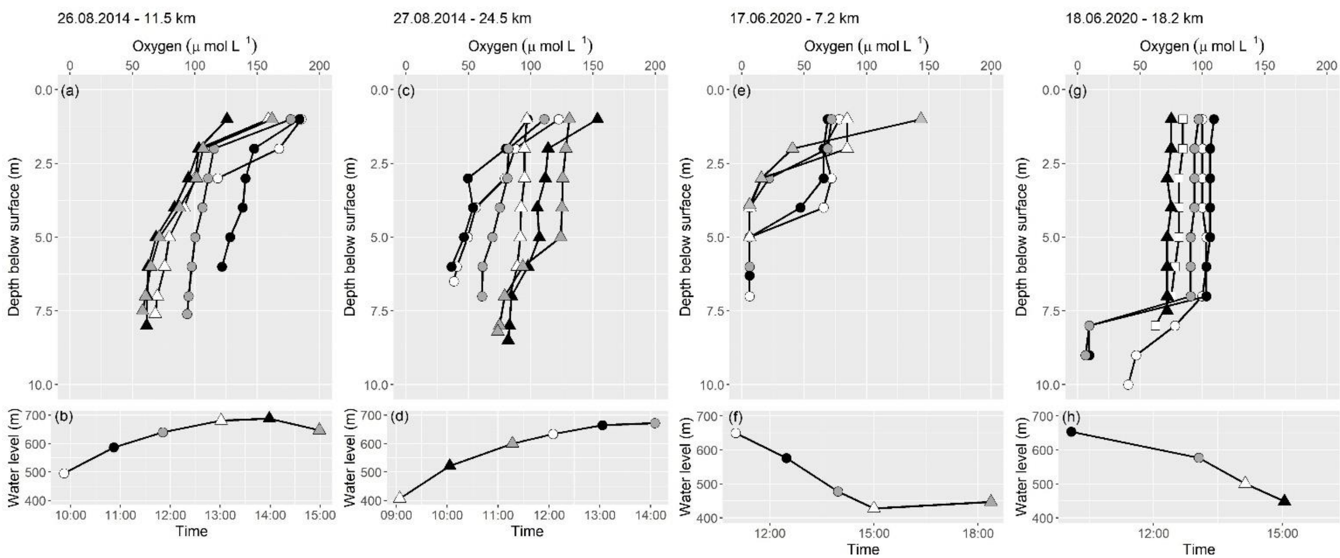

Figure S1: ***Vertical profiles of oxygen concentration in 2014 and 2020 at different stations along the Tidal River and Dollard Reach: a) vertical profile measured in 2014 at a station at stream kilometer 11.5, with b) the water levels over time at the station, c) vertical profile measured in 2014 at a station at stream kilometer 24.5, with d) the water levels over time at the station, e) vertical profile measured in 2020 at a station at stream kilometer 7.2, with f) the water levels over time at the station, g) vertical profile measured in 2020 at a station at stream kilometer 18.2, with h) the water levels over time at the station***

The lowest concentration in bottom water in 2014 was 58 µmol L^{-1} at stream km 11.5, and 36 µmol L^{-1} at km 24. In 2020, oxygen concentration was low in surface water at stream km 7.2, and anoxia occurred in bottom water over a full tidal cycle. Downstream, at km 18.2, O_2 concentration decreased 6 m below the surface, and anoxic conditions were reached at low water levels

S2: Plot of $\delta^{18}O\text{-}NO_3^-$ vs $\delta^{15}N\text{-}NO_3^-$ in the Tidal River of the Ems estuary

