

1 **Supplementary Material**

2 **1 Description of the model**

3 A zero-dimensional model describing the main diagenetic reactions affecting dissolved and
4 particulate nitrogen species in the sediment was constructed. The state variables, two derived
5 species and their units are presented in Table 1, a summary of the biochemical reactions is given
6 in Table 2, the model equations can be found in Table 3 and the parameters with their units are
7 listed in Table 4.

8 The biochemical equations were formulated as in the diagenetic model OMEXDIA (Soetaert et
9 al., 1996). The rates are proportional to the concentration of the dissolved or particulate species
10 taking part to the reaction; rate limiting terms are expressed by a Monod (hyperbolic) function;
11 inhibition terms are represented by a reciprocal hyperbolic function.

12 The model distinguishes between light and heavy nitrogen isotopes, denoted as ^{14}N and ^{15}N
13 respectively. It was assumed that isotope fractionation was insignificant, and the same values of
14 reaction rates, limitation and inhibition constants were used in the biogeochemical reactions
15 involving the two isotopic species.

16 OMEXDIA describes two fractions of organic matter which differ in degradability and C/N
17 ratio; a fast decaying fraction (see Table 1) with Redfield stoichiometry and a slow decaying
18 fraction with a higher C/N ratio of 7.5 (Soetaert et al., 1996). Due to the short time of the
19 experiment, the less labile fraction was not dynamically described here; rather a constant
20 degradation was assumed. The fast decaying fraction is dynamically modeled using a first order
21 degradation rate. The model includes three different mineralization pathways: oxic
22 mineralization (Tab 3.a, Eqs. 2 and 3), consuming oxygen (see Tab 2 and Tab 3), anoxic
23 mineralization (Tab 3.a, Eqs. 4 and 5), consuming oxidants other than oxygen and nitrate and
24 denitrification (Tab 3.a, Eqs. 6 and 7) using nitrate as the terminal electron acceptor.

25 As in OMEXDIA, all the organic matter oxidation pathways proceed at the same degradation
26 rate but unlike the original model formulation, the ammonium produced via oxic mineralization
27 is not directly oxidized to nitrate, so that competition between nitrification (Table 2 and Table
28 3.a, Eq.1) and ammonium uptake (Table 3.a, Eq. 8) can occur (Hochard et al., 2010).

29 Four different simulations were run to reproduce the different scenarios of the labeling study:
30 under dark and under light conditions, and for homogenized sediment spiked with, in the same
31 concentrations, $^{15}\text{N-NH}_4$ in combination with $^{14}\text{N-NO}_x$ (NO_3+NO_2), and $^{15}\text{N-NO}_x$ together with
32 $^{14}\text{N-NH}_4$ (see Materials and methods section for a complete description of the experimental set
33 up).

34 As bacterial and algal biomass is not explicitly modeled, it is assumed that nitrogen uptake
35 (Table 3a, Eqs. 8 and 10) is a function of the substrate concentration only. In order to clarify the
36 role of microphytobenthos, dark and light conditions were simulated using different expressions
37 for the microbial uptake rate. The initial value of nitrogen uptake rate was assumed to be the
38 same for both dark and light simulations but while it remains constant in the light, it tapers off
39 toward the end of the dark simulation (Table 3.a, Eq. 9).

40 Uptake of ^{15}N containing compounds produces labeled microbial biomass but, given the short
41 duration of the experiment, it is assumed that mortality of this biomass is negligible, thus there is
42 no production of ^{15}N -detritus. For this reason only the dead organic matter present at the
43 beginning of the experiment is mineralized, with production of $^{14}\text{N-NH}_4$ only.

44 $^{15}\text{N-NO}_x$ storage into diatoms cell, which selectively removes $^{15}\text{N-NO}_x$ from the dissolved pool,
45 is described by a simple first order reaction proportional to the difference between the internal
46 stored pool and the dissolved $^{15}\text{N-NO}_x$ (Table 3.a, Eq.11).

47 Ammonium adsorption to sediment particles (Table 3.a, Eq.12), and ammonium desorption
48 (Table 3.a, Eq.13), is assumed to affect only the new added $^{15}\text{N-NH}_4$, i.e. the adsorption-
49 desorption of unlabeled NH_4 is at equilibrium.

