Nitrogen removal in coastal sediments of the German Wadden Sea

Astrid Deek · Kay Emeis · Justus van Beusekom

Abstract Although sediments of the German Wadden Sea are suspected to eliminate a considerable share of nitrate delivered to the SE North Sea, their denitrification rates have not been systematically assessed. We determined N_2 production rates over seasonal cycles (February 2009–April 2010) at two locations with two sediments types each, the first site (Meldorf Bight) receiving nitrate during all seasons from the Elbe river plume, and a second site on the island of Sylt, where nitrate is depleted during summer months. In sediments from the Sylt site, N_2 production ranged from 15 to 32 μmol N_2 m^{-2} h^{-1} in the fine sand station and from 7 to 13 μmol N_2 m^{-2} h^{-1} in the coarse sand station; N_2 production was not detected when nitrate was depleted in May and July of 2009. N_2 production in the Meldorf Bight sediments were consistently detected at higher rates (58–130 μmol N_2 m^{-2} h^{-1} in the very fine sand station and between 14 and 30 μmol N_2 m^{-2} h^{-1} in the medium sand station). Analysis of ancillary parameters suggests that major factors controlling N_2 production in coastal sediments of the German Wadden Sea are the nitrate concentrations in the overlying water, the ambient temperature, and the organic matter content of the sediment. Extrapolating our spot measurements to the zone of nitrate availability and sediment types, we estimate an annual nitrogen removal rate around 16 kt N year^{-1} for the entire northern sector of the German Wadden Sea area. This corresponds to 14% of the annual Elbe river nitrogen load.

Keywords Denitrification · Nitrogen · Sediments · Wadden Sea · German Bight · Incubation

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Introduction

Reactive nitrogen (rN) inputs to the North Sea by rivers have increased 3-fold since the 1960's (van Beusekom and de Jonge 2002) and are considered to be a prime reason for environmental deterioration in offshore regions of the North Sea (Owens et al. 1990) and in the Wadden Sea area (van Beusekom et al. 2001; van Beusekom and de Jonge 2002). Mass balance estimates (Pätsch et al. 2010) suggest that increased atmospheric and riverine rN inputs are mitigated by effective sinks, such as sedimentary

denitrification in subtidal and intertidal sediments of the shallow North Sea. These sediments receive constantly high organic matter loadings (van Beusekom and de Jonge 2002) and are permeable, so that nitrate is readily advected to suboxic depth levels. These suboxic sediment layers thus are active sites of denitrification, during which NO_3^- and NO_2^- are reduced via NO and N_2O to N_2. The latter escapes from the sediment to the overlying water or atmosphere by diffusion (Nixon et al. 1996) and is lost from the system. Together with the process of anaerobic ammonia oxidation, which is less well studied, denitrification in sediments is the most important sink for nitrate on a global scale and a first order regulation mechanism for the nitrogen cycle of shelf seas (Middelburg et al. 1996; Middelburg and Soetaert 2004). Controlling factors for denitrification rates in coastal sediments are amount and quality of sedimentary organic matter, concentrations of nitrate in water overlying the sediment, permeability of the sediment, and secondary factors, such as bioturbation/ bio-irrigation, macrophytes and availability of manganese and iron (Cornwell et al. 1999). Although of considerable value as an ecosystem service that eliminates excess nitrate in and counteracts eutrophication of coastal zones, knowledge of denitrification rates in the sediments of the North Sea is limited. Experimental data were obtained in the 1990's with methods (the acetylene blocking technique) that may underestimate actual denitrification rates (Sørensen 1978; Lohse et al. 1993; Lohse et al. 1996). Budget estimates generally give higher N loss rates than available measurements (Brion et al. 2004). van Beusekom and van de Jonge (1998) estimated annual denitrification rates of about 900 mmol N m^{-2} $year^{-1}$ for the Ems estuary based on nutrient-salinity relations and input data. Hydes et al. (1999) regarded the Southern North Sea as a giant estuary and estimated an average denitrification in the Southern North Sea of about 256 mmol N m^{-2} $year^{-1}$. Brion et al. (2004) used similar rates for their North Sea budget. Only limited information using modern techniques like the N_2/Ar method is available to support the high denitrification rates as suggested by budget calculations. Two studies carried out within the Wadden Sea focussed on experimental techniques and reported potential denitrification rates (after addition of ^{15}N labelled nitrate) (Cook et al. 2006; Gao et al. 2010). These rates were among the highest ever measured in

the Wadden Sea, comparable to the above rates as derived from budgets, but lack spatial and temporal resolution.

Our main objective here was to improve the seasonal and spatial resolution of sedimentary N_2 production measurements as an indication of nitrogen removal in intertidal sediments of the Wadden Sea. Our approach was to determine nitrogen removal rates along gradients of nitrate availability, sediment type and organic matter content, and ambient temperature. By extrapolation of determined N_2 production rates, we estimated the overall nitrogen loss in the entire Wadden Sea.

Materials and methods

We quantified N_2 production rates using a direct flux approach by incubating undisturbed sediment cores in a flow-through incubation set-up (modified after Lavrentyev et al. 2000; McCarthy and Gardner 2003) and measuring N_2 fluxes by the N_2/Ar technique (Kana et al. 1994; An et al. 2001). The incubation design allowed us to quantify net N_2 fluxes in sediment cores incubated with ambient site water, and to calculate denitrification rates according to the isotope pairing technique in sediment cores incubated with site water enriched by $^{15}NO_3^-$. We conducted sampling campaigns in different seasons and at sites with varying sediment types to bracket the most significant gradients in environmental parameters listed in the introduction (Cornwell et al. 1999).

Study sites

In the northern Wadden Sea of the German Bight, nitrate gradients are steep and mirror the decreasing influence of the Elbe River plume. We selected two study sites along the nitrate gradient: Near the plume, at study site Büsum/Meldorf Bight (Fig. 1), nitrate is never depleted in waters in contact with sediments. The site near the island of Sylt has no or negligible nitrate and low ammonium concentrations between July and September (van Beusekom et al. 2009). At each site, two different sediment types in terms of grain size distribution and organic matter content were sampled (Table 1).

