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Nitrogen stable isotope dynamics in the central Baltic Sea: influence of deep-water renewal on the N-cycle changes

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ABSTRACT: The vertical profiles of NO_3^-, NH_4^+, O_2, and H_2S as well as the isotopic composition of particulate nitrogen and NH_4^+ were sampled yearly over a 5 yr period in the Gotland Basin to follow biochemical changes in N-cycling resulting from an inflow of saltwater. The water column has a pronounced interface at 80 to 120 m depth which separates warm (13°C) brackish surface waters (salinity 7 psu) and the underlying cold winter water layer from more saline (9 to 11 psu) bottom waters originating from irregularly occurring inflow events of oxygenated, nitrate-rich North Sea water masses. Anoxic conditions usually exist in the deep stagnant waters, where nutrients only occur as ammonia, which reaches concentrations of up to 30 µmol l^{-1}. In spring 1993 large amounts of nitrate- and oxygen-rich water were transported into the deep waters of the Gotland Basin, thus displacing the stagnant deep water body. With the inflow, oxygen and nitrate concentrations rose by 3 ml l^{-1} and more than 10 µmol l^{-1} respectively. During the following years the concentrations of oxygen in the near bottom layer decreased again. The isotope signature of the suspended particles in the layer below 120 m reflects these changes: in 1993 the mean stable nitrogen isotope value in the anoxic water was at 1.1‰. We assume bacterial incorporation of ammonia to be the mechanism producing isotopically light particles. A fractionation factor calculated for ammonia uptake of 11‰ supports this hypothesis. During the following years the particles in the oxygenated water column were around 8‰ which is characteristic for microbially degraded material. The surface sediment of the central Gotland Sea has a low isotope signal of 3 to 4‰. These findings might have consequences for the interpretation of sediment $\delta^{15}N$ data where low isotope contents are usually taken as an indicator of high nutrient concentrations in surface waters.

KEY WORDS: $\delta^{15}N$ · Nitrogen cycling · Baltic Sea

INTRODUCTION

In recent years, the isotopic composition of marine suspended particulate organic nitrogen (PON) has been used to study particle dynamics on a seasonal scale and as an indicator of the sources and sinks of organic nitrogen in planktonic systems (Altabet 1989, Schäfer & Ittekkot 1993, Voss et al. 1996). On a longer time scale, the isotopic composition of sedimentary organic nitrogen has been used to characterize the relative availability of nutrients needed to support primary production in the surface layer (Calvert et al. 1992, François et al. 1992, Montoya 1994).

The usefulness of nitrogen isotope measurements in ecosystem level studies results from the isotopic discrimination that accompanies most biological transformations of nitrogen. In general, molecules containing the light isotope (^{14}N) are slightly preferred in enzymatic reactions that make or break a bond to the nitrogen atom, resulting in the formation of a product that is isotopically depleted relative to the substrate. The degree of this isotopic depletion is constant for a given reaction and can be measured in the laboratory (Mariotti et al. 1981, Montoya & McCarthy 1995) as well as in natural systems (Cifuentes et al. 1988, Horrigan et al. 1990, Montoya et al. 1991). In general, dissimilatory reactions such as nitrification and denitrification show very strong isotopic discrimination (Cline & Kaplan

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1975, Mariotti et al. 1981), leading to a marked increase in the ^{15}N content of the residual substrate pool (NH_4^+ and NO_3^-, respectively) as the reaction proceeds. Although assimilatory reactions discriminate to a lesser degree (Cifuentes et al. 1989, Hoch et al. 1992, Montoya & McCarthy 1995), growth of phytoplankton and bacteria on NO_3^- and NH_4^+ will produce biomass that is depleted with respect to the available inorganic substrate. Finally, it is worth noting that N_2-fixation is accompanied by little isotopic discrimination (Hoering & Ford 1960, Macko et al. 1987) and produces organic matter with an isotopic composition very similar to that of atmospheric N_2. As a result of isotopic fractionation, nitrogen cycle processes create characteristic isotopic differences between interacting pools of inorganic and organic nitrogen; these differences in turn provide a record of the major processes acting within an ecosystem.