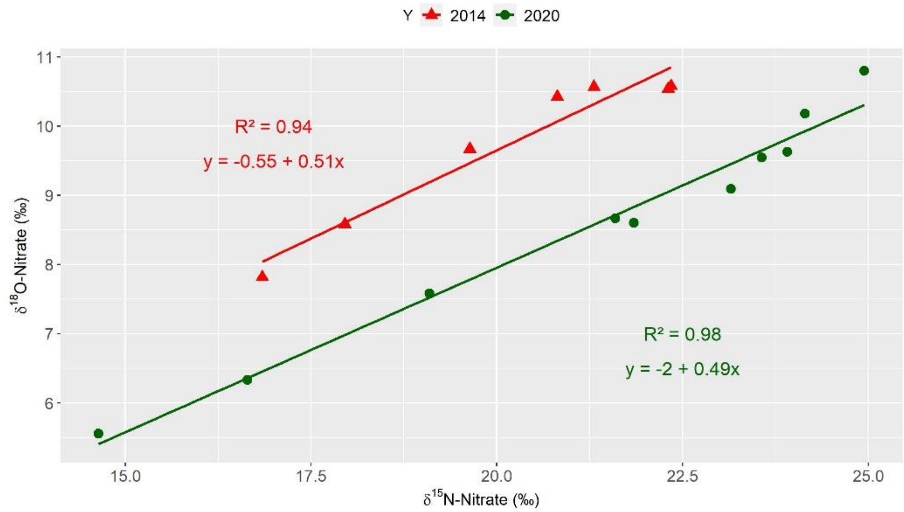

Figure S2: Plot of $\delta^{18}O\text{-}NO_3^-$ vs $\delta^{15}N\text{-}NO_3^-$ in the Tidal River of the Ems estuary in 2014 and 2020 with regression line, coefficient of determination and linear regression equation. Point shapes and colors mark stream kilometer of each sampling station.

S3: Open-system Mapping approach

S3.1 Explanation and exemplary application

We used a mapping approach inspired by Lewicka-Szczebak et al. (2017) using open-system approach for calculations of isotope effects. In an open-system approach the isotope effect can be described as the slope of the linear relationship between isotope values of nitrogen and fraction f (Chapter 2.8) (Sigman et al. 2009). The remaining fraction of substrate f is determined by $f = ([C]/[C_{initial}])$. We used our calculated mixing values (Chapter 2.7) as initial concentration and isotope values. In following, we explain our procedure in detail using a sample (ID: 702, km 41.50, 2020) as an example.

1. We calculated nitrate mixing concentration and the theoretical isotope value of samples following conservative mixing between two endmembers (Liss 1976). For our example we derived:

$$s = \frac{(32.31-18.64)}{(32.31-0)} = 0.423 \quad (1)$$

$$C_{Mix} = s \times C_R + (1-s)C_M = 0.423 \times 181.70\ \mu mol\ L^{-1} + (1-0.423) \times 3.60\ \mu mol\ L^{-1} = 78.94\ \mu mol\ L^{-1} \quad (2)$$

$$\delta_{Mix} = \frac{s \times C_R \times \delta_R + (1-s) \times C_M \times \delta_M}{C_{Mix}} = \delta_{Mix} = \frac{0.423 \times 181.70\ \mu mol\ L^{-1} \times 14.64\ ‰ + (1-0.423) \times 3.60\ \mu mol\ L^{-1} \times 14.23\ ‰}{78.94\ \mu mol\ L^{-1}} = 14.62\ ‰ \quad (3)$$

2. We use $\delta^{15}N\text{-}NO_3^-{}_{MIX}$ as initial isotope value and $c(NO_3^-)_{MIX}$. The remaining fraction for these values is f=1, since no nitrate is converted during pure mixing processes. Following figure is derived for our example:

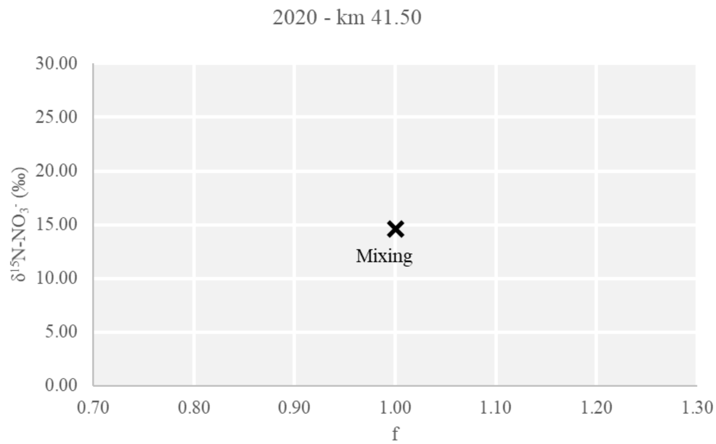

Figure S1: Calculates mixing $\delta^{15}N\text{-}NO_3^-$ versus the remaining fraction of nitrate for the example sample

3. We know that nitrogen isotopes ratios change along a specific isotope effect. We determined following relevant isotope effects for the Ems estuary: $^{15}\varepsilon_{DENIT}$ = -26 ‰, $^{15}\varepsilon_{REMIN}$ = -1.2 ‰. As long as ammonium concentration is low, we applied this $^{15}\varepsilon_{REMIN}$ value for nitrification with prior remineralisation. As soon as, ammonium and nitrite concentrations increased, we assume that remineralisation no longer determines the overall isotope effect of nitrification. We expect a combined influence of ammonium oxidation with an isotope effect $^{15}\varepsilon$ = -14 to -41 ‰ (Mariotti et al. 1981; Casciotti et al. 2003; Santoro and Casciotti 2011) and nitrite oxidation with $^{15}\varepsilon$ = +9 to +20 ‰ (Casciotti 2009; Buchwald and Casciotti 2010; Jacob et al. 2017) that leads to an assumed isotope effect of $^{15}\varepsilon_{NITRI}$ = -10 ‰ (Sanders unpublished data).