50 Anammox, the anaerobic formation of N_2 from NH_4 and NO_3 , is described by a first order
51 reaction proportional to dissolved NH_4 concentration, limited by nitrate and inhibited by oxygen
52 (Table 3.a, Eq. 14). Because of the presence of 15/14N ammonium and nitrate potentially
53 interacting with each other, anammox can lead to the formation of three species of molecular
54 nitrogen with a different isotopic composition: $^{28}\text{N}_2$, $^{29}\text{N}_2$ and $^{30}\text{N}_2$. Although only the production
55 of $^{29}\text{N}_2$ was measured, every possible anammox reaction that can potentially take place is
56 included in the model.

57 Because the data set was not detailed enough for the model to discriminate between the
58 contribution of anammox and denitrification to N_2 production, these two processes were lumped
59 together.

60 Dissimilatory nitrate reduction to ammonium, an anaerobic process inhibited by oxygen, is
61 modeled as a first order reaction proportional to the concentration of nitrate (Table 3.a, Eq. 15)

62 Oxygen is consumed by nitrification and oxic degradation of organic matter and is replenished
63 by diffusion at the sediment-water interface (Table 3.a, Eq. 16).

64 The concentrations of the dissolved species measured at the beginning of the study were used as
65 initial values for the state variables.

66 The model was implemented in the computing environment R (R Development Core Team,
67 2011), and solved with the ode function from R-package deSolve (Soetaert et al., 2010).

68 test

69 **2 Sensitivity analysis, parameters fitting and identifiability**

70 The R package FME (Soetaert and Petzoldt, 2009) was used to calibrate the model. The package
71 contains functions that, in a stepwise procedure allow fitting the model to data and assessing the
72 parameter uncertainty.

73 As a first step, model parameters were manually tuned until a good fit to the data was achieved.
74 From this set of parameters, first the ones to which the model is most sensitive to and which are
75 identifiable (i.e. can be uniquely estimated) were selected. The model was then fitted to the data
76 by finetuning the identifiable parameters, using the pseudo-random search algorithm of Price
77 (Price, 1977; Soetaert and Herman, 2009). Unidentifiable parameters (i.e. linearly correlated in a
78 way that an equal change in different directions has no effect on the model output) were kept at
79 the initial value assigned (Table 4).

80 Finally the parameter uncertainty was evaluated by a Markov Chain Monte Carlo (MCMC)
81 which generates a sample of the parameter probability density function (PDF, Soetaert and
82 Petzoldt 2009). The parameter PDF was then used to estimate the uncertainty on the model
83 variables. This was achieved running the model several times with a randomly chosen set of
84 parameters; the mean \pm standard deviation, and the minimum and the maximum value of model
85 variables at each time point are then represented as envelopes around the mean trajectory
86 (Soetaert and Petzoldt, 2009).

87

88 **References:**

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102

103 **Table 1a. State variables included in the model**

<u>NAME</u>	<u>UNITS</u>	<u>DESCRIPTION</u>
¹⁴ <i>NO_x</i>	μmol /slurry	Concentration of ¹⁴ N - nitrate+ nitrite
¹⁴ <i>NH</i> ₄	μmol /slurry	Concentration of ¹⁴ N – ammonium
¹⁴ <i>NDET_f</i>	μmol /slurry	Concentration of ¹⁴ N - fast decaying dead organic matter
¹⁵ <i>NOM</i>	μmol /slurry	Concentration of ¹⁵ N - organic nitrogen
¹⁵ <i>NO_x</i>	μmol /slurry	Concentration of ¹⁵ N - nitrate + nitrite
¹⁵ <i>NH</i> ₄	μmol /slurry	Concentration of ¹⁵ N - ammonium
¹⁵ <i>NH</i> _{4a}	μmol /slurry	Concentration of ¹⁵ N - adsorbed ammonium
¹⁵ <i>NO_{xs}</i>	μmol /slurry	Concentration of ¹⁵ N - internal stored nitrate
<i>N</i> ₂	μmol /slurry	Concentration of ²⁹ N - dinitrogen
<i>O</i> ₂	μmol /slurry	Concentration of oxygen