To date, most investigations of the nitrogen isotope systematics of marine systems have been carried out in oxic waters, and relatively few ^{15}N natural abundance studies have focused on anoxic environments. Many reactions in the marine nitrogen cycle are highly sensitive to the presence and concentration of oxygen and are generally restricted to hypoxic and anoxic portions of the water column. PON collected in anoxic environments like the Peru upwelling and the Black Sea is depleted in ^{15}N relative to PON from oxic water columns (Libes & Deuser 1988, Fry et al. 1991). However, the factors that generate the low ^{15}N abundances from anoxic waters are not well understood at present.

In this paper, we discuss a suite of nutrient, stable isotope, and hydrographic measurements collected during a multiyear field program in the central Baltic Sea. In recent years, the central Baltic has provided an unusual juxtaposition of environments in which many important biological transformations of nitrogen may occur in close spatial proximity and on time scales of only a few months. The Baltic is essentially a large estuarine system with a bottom topography characterized by sills and deep basins, of which the Gotland Basin is the largest, with a maximum water depth of 240 m. The water column in the Gotland Basin is typically well stratified, with a seasonal thermocline at 20 to 30 m depth and a permanent halocline at 80 to 120 m depth (Gundersen 1981, Matthäus 1996). Intense surface blooms of N_2-fixing cyanobacteria (*Aphanizomenon* spp., *Anabena flos-aquae*, *Nodularia spumigena*) frequently occur in summer and autumn (Leppänen et al. 1988), and N_2-fixation may be a seasonally important source of new nitrogen in the surface layer (D. G. Capone, Montoya & Voss unpubl. data). The physical properties of the water layer between the seasonal thermocline and the halocline are largely determined by deep convective mixing during the winter, and this water mass is consequently known as winter water. Anoxic conditions occur frequently in the deep waters below the halocline in the Gotland Basin. The development of an oxygen-deficient zone at depth leads to denitrification and to a reduction in the ambient concentration of NO_3^- at depth to levels well below the ca 12 µmol l^{-1} characteristic of North Sea water (Gundersen 1981).

Although the deep waters of the Gotland Basin are typically anoxic, periodic incursions of oxygenated bottom water from the North Sea can lead to temporarily oxic conditions. These inflows occurred fairly regularly during the first three-quarters of this century (Matthäus 1995), but only a few small incursions have occurred since 1975/1976 and there were none from 1983 to 1993. The most recent events, in the winters of 1992/1993 and 1993/1994, recharged the Baltic Sea with ca 310 km^3 of water (Matthäus & Lass 1995). The transport of significant quantities of oxygenated North Sea water into the deep Baltic actually occurred over 2 years, beginning with an incursion of high salinity water (>19 psu) into the bottom 40 m of the water column in May 1993. In December 1993 and March 1994, smaller inflow events brought 200 km^3 of water directly into the Gotland Basin. These events were the first major inflow since 1975, and they resulted in deep water oxygen concentrations as high as 3.8 ml l^{-1} and NO_3^- concentrations greater than 10 µmol l^{-1}, the highest values measured since the 1930s (Matthäus & Lass 1995). This physical forcing caused a profound change in the biogeochemical environment of the central Baltic, with significant implications for the nitrogen cycle.

In this paper, we describe the spatial and temporal variations in the isotopic composition of particulate organic nitrogen and NH_4^+ in the Gotland Basin that accompanied the recent major incursion of deep water from the North Sea. The isotopic measurements provide an integrative measure of the effects of diverse nitrogen cycle processes acting within the Gotland Basin, and we present our results in the context of the shift between anoxic and oxic conditions in the deep waters.