   In our example sample, ammonium and nitrite concentration already increased, so we used -10 ‰ as slope for our nitrification line and -26 ‰ for the denitrification line. Which leads to the following figure.

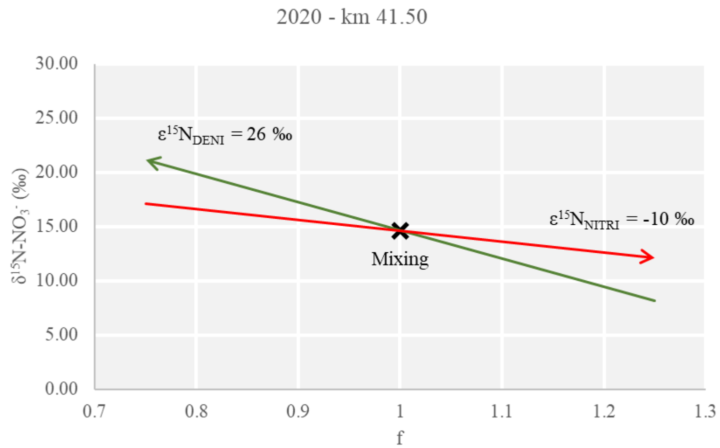

Figure S2: Calculated mixing $\delta^{15}N\text{-}NO_3^-$ versus the remaining fraction of nitrate for the example sample. The red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the mixing sample as intersect. The green lines indicates the isotope effect of denitrification with $^{15}{}_{DENI}$ = 26 ‰ with the mixing sample as intersect.

We can describe the linear equations for both processes ($EQ_{DENI,\ mix}$ and $EQ_{NITRI,mix}$) using the isotope effect as slope and the calculated mixing value as intersect. Leading to following equations:

$EQ_{DENI,mix:}$

$$\delta^{15}N_{mix,DENI} = m_{DENI} \times f + b_{DENI,mix} \rightarrow b_{DENI,mix} = \delta^{15}N_{mix,DENI} - m_{DENI} \times f = 14.64\ ‰ + 26 \times 1 = 40.64 \quad (4)$$

$EQ_{NITRI,,\ mix}$

$$\delta^{15}N_{mix,NITRI} = m_{NITRI} \times f + b_{NITRI,mix} \rightarrow b_{NITRI,mix} = \delta^{15}N_{mix,NITRI} - m_{NITRI} \times f = 14.64\ ‰ + 10 \times 1 = 24.64 \quad (5)$$

4. If only one of the processes occurred, the isotope value of our measured sample would lie on one of the regression lines. However, as the following plot shows, this is not the case for our example. The remaining fraction f of the sample is derived by $f = ([C]/[C_{initial}])$.

$$f = \frac{C_M}{C_{Mix}} = \frac{84.60\ \mu mol\ L^{-1}}{78.94\ \mu mol\ L^{-1}} = 1.07 \quad (6)$$

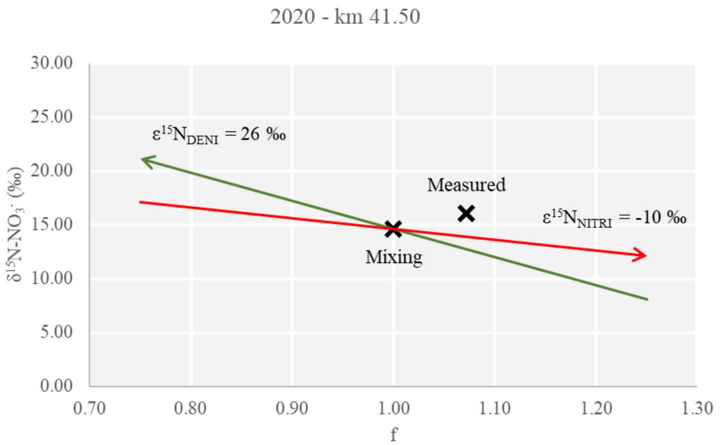

Figure S5: Calculated mixing $\delta^{15}N\text{-}NO_3^-{}_{mix}$ and measured $\delta^{15}N\text{-}NO_3^-{}_{sample}$ versus the remaining fraction of nitrate for the example sample. The red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the mixing sample as intersect. The green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the mixing sample as intersect.