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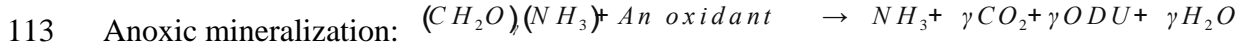
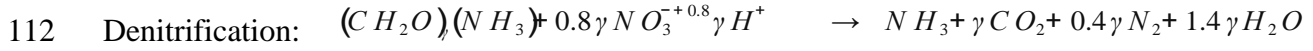
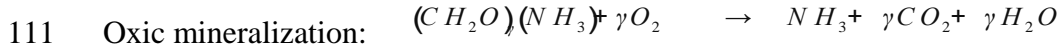
106 **Table 1b. Model derived species**

<u>NAME</u>	<u>UNITS</u>	<u>DESCRIPTION</u>
<i>NO_x</i>	μmol /slurry	Concentration of ¹⁴ N-NO _x + ¹⁵ N-NO _x
<i>NH</i> ₄	μmol /slurry	Concentration of ¹⁴ N-NH ₄ + ¹⁵ N-NH ₄

107

108 **Table 2. Chemical reactions included in the model**

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118 **Table 3.a Model equations**

119
$$Nit = R_{nit} * \frac{(O_2)}{(O_2) + K_{oxni}} * [NH_4] \quad (1)$$

120
$$NDETfmin = R_{denf} * \frac{[O_2]}{(O_2) + K_{oxmin}} * NDETf \quad (2)$$

121
$$NDETSmin = pdenf_s * R_{denf} * \frac{[O_2]}{(O_2) + K_{oxmin}} \quad (3)$$

122
$$NDETfamin = R_{denf} * \frac{K_{oxde}}{(O_2) + K_{oxde}} * \frac{K_{nitrde}}{[NO_x] + K_{nitrde}} * NDETf \quad (4)$$

123
$$NDETSamin = pdenf_s * R_{denf} * \frac{K_{oxde}}{(O_2) + K_{oxde}} * \frac{K_{nitrde}}{[NO_x] + K_{nitrde}} \quad (5)$$

124
$$NDETfden = R_{denf} * \frac{K_{oxde}}{(O_2) + K_{oxde}} * \frac{[NO_x]}{[NO_x] + K_{nitrde}} * NDETf \quad (6)$$

125
$$NDETSden = pdenf_s * R_{denf} * \frac{K_{oxde}}{(O_2) + K_{oxde}} * \frac{[NO_x]}{[NO_x] + K_{nitrde}} \quad (7)$$

126 (for i= light, dark)

$$127 \quad Ammu p = U p t a k e_i * \frac{[N H_4]}{[N H_4]^+ K_{amm}} \quad (8)$$

$$128 \quad U p t a k e_{i g h t}^{u p} = N \xi_w ; \quad U p t a k e_{d a r k} = (N_{u p} - N_{u p d k}) * \frac{0.1}{0.1 + t} + N_{u p d k} \quad (9)$$

$$129 \quad N i t u p = U p t a k e_i * \frac{[N O_x]}{[N O_x]^+ K_{nitr}} * \frac{K_{ammn}}{[N H_4]^+ K_{ammn}} \quad (10)$$

$$130 \quad N O_{xstor} = Rstor * (N O_x - N O_{xs}) \quad (11)$$

$$131 \quad N H_{4ads} = Rads * N H_4 \quad (12)$$

$$132 \quad N H_{4des} = Rdes * N H_{4a} \quad (13)$$

$$133 \quad Anmox = R_{anamox} * \frac{K_{oxax}}{(O_2)^+ K_{oxax}} * \frac{(N O_x)}{(N O_x)^+ K_{nitr}} * [N H_4] \quad (14)$$

134

$$135 \quad DNRA = R_{dnra} * \frac{K_{oxde}}{(O_2)^+ K_{oxde}} * [N O_x] \quad (15)$$

$$136 \quad OXrear = R O_2 * ([O_{2sat}] - [O_2]) \quad (16)$$

137

138 **Table 3.b Rate of change of the state variables**

$$139 \quad \frac{dNO_x}{dt} = Nit * \frac{[NH_4]}{[NH_4]} - (Nitup + DNRA + 0.8 * (NDETFden * CN_{rf} + NDETSden * CN_{rs}) + Anmox) * \frac{[NO_x]}{[NO_x]}$$