MATERIAL AND METHODS

We collected samples during 5 cruises to the Gotland Basin in the central Baltic Sea (Fig. 1, 57° 17' N, 20° 05' E): a Baltic Monitoring Program cruise in 1992 (RV 'Professor Albrecht Penck', 5 to 24 May), 2 GOBEX (Gotland Basin Experiment, Institut für Ostseeforschung, Warnemünde) cruises in 1993 (RV 'Professor Albrecht Penck', 26 June to 3 July) and 1994 (RV 'Alexander von Humboldt', 15 to 28 July), an IOW

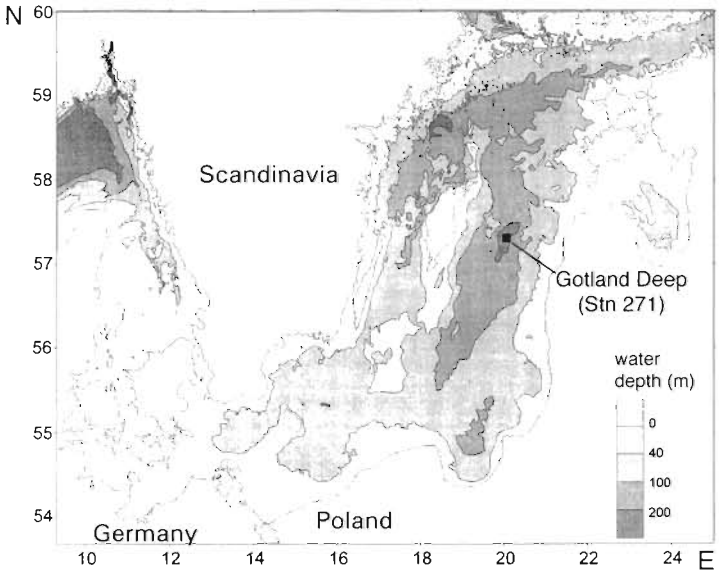

Fig. 1. Investigation area in the central Baltic Sea (Gotland Basin)

cruise in 1995 (RV 'Poseidon', 18 to 26 February), and a BASYS (Baltic System Studies, an EU project) cruise in 1996 (RV 'Professor Albrecht Penck', 1 to 7 July). In the years 1993 and 1994 drift studies in which a water body marked with a drifting buoy was sampled over several days was carried out. Details about the sampling strategy and methods will be described in detail elsewhere (F. V. Pollehne et al. unpubl.); here we describe only those methods relevant to this study.

Hydrographic data and discrete water samples were collected using a CTD-rosette system. The concentrations of NO_3^-, NO_2^-, NH_4^+, O_2, and H_2S were measured using standard protocols (Rhode & Nehring 1979, Grasshoff et al. 1983). At NH_4^+ concentrations above 20 µmol l^{-1} an extra calibration was carried out. The concentration of H_2S is expressed as a negative oxygen concentration (1 ml H_2S l^{-1} = −2.62 ml O_2 l^{-1}). Suspended particles were collected by gentle vacuum filtration (<25 cm Hg) through a 25 mm precombusted (450°C for 12 h) GF/F filter. The filters were frozen immediately and stored at −20°C while at sea. During the 1993 cruise, the filtrate was collected and frozen in polyethylene bottles that had previously been acidified and thoroughly rinsed.