5. After plotting the measured value, we calculate linear equations for each process using the measured values as the intersect ($EQ_{DENI,meas}$ and $EQ_{NITRI,meas}$). Leading to following results:

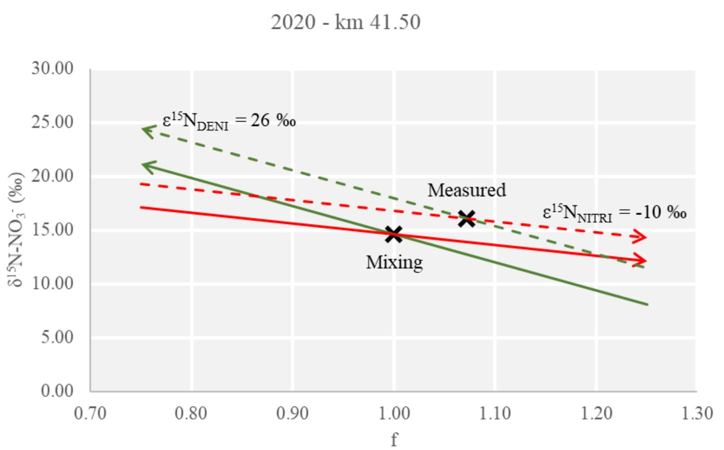

Figure S3: Calculated mixing $\delta^{15}N\text{-}NO_3^-{}_{mix}$ and measured $\delta^{15}N\text{-}NO_3^-{}_{sample}$ versus the remaining fraction of nitrate for the example sample. The red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the mixing sample as intersect. The green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the mixing sample as intersect. The dashed red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the measured sample as intersect. The dashed green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the measured sample as intersect.

$EQ_{DENI,meas:}$

$$\delta^{15}N_{meas,DENI} = m_{DENI} \times f + b_{DENI,meas} \rightarrow b_{DENI,meas} = \delta^{15}N_{meas,DENI} - m_{DENI} \times f = 16.09\ ‰ + 26 \times 1.07 = 43.95 \quad (7)$$

$EQ_{NITRI,\ meas}$

$$\delta^{15}N_{mix,NITRI} = m_{NITRI} \times f + b_{NITRI,meas} \rightarrow b_{NITRI,meas} = \delta^{15}N_{meas,NITRI} - m_{NITRI} \times f = 16.09\ ‰ + 10 \times 1.07 = 26.80 \quad (8)$$

6. We determine intersects of a linear equation derived by the mixing value as intersect and an equation derived by the measured sample to calculate the amount of nitrate processes via the different pathways. Therefore, two different approaches are possible for this purpose that lead to the same results.
   a. Calculating f_{DENI}: The intersection of $EQ_{DENI,mix}$ and $EQ_{NITRI,meas}$ leads to f_{DENI}. The difference between f and f_{DENI} results in f_{NITRI}. To calculate the amount of nitrate consumed/produced via each process, we use the deviation from the initial fractionation.

$f_{DENII:}$

$$\delta^{15}N_{DENI} = m_{DENI} \times f_{DENI} + b_{DENI,mix} = m_{NITRI} \times f_{DENI} + b_{NITRI,meas} \quad (9)$$

$$-26 \times f_{DENI} + 40.64 = -10 \times f_{DENI} + 26.80$$

$$f_{DENI} = \frac{40.64-26.80}{26-10} = 0.86$$

$f_{NITRI:}$

$$f_{NITRI} = f - f_{DENI} = 1.07 - 0.86 = 0.21 \quad (10)$$

$c(NO_3^-)_{DENI}$

$$C_{DENI} = (f_{ini} - f_{DENI}) \times C_{MIx} = (1 - 0.86) \times 78.94\ \mu mol\ L^{-1} = 10.73\ \mu mol\ L^{-1} \quad (11)$$

$c(NO_3^-)_{NITRI}$

$$C_{NITRI} = f_{NITRI} \times C_{MIx} = 0.21 \times 78.94\ \mu mol\ L^{-1} = 16.39\ \mu mol\ L^{-1} \quad (12)$$