$$140 \quad \frac{dNH_4}{dt} = NDETFmin + NDETFamin + NDETFden + NDETSmin + NDETSamin + NDETSden - DNRA * \frac{[NO_x]}{[NO_x]} - (Nit + Ammup + Anmox) * \frac{[NH_4]}{[NH_4]}$$

$$141 \quad \frac{dNDETF}{dt} = - NDETFmin - NDETFamin - NDETFden$$

$$142 \quad \frac{dNOM}{dt} = Ammup * \frac{[NH_4]}{[NH_4]} + Nitup * \frac{[NO_x]}{[NO_x]}$$

$$143 \quad \frac{dNO_x}{dt} = Nit * \frac{[NH_4]}{[NH_4]} - NOxstor - (Nitup + DNRA + 0.8 * (NDETFden * CN_{rf} + NDETSden * CN_{rs}) + Anmox) * \frac{[NO_x]}{[NO_x]}$$

$$144 \quad \frac{dNH_4}{dt} = DNRA * \frac{[NO_x]}{[NO_x]} - (Nit + Ammup + Anmox) * \frac{[NH_4]}{[NH_4]} - NH_{4ads} + NH_{4des}$$

$$145 \quad \frac{dNH_{4a}}{dt} = NH_{4ads} - NH_{4des}$$

$$146 \quad \frac{dNO_{xs}}{dt} = NOxstor$$

$$147 \quad \frac{dN_2}{dt} = Anmox * \frac{[NH_4]}{[NH_4]} * \frac{[NO_x]}{[NO_x]} + Anmox * \frac{[NH_4]}{[NH_4]} * \frac{[NO_x]}{[NO_x]} + 0.8 * \frac{[NO_x]}{[NO_x]} * (NDETFden * CN_{rf} + NDETSden * CN_{rs})$$

148

$$149 \quad \frac{dO_2}{dt} = Oxrae - Nit * 2 - NDETFmin * CN_{rf} - NDETSmin * CN_{rs}$$

150

151 **Table 4. List of all the parameters and the reaction rates**

Name	Value	Description	Unit	References
CNrf	6.6	C:N ratio for fast decay detritus	dimensionless	[Soetaert et al, 1996a]
CNrs	7.5	C:N ratio for slow decay detritus	dimensionless	[Soetaert et al, 1996a]
RO ₂	0.001	O reaeration coefficient	day ⁻¹	See text
Rnit	0.8	Nitrification rate	day ⁻¹	Fitted
Rdenf	1	Mineralization rate fast decay detritus	day ⁻¹	See text
pdenf_s	1.22	Production rate slow versus fast decay detritus	dimensionless	Fitted
Nup	5.26	DIN uptake in light	μmol L ⁻¹ day ⁻¹	See text
Nupdk	3.83	DIN uptake in the dark	μmol L ⁻¹ day ⁻¹	Fitted
Koxni	5	Monod constant for O ₂ limitation in nitrification	μmol L ⁻¹	[Soetaert et al, 1996a]
Koxde	10	Monod ct for O ₂ inhibition of denitrification	μmol L ⁻¹	[Soetaert et al, 1996a]
Koxax	12.8	Monod ct for O ₂ inhibition of anammox	μmol L ⁻¹	Fitted
Koxmin	33.7	Monod ct for O ₂ limitation oxic mineralization	μmol L ⁻¹	Fitted
Kamm	7.7	Ammonium half saturation constant	μmol L ⁻¹	Fitted
Knitr	36.4	Nitrate half saturation constant	μmol L ⁻¹	Fitted
Knitrde	2 33.7*10	Monod ct for NO ₃ inhibition of anaerobic mineralization and limitation in denitrification	μmol L ⁻¹	Fitted
Kammin	1.87	Monod ct for NH ₄ inhibition of NO ₃ uptake	μmol L ⁻¹	Fitted
Rdnra	0.012	DNRA rate	day ⁻¹	Fitted
Ranamox	0.149	Anammox rate	day ⁻¹	Fitted
Rads	0.5	Ammonium adsorption coefficient	day ⁻¹	See text
Rdes	0.2	Ammonium desorption coefficient	day ⁻¹	See text
Rstor	0.35	Selective removal of 15Nitrate	day ⁻¹	Fitted