Once ashore, the sample filters were dried at 60°C and prepared for isotope ratio analysis with a Finnigan Delta S mass spectrometer. Samples collected in 1993 were combusted in sealed glass tubes using a Dumas combustion technique (Minagawa et al. 1984); the C and N content of each sample was determined manometrically, and the sample gases formed were purified by cryogenic distillation before manual introduction to the mass spectrometer. All other samples were wrapped in tin cups (Heraeus CHN cups) and formed into pellets with a laboratory press for analysis by continuous flow isotope ratio mass spectrometry (CF-IRMS). We used a Carlo Erba 1108 CHN analyzer to measure the C and N content of each sample and to convert organic nitrogen to N_2 for isotopic analysis. A portion of the gas stream from the elemental analyzer was injected directly into the mass spectrometer ion source through a split interface for the isotope ratio measurement. All isotope abundances are expressed as per mil (‰) deviations from the isotopic composition of atmospheric N_2. Isotopically characterized organic standards (peptone, Merck Chemical) were analyzed with each batch of samples to provide an estimate of the accuracy and precision of our analytical protocols. The standard deviation of the $\delta^{15}N$ values measured for a day's standards was typically 0.05‰ (SD, N = 5), so our overall analytical error can be conservatively estimated as ±0.1‰. Both the reference gas and the organic standards have been intercalibrated with standards used at Harvard and can be related to the isotopic composition of the proposed NBS primary isotopic references for nitrogen.

Dissolved NH_4^+ was isolated for isotopic analysis using the distillation protocol of Horrigan et al. (1990) to trap the NH_4^+ on an ion sieve (Velinsky et al. 1989, Horrigan et al. 1990) and was then filtered onto precombusted GF/F filters. The subsequent isotope determinations were carried out as described for the particle samples. The distillation step reduced the overall precision of our measurements, and we estimate the overall error associated with the $\delta^{15}N$ of NH_4^+ to be <0.4‰. The mass requirements of the mass spectrometer prevented us from measuring the $\delta^{15}N$ of NH_4^+ in samples containing less than 2 µmol l^{-1} NH_4^+.

Isotopic fractionation. The isotopic fractionation associated with a reaction generates a predictable difference in isotopic composition between the substrate and product of that reaction, as well as a regular temporal change in the isotopic composition of both substrate and product as the reaction proceeds to completion. In this study, we measured isotope abundances relative to a working standard of ultra high purity N_2 (here: $\delta^{15}N$ = −8.09‰) and expressed them as per mil deviations from the isotopic composition of atmospheric N_2 using the $\delta^{15}N$ convention:

$$\delta^{15}N = \left(\frac{R_{sample}}{R_{atmosphere}} - 1\right) \cdot 1000 \quad (1)$$

where R is the isotope ratio (^{15}N:^{14}N). Although a number of conventions have been used to express the mag-