Fig. 1 Study sites in the North Frisian Wadden Sea
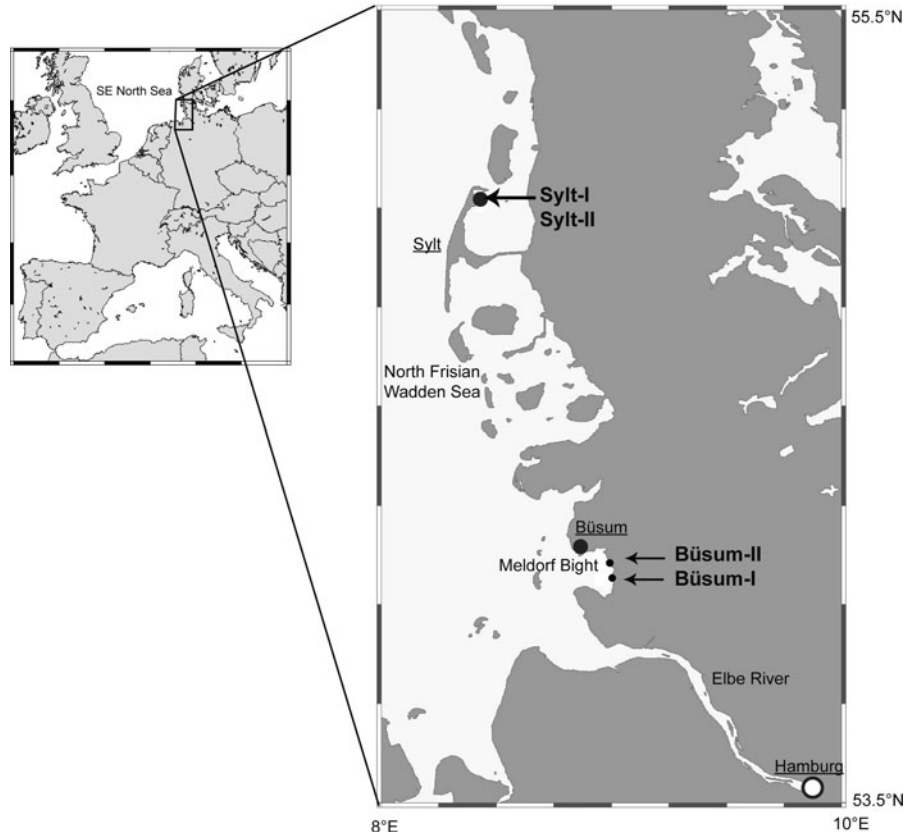
Table 1 Sediment characteristics for the study sites Sylt-I, Sylt-II, Büsum-I and Büsum-II (Porosity, carbon content (%C), nitrogen content (%N), organic carbon content (%C$_{org}$) and C/N given as average value with standard deviation for 10 sediment slices from one core of 10 cm length (depth resolution: 1 cm); amino acids (AA) were analyzed in surface sediment layers: 0–1 cm; median grain size given for 10 cm; sediments were classified according the Udden-Wenthworth size scale (Wentworth 1922; Pettijohn and Potter 1972)

Month	Station	Porosity (v/v)	C (%)	N (%)	C$_{org}$ (%)	C/N	AA (mg g^{-1})	Median grain size (μm)	Sediment classification
June'09	Büsum-I	0.67 ± 0.13	2 9 ± 0.4	0 22 ± 0 05	1 9 ± 0 4	9 5 ± 0 3	7	63	Very fine sand
	Büsum-ll	0.42 ± 0,02	0.5 ± 0.1	0.1 ± 0.04	0.1 ± 0.1	5.9 ± 0.4	0.6	250	Medium sand
July'09	Sylt-I	0.73 ± 0.06	2.1 ± 0.8	0.21 ± 0.12	1.6 ± 0.6	7.5 ± 0.7	14.9	125	Fine sand
	Sylt-II	0.42 ± 0.09	0.7 ± 0.4	0.05 ± 0.06	0.6 ± 0.4	13.7 ± 0.4	0.8	500	Coarse sand

Sampling

At least six sediment cores were taken during each sampling campaign at each of the sampling stations of the two sites. These stations were sampled at the beginning of high tide, when the sediment was covered by a water column of at least 10 cm and a maximum of 30 cm. Samples (around 20 cm length) were taken manually with polyvinylchloride (PVC) tubes (60 cm length and inner diameter of 10 cm) and stoppered. After sampling, the upper rubber stopper was removed and water level overlying the sediment was adjusted to 10 cm. Sediment cores were cooled and transported (<6 h) to the temperature-controlled

Table 2 Site parameters for the sampling campaigns in Sylt and Büsum

Station	Month	Water column		pH	PO_4^- (µmol/l)	Si (µmol/l)	NH_4^+ (µmol/l)	NO_3^- (µmol/l)	NO_2^- (µmol/l)	Sediment	
		T (°C)	Sal							Oxygen penetration depth (mm)	
Büsum										Büsum-I	Büsum-ll
	February'09	4	18	8.6	6.0	18.0	23.7	67.4	1.5	1.5	2.6
	June'09	23	30	8.0	2.6	18.9	14.2	6.8	1.3	0.8	2.2
	November'09	8	20	8.0	5.2	87.3	31.0	67.2	4.2	2.6	3.0
	April'10	9	30	8.5	n.d.	20.6	1.4	85.4	4.6	1.4	1.6
Sylt										Sylt-I	Sylt-ll
	February'09	1	30	8.2	2.3	10.4	4.7	31.9	n.d.	2.2	3.1
	May'09	15	30	–	1.2	7.8	n.d.	n.d.	n.d.	1.2	1.6
	July'09	24	32	8.3	0.6	3.8	1.3	n.d.	n.d.	1.2	1.7
	November'09	8	30	8.8	1.3	10.8	10.3	12.1	0.8	1.8	–
	April'10	8	30	8.5	0.2	1.7	4.8	13.0	1.3	1.2	2.0

(*n.d.* not detected; – data not available)

laboratory. At least 100 l of site water were collected in 25 l carboys to ensure sufficient site water with appropriate salinity and nutrient concentrations for the flow-through incubation.

At the sample site, 50 ml of site water were filtered through a 0.2 µm syringe filter (X50 Analypore, PVDF) and stored frozen until analysis of ambient nutrient concentrations as described in "Nutrient concentrations" section. O_2 saturation and temperature of site water were measured with an Ecoscan DO 6 (Eutech Instruments), salinity with a handheld refractometer (Atego), and pH with an Ecoscan Ion 6 (Eutech Instruments) (Table 2).

Sediment core incubation

After transferring the cores to a temperature controlled room or into a circulating water bath adjusted to the in situ temperature, the supernatant water was aerated with aquarium pumps for 6–12 h to reach oxygen saturation in the overlying water column before starting the incubation. The incubation set-up is a modified and larger version of the incubation set-ups described in Gardner et al. (1991), Lavrentyev et al. (2000) and McCarthy and Gardner (2003). The sediment cores were wrapped with aluminium foil to prevent variable light conditions, and were placed in

a flow-through incubation system, consisting of the cores, a 25 l carboy of site water, a peristaltic pump and an outflow collection vessel. The site water reservoir was aerated constantly, because the ambient water at the study sites was always oxygen saturated during all sampling campaigns. The sediment cores were locked with a gas tight plunger with two O-rings and Tygon inflow and outflow tubes. The plunger was positioned to 5 cm above the sediment–water interface, which is equal to a water volume of 785 ml over the sediment, continuously replaced by site water at a rate of 1–2 ml min^{-1}. Samples of inflow water and outflow water were taken every 12 h of incubation for N_2 and O_2 measurements and every 24 h of incubation for nutrient analysis. Two control cores without sediment were run in the same way to exclude water column denitrification. Whereas the incubations of Sylt-I and Büsum-I cores were run for 2 days, the incubations of Sylt-II and Büsum-II cores were run for 4 days. The different incubation times were necessary to account for the different sediment types: In preliminary incubation tests, sandy sediments had reached steady-state fluxes of nutrients, O_2 and N_2 only after longer incubation times, explained by deeper oxygen penetration (Table 2) and deeper denitrification zones in sandy sediments (Cook et al. 2006; Gihring et al. 2010).

Core incubations with site water (unamended cores)

Three of the 6 subcores were incubated with untreated site water. From the unamended cores, actual net N_2 fluxes (m28) and O_2 fluxes were measured under actual conditions with ambient nitrate concentrations at the sampling site (Table 2). Rate determinations were conducted as described in "Determination of actual net N_2 and O_2 fluxes" section.