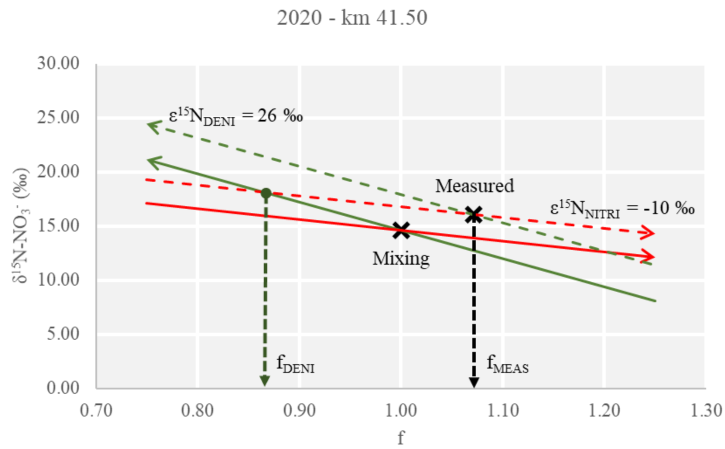

Figure S4: Calculated mixing $\delta^{15}N\text{-}NO_3^-{}_{mix}$ and measured $\delta^{15}N\text{-}NO_3^-{}_{sample}$ versus the remaining fraction of nitrate for the example sample. The red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the mixing sample as intersect. The green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the mixing sample as intersect. The dashed red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the measured sample as intersect. The dashed green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the measured sample as intersect.

b. Calculating f_{NITRI}: The intersection of $EQ_{REMIN,mix}$ and $EQ_{DENI,meas}$ leads to f_{NITRI}. The difference between f and f_{NITRI} results in f_{DENI}. To calculate the amount of nitrate consumed/produced via each process, we use the deviation from the initial fractionation.

$f_{NITRI:}$

$$\delta^{15}N_{NITRI} = m_{DENI} \times f_{NITRI} + b_{DENI,meas} = m_{NITRI} \times f_{NITRI} + b_{NITRI,mix} \quad (13)$$

$$-26 \times f_{NITRI} + 43.95 = -10 \times f_{NITRI} + 24.63$$

$$f_{NITRI} = \frac{43.95 - 24.63}{26 - 10} = 1.21$$

$f_{DENI:}$

$$f_{DENI} = f_{NITRI} - f = 1.21 - 1.07 = 0.14 \quad (14)$$

$c(NO_3^-)_{NITRI}$

$$C_{NITRI} = (f_{NITRI} - f_{ini}) \times C_{MIx} = (1.21 - 1) \times 78.94\ \mu mol\ L^{-1} = 16.39\ \mu mol\ L^{-1} \quad (15)$$

$c(NO_3^-)_{DENI}$

$$C_{DENI} = f_{DENI} \times C_{MIx} = 0.14 \times 78.94\ \mu mol\ L^{-1} = 10.73\ \mu mol\ L^{-1} \quad (16)$$

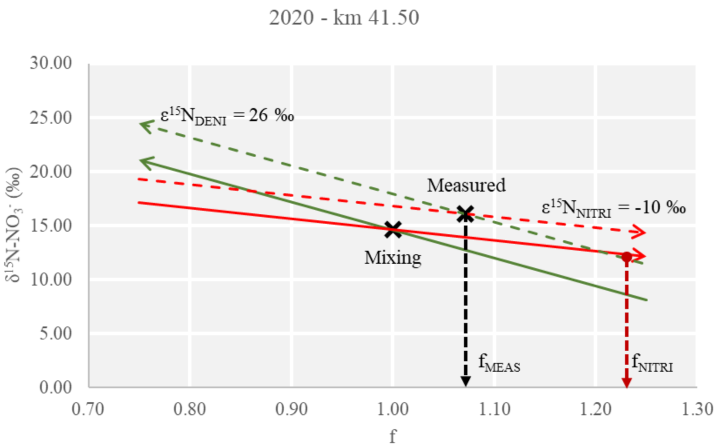

Figure S5: Calculated mixing $\delta^{15}N\text{-}NO_3^-{}_{mix}$ and measured $\delta^{15}N\text{-}NO_3^-{}_{sample}$ versus the remaining fraction of nitrate for the example sample. The red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the mixing sample as intersect. The green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the mixing sample as intersect. The dashed red line indicates the isotope effect of nitrification with $^{15}\varepsilon_{NITRI}$ = -10 ‰ with the measured sample as intersect. The dashed green lines indicates the isotope effect of denitrification with $^{15}\varepsilon_{DENI}$ = 26 ‰ with the measured sample as intersect.