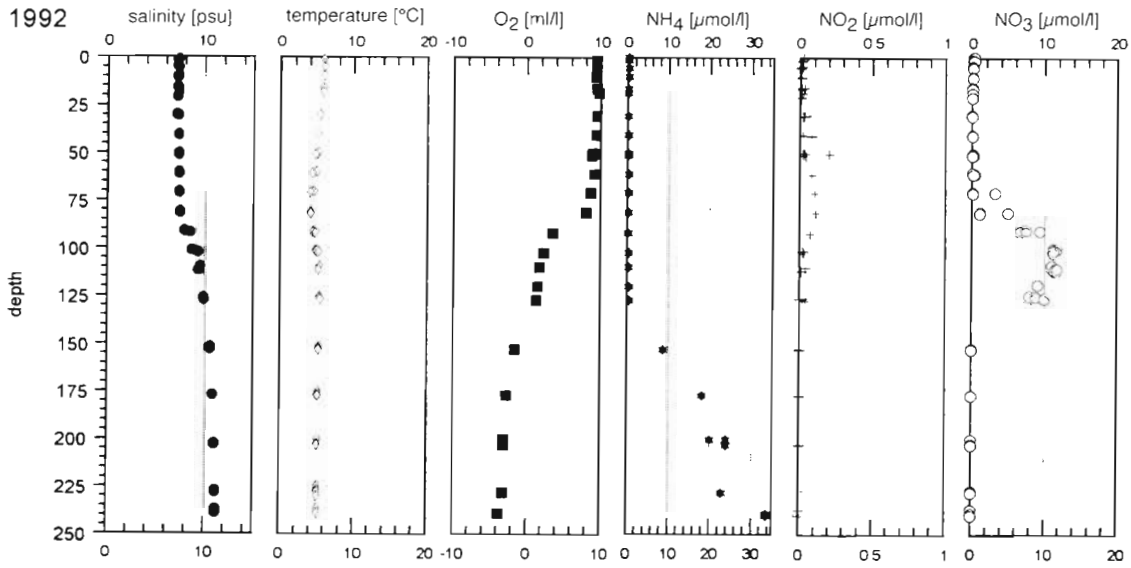

Fig. 2. Depth profiles of salinity, temperature, oxygen, NH_4^+, NO_2^- and NO_3^- from May 1992 in the central Gotland Basin. Note that hydrogen sulfide is expressed as negative oxygen concentration (conversion factor: –2.62)

nitude of isotopic discrimination associated with a reaction, we will do so in terms of the isotopic enrichment factor, ε, which is equal to the instantaneous difference in $\delta^{15}N$ between the available substrate and the product formed in a single step, unidirectional reaction (for further explanations see Mariotti et al. 1981). The enrichment factor is a linear transform of the isotopic fractionation factor, α, which is the ratio of rate constants for reaction of molecules containing the light (^{14}N) and heavy (^{15}N) isotopes ($\alpha = {}^{14}k:{}^{15}k$):

$$\varepsilon = (\alpha - 1) \cdot 1000 \quad (2)$$

We can use our data to estimate the magnitude of isotopic fractionation associated with a number of nitrogen cycle processes in the Gotland Basin, including NH_4^+ uptake. For a unidirectional process consuming a finite pool of substrate, the $\delta^{15}N$ of the residual substrate will be a function of the substrate concentration (Mariotti et al. 1981):

$$\delta^{15}N_{substrate} = \delta^{15}N_{substrate,\ initial} + \varepsilon \ln f \quad (3)$$

where f is the fraction of the original substrate pool left unreacted. In many field studies, only the relationship between $\delta^{15}N$ and pool size is measured, i.e. the initial pool size is unknown. In this case, Eq. (3) can be rewritten as:

$$\delta^{15}N_{substrate} = \delta^{15}N_{substrate,\ initial} + \varepsilon \ln[substrate] - \varepsilon \ln[substrate,\ initial]$$

or

$$\delta^{15}N_{substrate} = A + \varepsilon \ln[substrate] \quad (4)$$

where

$$A = \delta^{15}N_{substrate,\ initial} - \varepsilon \ln[substrate,\ initial]$$

Thus, a regression of $\delta^{15}N_{substrate}$ as a function of ln[substrate] will have a slope numerically equal to the isotopic enrichment factor. Note that A, the ordinal intercept in Eq. (4), is a composite term containing information on the initial isotopic composition and concentration of the substrate pool. We used this approach to estimate the enrichment factors for a number of processes acting within the Gotland Basin during our study (see Table 3).

RESULTS

Vertical profiles of temperature, salinity, and nutrient and O_2 concentrations during the study period clearly reveal the influence of North Sea water on the chemical environment below the thermocline. Our first data set, obtained in May 1992, showed O_2 concentrations to be decreasing steadily below ca 80 m, and a fully anoxic water column below 120 m with a salinity of 11.2 psu and a temperature of 5°C (Fig. 2). The concentration of NH_4^+ was below the limit of detection between the surface and 100 m, then increased steadily with depth to values above 30 µmol l^{-1} at 240 m. The concentration of NO_3^- was also below the limit of detection in the surface layer, then increased to a maximum of ca 12 µmol l^{-1} at 100 to 120 m before decreasing to zero in the deep layer. The concentration of NO_2^- reached a maximum of 0.21 µmol l^{-1} slightly above the NO_3^- peak. Samples were not collected for stable isotope analysis during this cruise; nonetheless, the data provide a baseline of physical and chemical conditions prior to the incursion of North Sea water.