Core incubation with site water enriched with $^{15}NO_3^-$ (amended cores)

Three of the 6 subcores were incubated with site water enriched with $Na^{15}NO_3^-$ (98 atom % ^{15}N, Isotec). Whereas natural nitrate concentration of site water varied with sampling campaign as documented in Table 2, labeling of $^{15}NO_3^-$ was always to a 40–50 μmol/l $^{15}NO_3^-$ concentration, resulting in varying relative nitrate enrichment over natural conditions.

N_2 fluxes (m29 + m30) from these amended cores were used (a) to estimate N_2 fluxes under conditions when actual NO_3^- limitation is alleviated through artificial $^{15}NO_3^-$ addition, and (b) to calculate denitrification rates according to the isotope pairing technique (Nielsen 1992), as described in "Calculation of N_2 production rates from amended cores according to IPT" section.

Laboratory analyses and rate determinations

Triplicate samples from the inflow water and outflow water of unamended and amended cores were taken every 12 h with cylindrical glass vials during incubations and were analyzed immediately after collection for N_2 and O_2. The concentrations of N_2 and O_2 were calculated from N_2/Ar and O_2/Ar ratios measured with a membrane inlet mass spectrometry (MIMS) system consisting of a Quadrupole mass spectrometer (QMS) (GAM 200, InProcess Instruments) with a modified membrane inlet from Bay Instruments. The MIMS was standardized with air saturated temperature-salinity-standards according to Kana et al. (1994) to correct measured N_2/Ar and O_2/Ar ratios to actual ratios. Concentrations were calculated by multiplying the actual Ar concentration from solubility tables (Weiss 1970) with the corrected ratio. The QMS ion source produces O^+ ions by ionizing sample gas; O^+ ions may react with N_2 forming NO_x (Eyre et al.

2002). This so called NO_x-effect results in higher signals on mass 28 (m28) at low oxygen concentrations and low m28 signals at high oxygen concentrations, which may lead to a misinterpretation of actual denitrification. The NO_x-effect was tested for the MIMS system used here and the extent of this effect was found to be below the standard deviation of the method (0.1%) (J. Pohlmann, pers. comm. 2010).

N_2 and O_2 fluxes (fl; in μmol m^{-2} h^{-1}) were calculated by subtracting inflow concentration (C_i) from outflow concentration (C_o) multiplied with the flow rate (f: 1 h^{-1}) and sediment surface area (a: m^2) ratio:

$$fl = (C_o - C_i) \times f/a. \tag{1}$$

Determination of actual net N_2 and O_2 fluxes

Net N_2 and O_2 fluxes were determined from the water samples of the unamended cores by MIMS measurements of N_2 on mass 28 and O_2 on mass 32, and inserting respective inflow and outflow concentration data into Eq. 1. In the following, net N_2 and O_2 fluxes are also referred to as actual fluxes.

Calculation of N_2 production rates from amended cores according to IPT

From the water samples of the amended cores N_2 fluxes were determined by MIMS measurements on mass 29 and mass 30. Production rates of $^{29}N_2$ and $^{30}N_2$ were calculated according to Eq. 1.

The obtained $^{29}N_2$ and $^{30}N_2$ production rates allow estimates of N_2 production based on (natural) $^{14}NO_3^-$ (D_{14}) and (added) $^{15}NO_3^-$ (D_{15}) to be made with equations used in the isotope pairing technique (IPT) (Nielsen 1992). D_{15} is calculated as follows:

$$D_{15} = 2 \times N_2(m30) + N_2(m29), \tag{2}$$

where N_2 (m30) is the net N_2 production rate on m30 and N_2 (m29) is the net N_2 production rate on m29.

For the calculation of the denitrification rate based on ambient $^{14}NO_3^-$ (D_{14}) Eq. 3 applies:

$$D_{14} = [N_2(m29)/(2 \times N_2(m30))] \times D_{15}. \tag{3}$$

The total denitrification rate is:

$$D_{tot} = D_{14} + D_{15}. \tag{4}$$

We also estimated the proportion of coupled nitrification–denitrification (D_n) as opposed to

denitrification of nitrate from the overlying water (D_w) (Nielsen 1992; Steingruber et al. 2001):

$$D_w[\%] = (D_{15}/\varepsilon) \times 100/D_{tot}, \qquad (5)$$

where ε represents the relative $^{15}NO_3^-$ enrichment during the core incubation.

From Eqs. 4 and 5, coupled nitrification–denitrification can be estimated:

$$D_n[\%] = 100 - D_W. \qquad (6)$$

The parallel measurement of net N_2 production rates measured in unamended cores and the calculation of N_2 production rates (D_{14}) from amended cores of the same site furthermore allowed estimation of N_2 production and consumption rates based on calculations proposed by An et al. (2001).

Nutrient concentrations

During each sampling campaign, a sample of 50 mL site water was collected to determine ambient nutrient concentrations of inflow water at the beginning of flow-through incubation. Every 24 h during incubation, a 50 ml sample from each inflow and outflow of unamended and amended cores was collected for nutrient analyses. Water samples were filtered using a 0.2 μm syringe filter (X50 Analypore, PVDF) and were stored frozen in Falcon tubes until analysis.

Nutrient concentrations were determined photometrically on an automated continuous flow system (Bran & Luebbe Auto Analyzer 3) using standard methods of seawater analysis: nitrate and nitrite concentrations were determined according to Armstrong et al. 1967), ammonium concentrations according to Grasshoff et al. (1983), phosphate concentrations according to Murphy and Riley (1962), and silicate concentrations according to Brewer and Riley (1966). The relative error of triplicate sample measurements was below 1.5% for nitrate, nitrite and phosphate concentrations, and below 4.8% for ammonium concentrations.

Nutrient fluxes were calculated according to Eq. 1. In the following, nutrient fluxes in the unamended cores are also referred to as actual fluxes.

Sediment characteristics

Before incubation, oxygen penetration depths were determined for one sediment core from each sampling station. Triplicate profiles were performed at three different positions in each sediment core (a total of nine profiles); the precision of this determination was better than 0.5 mm. Two different types of oxygen profiling setups were used during this study: A Clark-type O_2 micro electrode (OX 50 and OX 100) in connection with a picoamperemeter (Unisense A/S), and an oxygen micro-optode (Oxy 50 M, Pyro Science) in connection with a Microx TX3-USB (Pyro Science, Presens).

Sediment characteristics were determined on sediment cores from sampling campaigns in June 2009 (Büsum-I and Büsum-II) and in July 2009 (Sylt-I and Sylt-II). For each sampling site, a sediment core with a length of 10 cm was sliced into 1 cm slices of known volume and frozen for transport.

Water content and porosity were calculated from the weight before and after freeze drying and assuming a grain density of 2.65 g cm^{-3}. Grain size distribution was then determined on dry residues by sieving through mesh sizes of 1000, 500, 250, 125 and 63 μm. The weight percentage of total nitrogen (TN) and organic carbon (C_{org}) were determined with an Elemental Analyzer (Thermo Flash EA) calibrated against acetanilide on dried and homogenised subsamples of unsieved sediments (in 1 cm slices). The standard deviation of duplicate analyses was below 0.05% for carbon and below 0.005% for nitrogen.

Total hydrolysable amino acids (AA) were analysed in surface sediments (0–1 cm) with a Biochrom 30 AA Analyser after hydrolysis of 30–50 mg of sediment with 6 N HCl for 22 h at 110°C. HCl was then removed by evaporation and the residue was taken up by acidic buffer and injected into the analyser. The relative error for total AA was 4%.