S3.2 Results

Table 1: Measured and calculated mixing values for samples in zone 2 in 2014 used for the open-system mapping approach

2014	MEAS		MIX	
km	$c(NO_3^-)$ µmol L^{-1}	$\delta^{15}N\text{-}NO_3\text{-}$ ‰	$c(NO_3^-)$ µmol L^{-1}	$\delta^{15}N\text{-}NO_3^-$ ‰
26.20	151.00	20.81	167.32	16.83
30.73	161.00	19.64	172.84	16.84
66.18	31.10	14.67	23.86	16.04
77.62	21.30	13.56	17.17	15.68
87.53	23.30	14.25	18.40	15.77
88.80	15.10	12.70	13.16	15.29

Table 2: Results of the open-system mapping approach for samples in zone 2 in 2014

2014			DENI			NITRI			DENI		NITRI	
Km	F_{mix}	F_{Meas}	m	$b_{mix,}$	b_{meas}	m	$b_{mix,}$	b_{meas}	f	$c(NO_3^{-})$ µmol L^{-1}	f	$c(NO_3^{-})$ µmol L^{-1}
26.20	1.00	0.90	-26.00	42.83	44.28	-1.20	18.03	21.90	0.84	26.07	1.06	9.75
30.73	1.00	0.93	-26.00	42.84	43.85	-1.20	18.04	20.75	0.89	18.92	1.04	7.08
66.18	1.00	1.30	-26.00	42.04	48.56	-10.00	26.04	27.71	0.90	2.48	1.41	9.72
77.62	1.00	1.24	-26.00	41.68	45.81	-10.00	25.68	25.96	0.98	0.30	1.26	4.43
87.53	1.00	1.27	-26.00	41.77	47.18	-10.00	25.77	26.92	0.93	1.32	1.34	6.23
88.80	1.00	1.15	-26.00	41.29	42.54	-10.00	25.29	24.18	1.07	-0.91	1.08	1.03

Table 3: Measured and calculated mixing values for samples in zone 2 in 2020 used for the open-system mapping approach

2020	MEAS		MIX	
km	$c(NO_3^{-})$ µmol L^{-1}	$\delta^{15}N\text{-}NO_3^{-}$ ‰	$c(NO_3^{-})$ µmol L^{-1}	$\delta^{15}N\text{-}NO_3^{-}$ ‰
30.64	141.80	21.85	151.83	14.64
35.25	119.50	19.62	120.89	14.64
41.50	84.60	16.09	78.94	14.63
41.50	76.60	15.73	67.08	14.63
44.83	67.50	14.39	64.45	14.63
48.12	51.90	12.49	48.02	14.62
48.37	62.20	13.23	60.35	14.62
55.61	48.90	11.96	48.58	14.62

Table 4: Results of the open-system mapping approach for samples in zone 2 in 2020

2020			DENI			NITRI			DENI		NITRI	
Km	F_{mix}	F_{Meas}	m	$b_{mix,}$	b_{meas}	m	$b_{mix,}$	b_{meas}	f	$c(NO_3^{-})$ µmol L^{-1}	f	$c(NO_3^{-})$ µmol L^{-1}
30.64	1.00	0.93	-26.00	40.64	46.13	-10.00	24.64	31.18	0.59	62.11	1.34	52.08
35.25	1.00	0.99	-26.00	40.64	45.32	-10.00	24.64	29.50	0.70	36.76	1.29	35.36
41.50	1.00	1.07	-26.00	40.63	43.95	-10.00	24.63	26.80	0.86	10.727	1.208	16.387
41.50	1.00	1.14	-26.00	40.63	45.42	-10.00	24.63	27.15	0.84	10.56	1.30	20.08
44.83	1.00	1.05	-26.00	40.63	41.62	-10.00	24.63	24.86	0.99	0.96	1.06	4.02
48.12	1.00	1.08	-26.00	40.62	40.59	-10.00	24.62	23.30	1.08	-3.96	1.00	-0.09
48.37	1.00	1.03	-26.00	40.62	40.03	-10.00	24.62	23.54	1.07	-4.10	0.96	-2.25
55.61	1.00	1.01	-26.00	40.62	38.13	-10.00	24.62	22.03	1.16	-7.87	0.84	-7.55