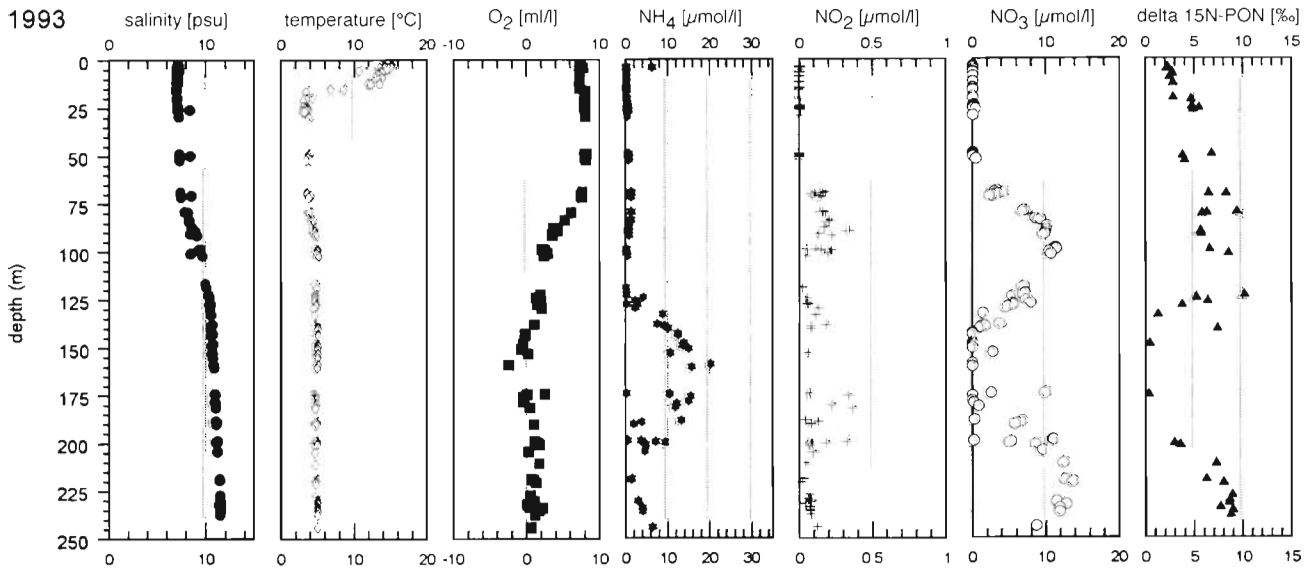

Fig. 3. Depth profiles of salinity, temperature, oxygen, NH_4^+, NO_2^- and NO_3^- and $\delta^{15}N$ of PON from June/July 1993. Note the coverage of data points within the anoxic zone is sparse

Following the first bottom water incursion in early 1993, oxygenated water (up to 2 ml l^{-1}) was found between 185 m and the bottom, underlying a zone with hydrogen sulfide (see also Nausch & Nehring 1994) (Fig. 3). The concentration of NH_4^+ showed a distinct maximum in the anoxic interval around 160 m and was 2 to 4 µmol l^{-1} in the bottom layer. In general, the $\delta^{15}N$ of NH_4^+ varied inversely with NH_4^+ concentration in the anoxic zone (Fig. 4); a minimum of 5‰ was measured around 160 m. The concentration of NO_3^- reached a peak at around 100 m, with a second peak in concentration at the bottom. The NO_2^- profile included 3 distinct local maxima, one just above and another just below the mid-depth NO_3^- maximum, and a third at the base of the NH_4^+ maximum. The concentrations of NO_3^-, NO_2^- and NH_4^+ all showed significant variation in the depth intervals between 125 and 130, and between 170 and 200 m, reflecting a complex horizontal interleaving of oxygenated water with the remaining anoxic water mass (Fig. 3). The $\delta^{15}N$ of PON shows marked vertical structure throughout the water column, with low values at the surface, high variability around 80 to 100 m, and a broad minimum in the NH_4^+-rich layer between 120 and 200 m.