Results

Sediment characteristics at the different sites

Properties of sediments at each of the sample locations are listed in Table 1. These parameters were only determined once on one core from each sediment facies (see sections "Study sites", "Sediment characteristics"). The highest organic carbon content together with highest total hydrolysable AA concentrations were measured in cores of Sylt-I and

Büsum-I, which suggests that substrate quality in terms of labile particulate matter available for bacterial respiration was highest at these stations.

Seasonal variation of the site parameters

Stations in the Meldorf Bight (Büsum-I and Büsum-II) and on Sylt (Sylt-I and Sylt-II) experienced a pronounced seasonal variability of site parameters (Table 2). High water temperatures as measured during summer months (May–July) were accompanied by comparatively low nutrient concentrations in the water column. At Sylt sites, NO_3^-, NO_2^- and NH_4^+ concentrations were even below detection limit in May 2009. The oxygen penetration depth of the sediments varied similarly with season, lowest oxygen penetration depths were measured at high water temperatures during summer months.

Nutrient fluxes

Nutrient fluxes across the sediment–water interface of incubated cores were determined every 24 h of incubation. For simplification, nutrient fluxes are only given for the steady state. Table 3 depicts NO_3^- and NH_4^+ fluxes (mean value of three cores) for the unamended cores on day 2 of incubation for Büsum-I and Sylt-I and on day 4 of incubation for Büsum-II and Sylt-II (all at steady state). In the following, NO_3^- and NH_4^+ fluxes from the unamended cores are referred to as actual fluxes. In Table 4, NO_3^- and NH_4^+ fluxes from $^{15}NO_3^-$ amended cores are listed for the same situation (with negative values denoting flux into the sediment).

Highest ammonium fluxes were measured in the Büsum-I and Sylt-I cores which both have a relatively high organic carbon content (Table 1).

Highest actual NO_3^- fluxes were measured in the Büsum-I and Büsum-II cores in June 2009 and April 2010 (Table 3). In the Sylt cores, no significant actual nitrate fluxes were measured during May and July 2009 (Table 3) when water temperature was highest, but no ambient nitrate was left in the water column (Table 1), whereas significant NO_3^- fluxes in the $^{15}NO_3^-$ amended incubations were measured during summer months at the Sylt sites (Table 4).

Table 3 Nitrate and ammonium fluxes for the unamended cores

Station	Month	NO_3^- flux	NH_4^+ flux
		(μmol m^{-2} h^{-1})	
Büsum-I	February'09	-255 ± 28	-150 ± 12
	June'09	-412 ± 10	203 ± 30
	November'09	-71 ± 2	135 ± 40
	April'10	-298 ± 70	32 ± 5
Büsum-II	February'09	-62 ± 9	-78 ± 7
	June'09	-197 ± 3	-101 ± 15
	November'09	-54 ± 10	-178 ± 30
	April'10	-246 ± 67	26 ± 8
Sylt-I	February'09	-24 ± 9	88 ± 23
	May'09	n.d.	335 ± 104
	July'09	n. d.	515 ± 36
	November'09	-48 ± 5	487 ± 78
	April'10	-19 ± 11	35 ± 15
Sylt-II	February'09	-23 ± 4	-10 ± 24
	May'09	n.d.	83 ± 3
	July'09	n.d.	0 ± 69
	November'09	-184 ± 26	4 ± 8
	April'10	-16 ± 4	52 ± 30

Data are mean values of three cores ($n = 3$) with standard deviation

n.d. not detected

N_2 and O_2 fluxes

N_2 and O_2 fluxes (mean value of three cores) are given in Tables 5 and 6; again fluxes refer to day 2 of incubation for Büsum-I and Sylt-I and to day 4 of incubation for Büsum-II and Sylt-II (all at steady state).

Sedimentary oxygen demand (SOD) was highest in November 2009 at all sampling stations and lowest in February 2009 (Table 5).

Tables 5 and 6 also compare four different N_2 fluxes obtained simultaneously by the incubation of six cores: (1) Actual net N_2 fluxes on mass 28 from three unamended cores (Table 5), incubated as described in "Core incubations with site water (unamended cores)". (2) and (3) N_2 fluxes on mass 29 and mass 30 from three amended cores (Table 6), in which ambient $^{14}NO_3^-$ is partially combined with added $^{15}NO_3^-$ as described in "Core incubation with site water enriched with $^{15}NO_3^-$ (amended cores)" section. (4) the N_2 production rate (D_{14}) calculated

Table 4 Nitrate and ammonium fluxes for the $^{15}NO_3^-$ amended cores

Station	Month	NO_3^- flux (μmol m^{-2} h^{-1})	NH_4^+ flux
Büsum-I	February'09	−214 ± 73	−60 ± 26
	June'09	−763 ± 309	537 ± 168
	November'09	−163 ± 9	182 ± 4
	April'10	−177 ± 56	33 ± 5
Büsum-II	February'09	−90 ± 16	87 ± 3
	June'09	−258 ± 15	−56 ± 12
	November'09	−141 ± 3	71 ± 15
	April'10	−	−
Sylt-I	February'09	−64 ± 12	107 ± 14
	May'09	−130 ± 37	278 ± 93
	July'09	−219 ± 33	490 ± 132
	November'09	−228 ± 95	275 ± 49
	April'10	−52 ± 2	22 ± 10
Sylt-II	February'09	−44 ± 6	−24 ± 6
	May'09	−102 ± 37	67 ± 22
	July'09	−55 ± 20	69 ± 10
	November'09	−41 ± 60	−5 ± 4
	April'10	−74 ± 20	31 ± 20

Data are mean values of three cores ($n = 3$) with standard deviation

– data not available

Table 5 N_2 (m28) and O_2 fluxes for the unamended cores

Station	Month	N_2 (m28) flux (μmol m^{-2} h^{-1})	O_2 flux
Büsum-I	February'09	120 ± 30	−823 ± 106
	June'09	70 ± 10	−2165 ± 15
	November'09	130 ± 37	−2430 ± 217
	April'10	58 ± 20	−1360 ± 9
Büsum-ll	February'09	30 ± 8	−220 ± 30
	June'09	30 ± 5	−876 ± 288
	November'09	14 ± 8	−1265 ± 182
	April'10	29 ± 8	−564 ± 238
Sylt-I	February'09	32 ± 10	−981 ± 214
	May'09	n.d.	−904 ± 110
	July'09	n.d.	−1048 ± 135
	November'09	15 ± 4	−2044 ± 337
	April'10	16 ± 7	−1392 ± 255
Sylt-ll	February'09	13 ± 3	−228 ± 47
	May'09	n.d.	−892 ± 70
	July'09	n.d.	−645 ± 1
	November'09	3 ± 2	−1070 ± 137
	April'10	7 ± 2	−474 ± 127

Data are mean values of three cores ($n = 3$) with standard deviation

n.d. not detected

according the isotope pairing technique (IPT) (Nielsen 1992) from the N_2 fluxes (m29 + m30) of the $^{15}NO_3^-$ amended cores (Table 6) as described in "Calculation of N_2 production rates from amended cores according to IPT" section. The differences between calculated N_2 production rates on mass 28 (D_{14}) and actual N_2 production rates on mass 28 are small and within the standard deviation for the respective mean N_2 production rates (Tables 5, 6). Exceptions are fluxes during a sampling campaign at Sylt-II in February 2009, when fluxes calculated by IPT were higher (Table 6).