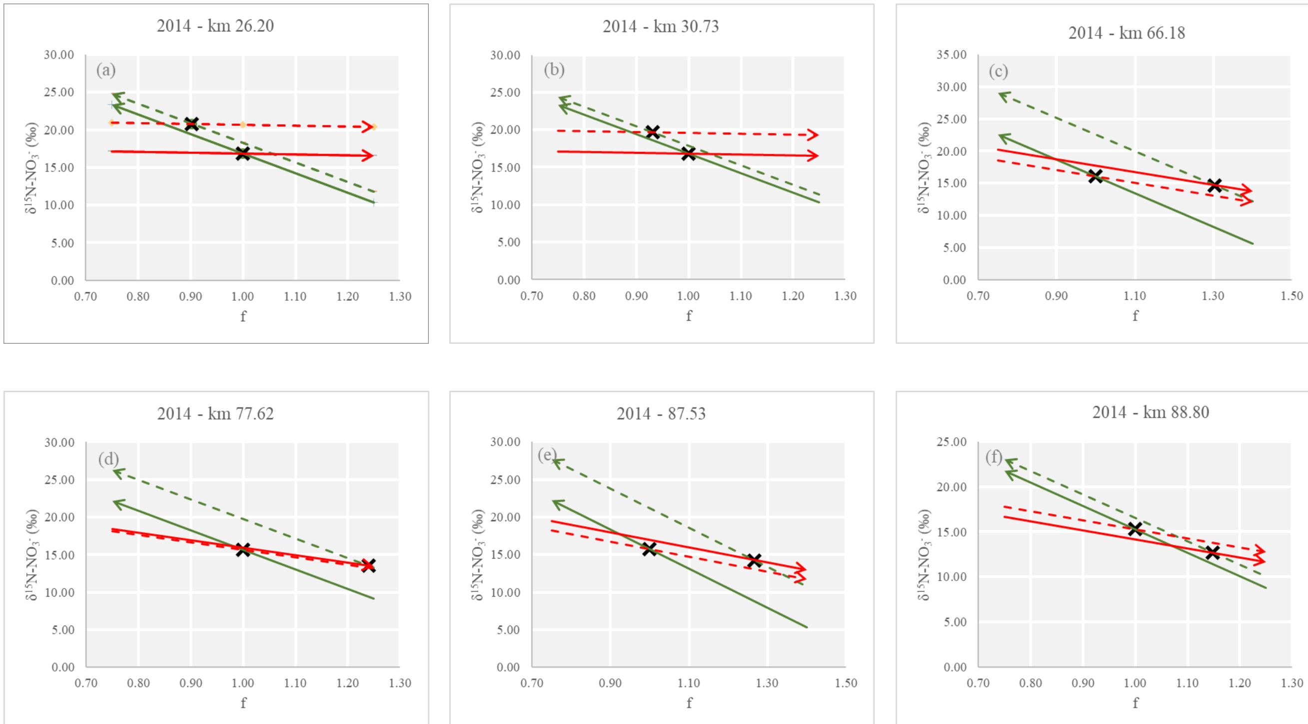

Figure S6: Plots derived by our open-system mapping approach for samples in 2014 (a) at stream kilometer 26.20, (b) at stream kilometer 30.73 (c) at stream kilometer 66.18, (d) at stream kilometer 77.62, (e) at stream kilometer 87.53, (f) at stream kilometer 88.80. Line color and arrow direction indicate the turnover-process. Green lines with arrows oriented upwards to the left are denitrification lines. Red lines with arrows oriented downwards to the right are nitrification lines. Straight lines indicate linear regression with mixing values as intercept and dashed lines linear regressions with the measured samples as intersect.