During the winter of 1993/1994, 2 subsequent North Sea water inflows penetrated into the remainder of the water column below 120 m, resulting in low NH_4^+ concentrations and a total absence of anoxia throughout the water column (Fig. 5) by the summer of 1994. The

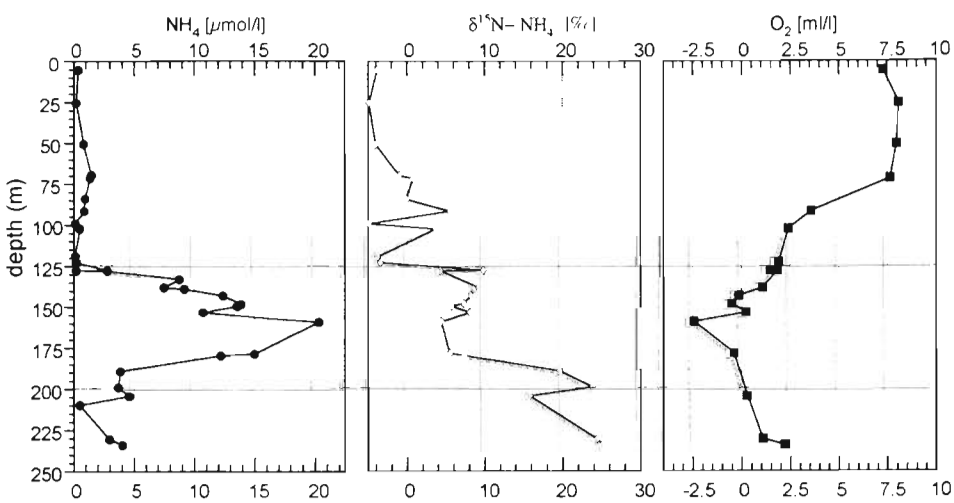

Fig. 4. Depth profiles of NH_4^+, $\delta^{15}N$ of NH_4^+ and oxygen. Note that hydrogen sulfide is expressed as negative oxygen concentration (conversion factor: –2.62)