Actual N_2 fluxes (m28) were highest in Büsum-I cores (130 μmol N_2 m^{-2} h^{-1}) in November 2009, and varied between 15 and 30 μmol m^{-2} h^{-1} in Büsum-II cores. We registered no actual N_2 flux (m28) during May and July 2009 in Sylt-I and Sylt-II sediments; these sediments produced N_2 at rates of 32 and 13 μmol N_2 m^{-2} h^{-1}, respectively, in February 2009. In all sediment cores, N_2 fluxes in the $^{15}NO_3^-$ amended cores were higher than actual rates during

summer, as indicated by maximum rates on m29 and m30 in Büsum-I (m30: 176 μmol N_2 m^{-2} h^{-1}; m29: 67 μmol N_2 m^{-2} h^{-1}) in June 2009. Büsum-II yielded 69 μmol N_2 (m30) m^{-2} h^{-1} and 32 μmol N_2 (m29) m^{-2} h^{-1} during the same sampling campaign (Table 6). Maximum N_2 fluxes in the $^{15}NO_3^-$ amended cores were 104 μmol N_2 (m30) and 13 μmol N_2 (m29) m^{-2} h^{-1} in Sylt-I cores in May 2009. Highest N_2 fluxes of 29 μmol N_2 (m30) m^{-2} h^{-1} were measured in Sylt-II cores in May 2009, whereas fluxes on m29 were not observed (Table 6).

Discussion

Our data bracket a range of environmental conditions and several salient features emerge on controls of N_2 fluxes out of Wadden Sea sediments. We focus our discussion on effects of nitrate concentrations and temperature, and further evaluate the influence of sedimentary C_{org} content on N_2 production rates. But the experimental data are also suited to differentiate

Table 6 N_2 (m29 + m30) fluxes and calculated N_2 fluxes (m28) according to IPT (D_{14}) for the $^{15}NO_3^-$ amended cores

Station	Month	N_2 (m29) flux ($\mu mol\ m^{-2}\ h^{-1}$)	N_2 (m30) flux	N_2 flux (IPT: D_{14})
Busum-I	February'09	33 ± 12	6.2 ± 2.4	120 ± 43
	June'09	67 ± 4	176 ± 7.3	79 ± 6
	November'09	42 ± 14	9.8 ± 9	132 ± 7
	April'10	19 ± 3	5.8 ± 0.8	52 ± 10
Busum-ll	February'09	12 ± 4	2.3 ± 1.6	46 ± 8
	June'09	32 ± 2	69 ± 0.3	38 ± 4
	November'09	4 ± 1	1.8 ± 0.7	9 ± 2
	April'10	7 ± 4	1.4 ± 0.5	26 ± 18
Sylt-I	February'09	11 ± 2	6 ± 0.8	20 ± 5
	May'09	13 ± 6	104 ± 33	14 ± 7
	July'09	1 ± 1	46 ± 12	1 ± 1
	November'09	20 ± 5	35 ± 13	26 ± 6
	April'10	10 ± 2	13 ± 6	14 ± 6
Sylt-ll	February'09	11 ± 2	1.5 ± 0.1	47 ± 19
	May'09	n. d.	29 ± 1.8	–
	July'09	n. d.	16 ± 0.3	–
	November'09	2 ± 1	2.4 ± 0.3	2 ± 1
	April'10	4 ± 2	6.2 ± 2.2	6 ± 4

Data are mean values of three cores ($n = 3$) with standard deviation

n.d. not detected

between different processes in the sediments at each individual location and facies. This is because the actual N_2 fluxes (m28) measured in unamended sediment cores quantify net N_2 production rates; whereas the mass distribution of N_2 produced in $^{15}NO_3^-$ amended cores permit application of concepts developed for the IPT. Together with observed nutrient fluxes, the data give insight into and help to quantify individual processes of the nitrogen cycle. In the first part of our discussion, we estimate the proportion of alternative pathways of N_2 production and N_2 consumption, such as nitrification–denitrification and N_2 fixation that contribute to the net N_2 flux. Because the sediment types analysed represent the dominant sedimentary facies of the Wadden Sea, we finally estimate annual nitrogen removal rates by sediments in the northern German Wadden Sea.

Insights into N-cycling from nutrient fluxes and denitrification rates obtained by IPT

The calculated N_2 fluxes on mass 28 (D_{14}) obtained by IPT are in good agreement with the actual N_2 fluxes (m28) measured in unamended cores (Tables 5, 6), indicating that both methods yield comparable results. Moreover, the simultaneous measurement of actual

N_2 (m28) and nutrient fluxes, together with N_2 fluxes from $^{15}NO_3^-$ amended cores (m29 + m30) allows us to estimate the amount of coupled nitrification–denitrification in the sediment, and the amount of N_2 fixation.

According to An et al. (2001), N_2 production rates on mass 28 (D_{14}) estimated from IPT match actual N_2 fluxes (m28) when no N_2 fixation occurs. We applied equations derived by An et al. (2001) to calculate the amount of N_2 fixation, and obtained negative solutions, which denote zero N_2 fixation rates. N_2 fixation thus plays no or only a minor role in sediments under study. In consequence, the net N_2 flux (m28) measured in the unamended cores is a good approximation of the gross N_2 production, because N_2 consumption can be neglected. In our approach, we do not differentiate between the two different mechanisms of N_2 production (anammox and denitrification). Anammox has been shown to play an important role in removing rN from marine ecosystems (Thamdrup and Dalsgaard 2002; Trimmer and Nicholls 2009), but within this study its contribution to the total N_2 production cannot be estimated. This is because our experimental set-up did not include incubations with increasing concentrations of $^{15}NO_3^-$ concentrations as described in Risgaard Petersen

et al. (2003). In the following, we thus defined the gross denitrification rate as the net N_2 flux (m28) or net N_2 production rate (m28) that includes an unknown proportion of anammox.

Because our experiments did not include incubations with different $^{15}NO_3^-$ concentrations, requirements of the classical IPT (Nielsen 1992; Steingruber et al. 2001) have not been met. But by applying the IPT equations and evaluating the N_2 fluxes on m29 and m30 in the $^{15}NO_3^-$ amended cores (Table 6), we can calculate the contribution of coupled nitrification–denitrification to the total N_2 production (Nielsen 1992) as described in "Calculation of N_2 production rates from amended cores according to IPT" section. The calculations suggest that between 5 and 60% of N_2 flux out of Büsum and Sylt cores derives from coupled nitrification/denitrification. That significant nitrate additions are generated by nitrification corresponds to N_2 fluxes observed exclusively on m29 in the Sylt-I cores in May 2009 (Table 6), when ambient $^{14}NO_3^-$ was not available (Table 2). Fluxes on m29 are only generated when $^{15}NO_3^-$ is mixed with $^{14}NO_3^-$ before being reduced to $^{29}N_2$. The only possible source of $^{14}NO_3^-$ to the sediment in the Sylt area in May 2009 is nitrification within the sediment, and at rates calculated to around 15 µmol m^{-2} h^{-1}. The ammonium fluxes (up to 500 µmol m^{-2} h^{-1}, Table 3) from Sylt-I and Büsum-I cores to oxic supernatant water suggest even higher nitrification rates, because it is estimated that 27–36% of the actual ammonium efflux may be re-oxidised to nitrate within the sediment (Sweerts et al. 1991).