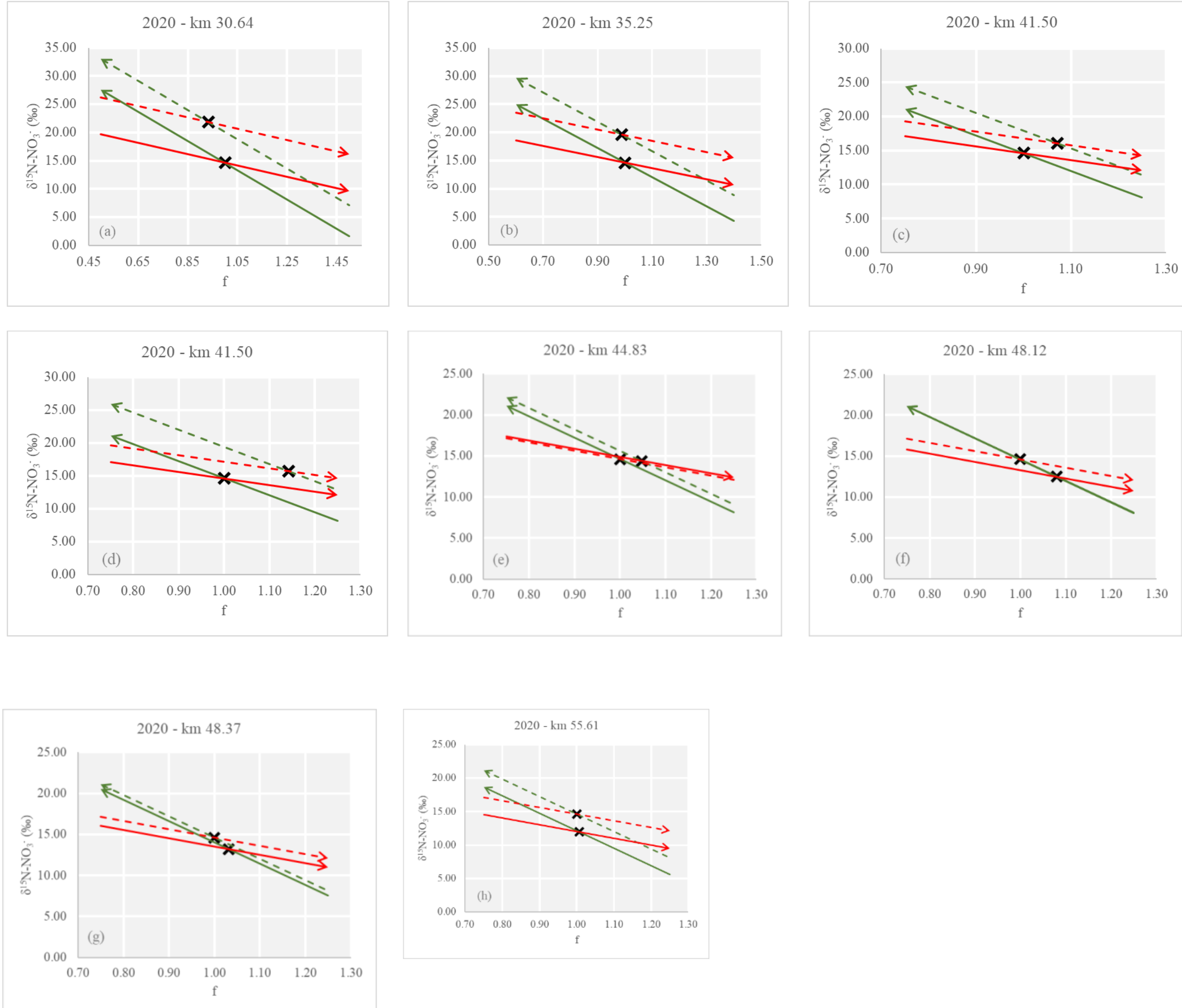

Figure S7: Plots derived by our open-system mapping approach for samples in 2020 (a) at stream kilometer 30.64, (b) at stream kilometer 35.25, (c) at stream kilometer 41.50, (d) at stream kilometer 41.50, (e) at stream kilometer 44.83, (f) at stream kilometer 48.12, (g) at stream kilometer 48.37, (h) at stream kilometer 55.61. Line color and arrow direction indicate the turnover-process. Green lines with arrows oriented upwards to the left are denitrification lines. Red lines with arrows oriented downwards to the right are nitrification lines. Straight lines indicate linear regression with mixing values as intercept and dashed lines linear regressions with the measured samples as intersect.

S4: Chlorophyll a concentration in the Outer Reaches

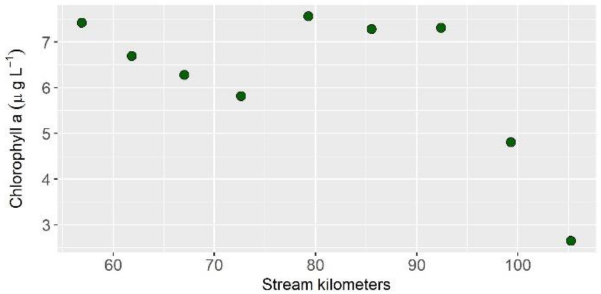

Figure S9: Chlorophyll a concentration in the Outer Reaches /zone 3 during our cruise in 2020. For chlorophyll a, water was filtered through combusted, pre-weighted GF/F Filters (4 h, 450 °C) and stored frozen for later analysis in our laboratory. The slight chlorophyll maximum supports our assumption of nitrate uptake due to ongoing primary production in the Outer Reaches.