The very high ammonium fluxes in the Sylt-I and Büsum-I cores coincide with the highest AA concentrations in the host sediments (Table 1), which apparently support extraordinary rates of ammonification. But dissimilatory nitrate reduction to ammonium (DNRA) may also be responsible for the high ammonium fluxes in Sylt-I and Büsum-I cores (Table 3) during summer: At that time, high nitrate fluxes (around 400 µmol m^{-2} h^{-1}) into the unamended Büsum-I sediment cores (Table 3) resulted in comparatively small N_2 fluxes (m28) of 70 µmol m^{-2} h^{-1} out of the sediment (Table 5). This is good evidence that DNRA is a process that may compete with denitrification for nitrate as a substrate, but it is not a sink for reactive nitrogen as are denitrification and anammox. Up to date, DNRA has not been quantified for sediments of our study site, but

it has been previously shown that carbon-rich environments (Tiedje 1987; Christensen et al. 2000) and a low redox potential of the sediments favour DNRA over denitrification (Burgin and Hamilton 2007; Gardner and McCarthy 2009). Scott et al. (2008) reported high DNRA rates in sediments related to a high SOD. Coincident with a relatively high SOD in the Sylt-I and Büsum-I sediments (Table 5) and high organic matter content (Table 1), we observed a dark sediment colour in Sylt-I and Büsum-I sediments which is a visual indication for a low redox potential that may stimulate DNRA. Apparently, a significant share of nitrate may be reduced by DNRA within these sediments and is subsequently released back into the water column as ammonium (An and Gardner 2002; Giblin et al. 2010).

Compared to the high fluxes in Sylt I and Büsum I sediments, ammonium and nitrate fluxes in Sylt-II and Büsum-II cores were much smaller (Table 3), and indicated lower overall N-cycling rates; in these sediments, O_2 penetration also was much deeper. Although an extended gradient of O_2 in the Sylt-II and Büsum-II cores should favour complete nitrification of effluxing ammonium, substrate limitation and a low rate of ammonification in the organic-poor sediments (Table 1) limits nitrification: According to IPT calculations, coupled nitrification–denitrification in Sylt-II cores accounts for around 10% in November 2009 and April 2010, whereas no denitrification has been detected in summer (Table 5). It is of note that we never registered a net flux of nitrate out of the sediments under study, which means that nitrification never adds more nitrate than is being immediately reduced again. Furthermore, nitrogen consumption by N_2 fixation is a negligible process (see above) and net N_2 fluxes are always directed out of the sediments, meaning that most of the sediments are a sink for reactive nitrogen. Exceptions are the Sylt-I sediments with ammonium fluxes of up to 500 µmol m^{-2} h^{-1} that exceed the nitrogen consumption rate most time of the year (Table 3). Hence, Sylt-I sediments are not a net sink, but a source of reactive nitrogen.

Nitrate and temperature effects on N_2 production rates in Büsum and Sylt sediments

N_2 production rates at both locations were lowest in summer (Table 5), despite of higher temperatures favouring most microbial processes (Koch et al.
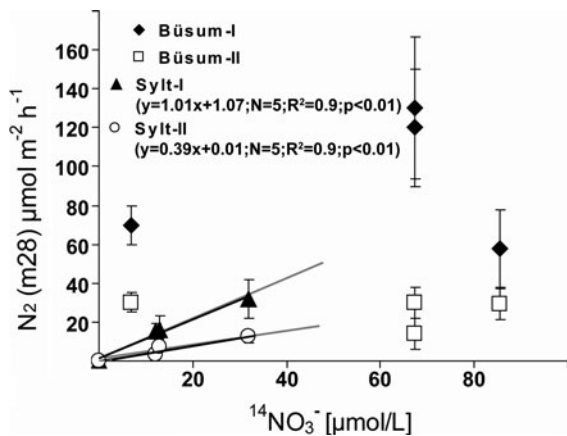
Fig. 2 Actual N_2 production rates versus ambient $^{14}NO_3^-$

1992). On the other hand, nitrate was abundant in overlying waters at these times. This observation and the fact that we did not measure actual N_2 (m28) production in sediments at Sylt sites when NO_3^- was depleted in summer identify nitrate availability as one of the overriding control factors on denitrification rates. Even in the small range of ambient NO_3^- concentrations encountered at Sylt-I and Sylt-II sampling sites (Fig. 2), actual N_2 (m28) production appears to increase linearly with nitrate availability, which here is relatively low (<30 μmol l^{-1}) and restricted to the cold seasons (Table 2). The linear positive relationships between actual N_2 production (m28) and nitrate concentrations for the four data points for Sylt I and II sampling in Fig. 2 are significantly correlated for each sampling site ($r^2 \geq 0.9$; $P < 0.01$ for Sylt-I and Sylt-II), but our data lack true seasonal resolution and doe not cover maximum NO_3^- concentrations of about 50 μmol l^{-1} (van Beusekom et al. 2009). NO_3^- availability is a major limiting factor in denitrification (Nedwell 1982; Seitzinger and Nixon 1985; Christensen et al. 1990), when other factors, such as organic matter or temperature, are not limiting (Golterman 2004).

In the $^{15}NO_3^-$ amended incubations of Sylt cores, the N_2 fluxes (m29 + m30) had maxima in summer months (Table 6), emphasizing that Sylt sediments are highly active when NO_3^- substrate is available. In response to decreasing riverine nitrogen loads, NO_3^- concentrations have steadily decreased during summer months over the last years in the Sylt area (van Beusekom et al. 2009), and nitrate availability was found to limit denitrification in previous investigations

(Jensen et al. 1996). We conclude that denitrification (fuelled by nitrate from the overlying water column) in the nitrate-limited sector of the Wadden Sea occurs only between November and April (fall, winter and early spring) at rates between 3 and 13 μmol N_2 m^{-2} h^{-1} in coarse sands (Sylt-II), and between 15 and 32 μmol N_2 m^{-2} h^{-1} in fine sands (Sylt-I).

In contrast to the Sylt stations, the Büsum stations are situated near the Elbe river plume and thus permanently influenced by river-borne nitrate, which is never completely depleted (Table 2), nitrate reduction in sediments proceeds during all seasons. Still, N_2 fluxes (m29 + m30) in the $^{15}NO_3^-$ amended cores were higher than actual N_2 (m28) fluxes in unamended cores in June 2009 (Table 5, 6), indicating that ambient nitrate concentrations around 7 μmol l^{-1} (Table 2) already limit actual N_2 production during summer months. In contrast to the Sylt stations, there is no firm relationship between maximum NO_3^- concentrations and maximum actual N_2 production (Fig. 2): Maximum nitrate concentrations occurred in April 2010, but were not associated with maximum actual N_2 (m28) production rates, which were instead measured in November 2009 (Table 5). One explanation is that temperature may control N_2 production in the absence of NO_3^- limitation: Although water temperature was similar during sampling campaigns in November 2009 and April 2010, lower rates in April 2010 followed an exceptionally long and cold winter during this study, during which the sampling site was covered with ice and mean air temperatures of −1.3°C chilled the coast until March (DWD 2010).

The effect of temperature on N_2 production is apparent from the results of the $^{15}NO_3^-$ amended cores. Because all $^{15}NO_3^-$ amended cores received 40–50 μmol l^{-1} of $^{15}NO_3^-$, substrate limitation can be excluded. During sampling campaigns with temperatures above 15°C, N_2 (m29 + m30) fluxes in $^{15}NO_3^-$ amended cores were highest (Table 6), indicating the positive effect of temperature on denitrification rates as shown elsewhere (Seitzinger 1988; van Luijn et al. 1999). But the effect observed in our study may be influenced by additional factors, such as oxygen penetration depth, nitrate availability, primary productivity and hence substrate availability, which are all linked with temperature. Competing heterotrophic processes for organic matter, especially sulphate reduction and aerobic respiration, may also

bias denitrification rates, so that isolated consideration of temperature only does not reflect interrelated in situ conditions. Sørensen et al. (1979), for example, observed that denitrification rates were low at times of high temperature, but attributed this to low nitrate concentrations co-occurring with higher temperatures. Because the Büsum-II and Sylt-II cores generally have lower denitrification rates than the Büsum-I and Sylt-I cores, even at high nitrate levels in the $^{15}NO_3^-$ amended cores at in situ temperatures above 15°C, we assume that other factors besides substrate availability and temperature may be rate-determining including organic matter content or the permeability-dependent transport of nitrate to the active site of denitrification in the sediment.

Influence of sedimentary TOC content

Maximum actual N_2 fluxes (m28) for the four study sites ranged between 13 and 130 µmol N_2 m^{-2} h^{-1}. These rates were largely determined by nitrate availability and temperature. To assess the influence of sediment type, we grouped sediments Büsum-I and Sylt-I into one sediment facies (group I) with moderate TOC content (1.6–1.9%; Table 1), high bulk AA concentrations (7–15 mg g^{-1}; Table 1), and low oxygen penetration depths (Table 2). In contrast, the two sediment facies Büsum-II and Sylt-II have low TOC concentrations (0.1–0.6%), low bulk AA concentrations (<1 mg g^{-1}, Table 1), and higher oxygen penetration depths (Table 2). When nitrate limitation (excluded in $^{15}NO_3^-$ amended cores) and temperature-related suppression of bacterial processes can be excluded as influences on denitrification rates in summer months (water temperatures above 15°C; Table 2), we expected effects of sediment characteristics to emerge from the data. Indeed, N_2 fluxes (m30) were highest during summer in group I sediments (104–176 µmol N_2 m^{-2} h^{-1}; Table 6). In group II sediments, N_2 fluxes (m30) were also highest in summer, but significantly lower than in group-I sediments (29–69 µmol N_2 m^{-2} h^{-1}; Table 6). There seems to be a trend of increasing N_2 production with increasing TOC content in the group I sediments, although our data set cannot be statistically evaluated because it encompasses only four different sediment types and TOC concentrations. But the tentative trend is consistent with numerous previous studies, where denitrification rates correlated positively with the TOC content (Seitzinger 1988; Sundbäck et al. 2000; Piña-Ochoa and Álvarez-Cobelas 2006; Deutsch et al. 2010).

A characteristic of sediments high in TOC and high denitrification rates is a thin oxygen penetration layer. It reflects a high sedimentary oxygen demand (SOD), high microbial biomass and a high overall microbial activity. Consistent with higher SOD in group I sediments (Table 5), the oxic zone in these cores is comparatively thin (Table 2). A thin oxic zone raises the denitrifying zone towards the sediment–water interface and implies a shorter diffusion path of nitrate, which increases the denitrification rate (Christensen et al. 1990), so that denitrification rates are inversely correlated with the oxygen penetration depth. We note that this relationship only holds for systems with high nitrate concentrations in overlying waters. In systems where nitrification in the sediments is the major source of nitrate for denitrification, a thick oxic layer may favour nitrification (Seitzinger 1988) where organic matter as a substrate for ammonification is available. In our case, deep oxygen penetration in group II sediments is accompanied by low ammonium fluxes (Table 3, 4) due to organic matter limitation, which in turn may limit nitrification.

In conclusion, denitrification rates are significantly higher in group I cores that have higher TOC content and AA concentrations, but the same supply of nitrate ($^{15}NO_3^-$ amended cores) does not necessarily induce the same rate of denitrification. Consistent with assumptions made by Duff et al. (1984), we suggest that low TOC concentrations in Büsum-II and Sylt-II cores cause low denitrification rates, whereas high TOC amount and AA concentrations induce high denitrification rates in Büsum-I and Sylt-I.

An estimate of annual nitrogen removal in the northern German Wadden Sea

Our ultimate goal was to quantify the role of sedimentary nitrogen loss in the Wadden Sea, which is suspected to be a major sink for riverborne nitrate loads due to favourable boundary conditions of nitrate availability and sediment facies. Ours are the first systematic data, and although spatial and temporal resolution is admittedly rather low, we feel justified to use this data set for a first estimate of annual nitrogen removal. When calculating annual nitrogen removal, our data suggest that the
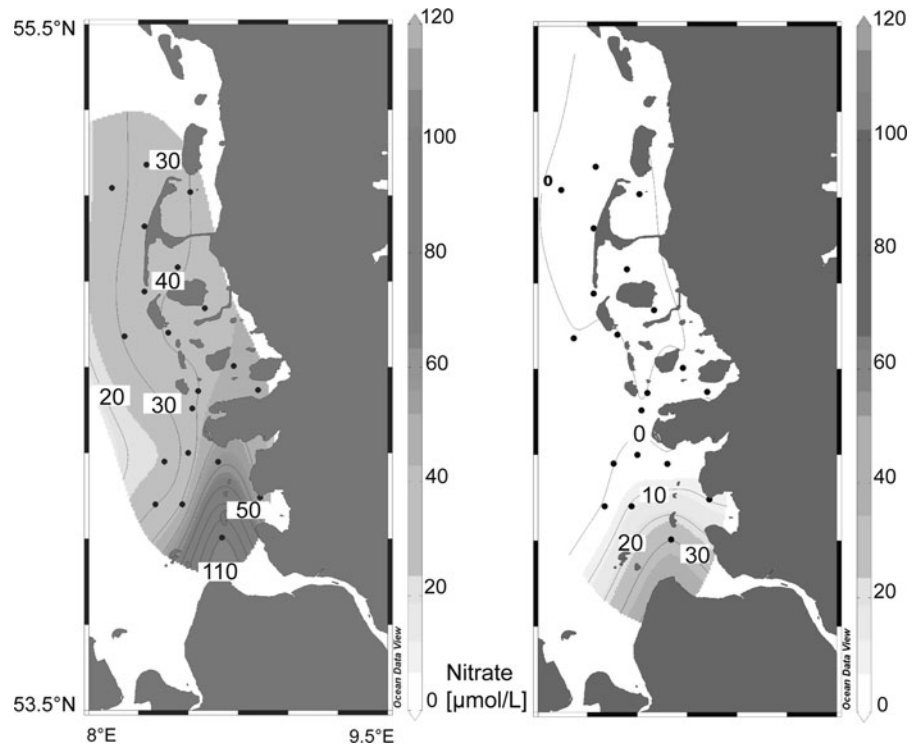
seasonality of NO_3^- concentrations and temperatures in the water body, and the sedimentary TOC content must be considered. Of these, the dominant controlling factor in the sense of net removal from the water column is the nitrate concentration in the water column. In Fig. 3 we show nitrate concentrations in the northern German Wadden Sea for February 2009 and August 2009. The nitrate distribution in February 2009 is representative for the winter months (October–April) and the nitrate distribution in August 2009 represents respective concentrations for summer months (May–September). In winter, denitrification is obviously not nitrate limited, and denitrification is potentially active in the entire north German Wadden Sea area (2.100 km^2), whereas ¾ of the area is nitrate limited in summer (Fig. 3). From these observations we assume that actual net-removal in summer is only effective in an area of about 525 km^2. Because denitrification rates also depend on permeability and the sedimentary TOC content, we have to consider the share of the individual sediment types. In the Wadden Sea, 65% of the sediments are sandy (proportion of grain size <63 μm is less than 10%), 30% are fine sand (proportion of grain size <63 μm is between 10 and 50%), and 5% are muddy (proportion

of grain size <63 μm is more than 50%) (Köster 1998).

To relate the rates determined in the different sediment types studied to the gross Wadden Sea sediment composition, we assigned rates determined in group-II sediments (Büsum-II and Sylt-II) to the sandy sediment facies, and rates in the group-I sediments (Büsum-I and Sylt-I) to the fine sand facies; muddy sediments were neglected due to their limited extent. Initially, we consider the sediments detached from the actual location, so that we can estimate denitrification rates for a certain sediment group at a given season. In the present study, measured actual N_2 production rates (m28) were highly dependent on location. Nitrate availability decreased with distance of the sampling site to the Elbe Estuary, which is the dominant source of nitrate in the northern Wadden Sea (van Beusekom et al. 2001). By considering total N_2 production rates (m28 + m29 + m30) from the $^{15}NO_3^-$ amended cores, we are able to estimate denitrification rates uncoupled from nitrate limitation and location. To account for seasonal temperature variation, we separated summer (sampling campaigns in May, June and July) and winter (sampling campaigns in

February, November and April) N_2 production rates. From this, we calculated mean total N_2 production rate (m28 + m29 + m30) of 49 µmol N_2 m^{-2} h^{-1} (sand) and 159 µmol N_2 m^{-2} h^{-1} (fine sand) in the summer, and 25 and 97 µmol N_2 m^{-2} h^{-1}, respectively, in the winter seasons. We then assigned these mean seasonal N_2 production rates to the percentage of sediment types as described in Köster (1998), and obtained a nitrogen removal rate of 3.8 mmol N m^{-2} day^{-1} in the summer and of 2.1 mmol N m^{-2} day^{-1} in the winter. Multiplied by the area (2,100 km^2 in winter and 525 km^2 in summer) of nitrate availability, we calculated an annual nitrogen removal rate of 16 kt N $year^{-1}$ for the north German Wadden Sea area. Given a total nitrogen load of the Elbe River of 116 kt in 2006 (Radach and Pätsch 2007), about 14% of the annual nitrogen load is removed by sedimentary N_2 production. But we only calculated loss in intertidal and shallow subtidal sediments of the northern sector: Considering that the entire North Sea Wadden Sea covers an area around 8,000 km^2 (including the Wadden Sea south of the Elbe River and the Dutch and Danish Wadden Sea), our estimate covers only 25% of the total Wadden Sea area. And nitrogen inputs from land may in addition be reduced by sedimentary denitrification, and anammox as well, in estuaries, and over an area of 750.000 km^2 in the offshore North Sea that have not been evaluated in this study.

Our estimate of an annual nitrogen removal rate is poorly constrained, mainly because of the low number of sites examined. Furthermore, there is a source of potentially significant error in our approach and the data set, because the applied experimental set-up did not imitate advective pore water transport and the overall influence of tidal cycling. The Wadden Sea is characterized by semi-diurnal tides and highly dynamic transport processes with different exposure times during low tide comparing subtidal and intertidal sediments. Advective pore water transport dominates during high tide and diffusive transport takes over during low tide exposure of permeable sediments (Huettel et al. 2003). Nutrient supply from the overlying water column and nutrient pore water concentrations show high variability over the tidal cycle, so that in situ denitrification rates are expected to be more variable than under steady state conditions assumed in the experimental design used here.

The rates measured in the sandy sediment facies in particular are likely to underestimate in situ rates at the sea floor. During high tide, nitrate is permanently flushed into permeable sediments (de Beer et al. 2005; Cook et al. 2006; Rao et al. 2007; 2008), which was not sufficiently imitated in the set-up of our flow-through incubations. According to Gihring et al. (2010), denitrification rates are up to ten times higher in percolated than in stirred set-ups. Recently, Gao et al. (2010) measured high potential denitrification rates (>80 µmol N_2 m^{-2} h^{-1}) in a percolation experiment with sediment cores from permeable sand in the southern German Wadden Sea area.

Nevertheless, N_2 production rates obtained in this study and the estimated annual nitrogen loss are relatively high compared to other studies from the 1990's, which were mostly conducted in offshore sediments (van Raaphorst et al. 1990, 1992; Law and Owens 1992; Lohse et al. 1993, 1996). Comparable studies by Kieskamp et al. (1991) in intertidal Wadden Sea sediments estimated an average denitrification potential of 0.3 mmol N m^{-2} day^{-1}. We attribute these relatively low older rate estimates to the acetylene-block-technique used at the time. Higher denitrification rates in Wadden Sea sediments have since been obtained with the isotope pairing technique (Jensen et al. 1996), who measured maximum rates of 1.2 mmol N m^{-2} day^{-1} in spring. In a mass-balance estimate by van Beusekom et al. (1999), the Wadden Sea was expected to remove nitrate at a (then considered excessive) rate of approximately 1.6 mmol N m^{-2} day^{-1}. Our findings suggest even higher nitrogen removal rates of 2.1–3.8 mmol N m^{-2} day^{-1} estimated from sediment core incubations. As explained above, this estimate may still be too low, because actual rates in permeable sands are likely to be higher than in our experiments (Gao et al. 2010). Therefore, we feel that our estimate for the Wadden Sea probably underestimates the potential of sediments in the coastal ocean for nitrogen removal and mitigation of river-induced eutrophication.

Conclusions

In this study we presented a first data set about seasonally varying N_2 production rates in typical sediments of the German Wadden Sea. The results of

our study suggest that major factors controlling nitrogen loss in coastal sediments of the German Bight are the nitrate concentrations in the water body, the temperature influencing biological activity, and the organic matter content of the sediment type. Sedimentary N_2 production rates increase with increasing nitrate delivery, suggesting that gross N loss increases in the vicinity of the nitrate advecting Elbe River plume. Whereas nitrogen removal in sediments at the island of Sylt are nitrate limited during summer months, N_2 production proceeds during all season in the Meldorf Bight situated in the outer Elbe Estuary. Experiments with $^{15}NO_3^-$ revealed that Wadden Sea sediments have a high potential to remove nitrate if it is abundant and available, thus providing an internal mechanism counteracting eutrophication by high river loads. In an extrapolation of seasonal mean N_2 production rates classified into different sediment types, we arrive at weighted nitrogen removal rates between 2.1 and 3.8 mmol N m^{-2} day^{-1}, suggesting intense nitrogen turnover in the entire Wadden Sea. Although further experiments in different sediment types and under varying conditions are required to exactly quantify nitrogen removal rates for the Greater North Sea area, it is clear that the near shore sedimentary nitrate sink accounts for a large share of the overall natural attenuation of man-induced eutrophication.

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