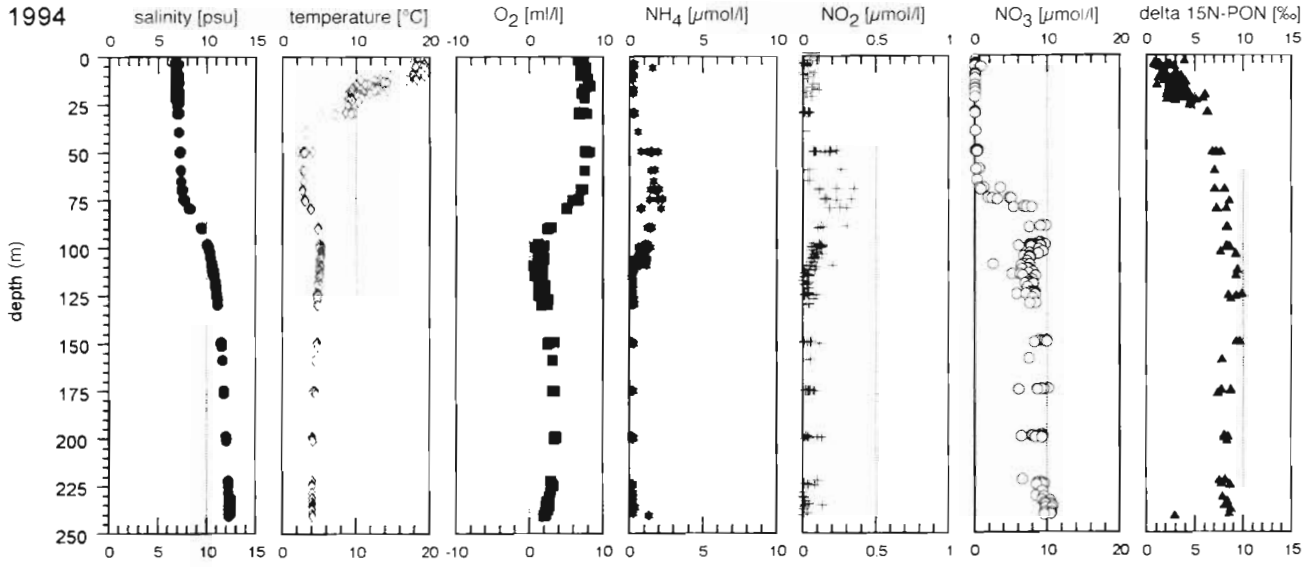

Fig. 5. Depth profiles of salinity, temperature, oxygen, NH_4^+, NO_2^- and NO_3^- and $\delta^{15}N$-PON from July 1994. At 240 m water depth, 1 sample with very low $\delta^{15}N$ of PON contained resuspended sediments

temperature was then at 4.1°C and the salinity had increased to 12.3 psu in the bottom water, leading to an overall increase in salinity of less than 1 psu. The mid-water peak in NO_3^- concentration was also absent from the layer immediately above the bottom water. The $\delta^{15}N$ of surface PON was again low, but then became rather uniform with depth below 50 m, varying between about 8 and 10‰, with maximum values clustered around 120 m depth.

In the 2 yr after the deep water inflow in 1994, the concentration of O_2 decreased slowly in the bottom water, a change accompanied by a small increase in NH_4^+ concentrations (Tables 1 & 2, Figs. 5 & 6; see also Nehring et al. 1995). The increase in NO_3^- concentra-

Table 1. Mean concentrations of NH_4^+, NO_3^- + NO_2^-, O_2, and PON and $\delta^{15}N$ of PON in 3 segments of the water column below the thermocline (see 'Discussion' for details). Values shown are means calculated from discrete measurements weighted by the depth interval represented by each individual sample. Not all data were used for the calculation, but rather 1 representative profile. The number of values used in each layer is given in parentheses

Year	Depth interval (m)	NH_4^+ (µmol l^{-1})	NO_3^- + NO_2^- (µmol l^{-1})	O_2 (ml l^{-1})	PON (µmol l^{-1})	$\delta^{15}N$-PON weighted mean (‰)
1992	50–120	0.39 (8)	5.02 (7)	5.23 (8)	No	No
May	120–200	11.08 (4)	1.58 (3)	−3.08 (5)	data	data
	200–240	23.62 (3)	0 (4)	−8.29 (5)		
1993	50–120	0.66 (4)	5.78 (4)	5.22 (4)	2.35 (4)	7.38 (4)
June	120–200	10.13 (9)	3.12 (9)	0.38 (9)	2.85 (6)	1.11 (6)
	200–240	3.39 (6)	11.44 (6)	1.05 (6)	2.65 (2)	5.30 (2)
1994	50–120	1.14 (7)	5.91 (7)	4.38 (7)	1.22 (7)	8.28 (7)
July	120–200	0.18 (6)	9.36 (6)	2.85 (6)	0.90 (8)	8.94 (8)
	200–240	0.22 (8)	9.13 (8)	2.99 (8)	1.61 (9)	8.15 (9)
1995	50–120	0.04 (5)	7.98 (5)	4.33 (5)	0.55 (5)	6.87 (5)
March	120–200	0.05 (2)	11.96 (2)	1.39 (2)	0.71 (5)	8.19 (5)
	200–240	0.05 (2)	14.52 (2)	1.28 (2)	0.99 (3)	8.26 (3)
1996	50–120	0.32 (8)	7.10 (8)	4.55 (8)	0.84 (6)	10.62 (6)
June	120–200	0.20 (3)	9.34 (3)	0.57 (3)	0.83 (3)	9.50 (3)
	200–240	1.49 (3)	3.72 (3)	0.15 (3)	1.49 (3)	6.62 (3)

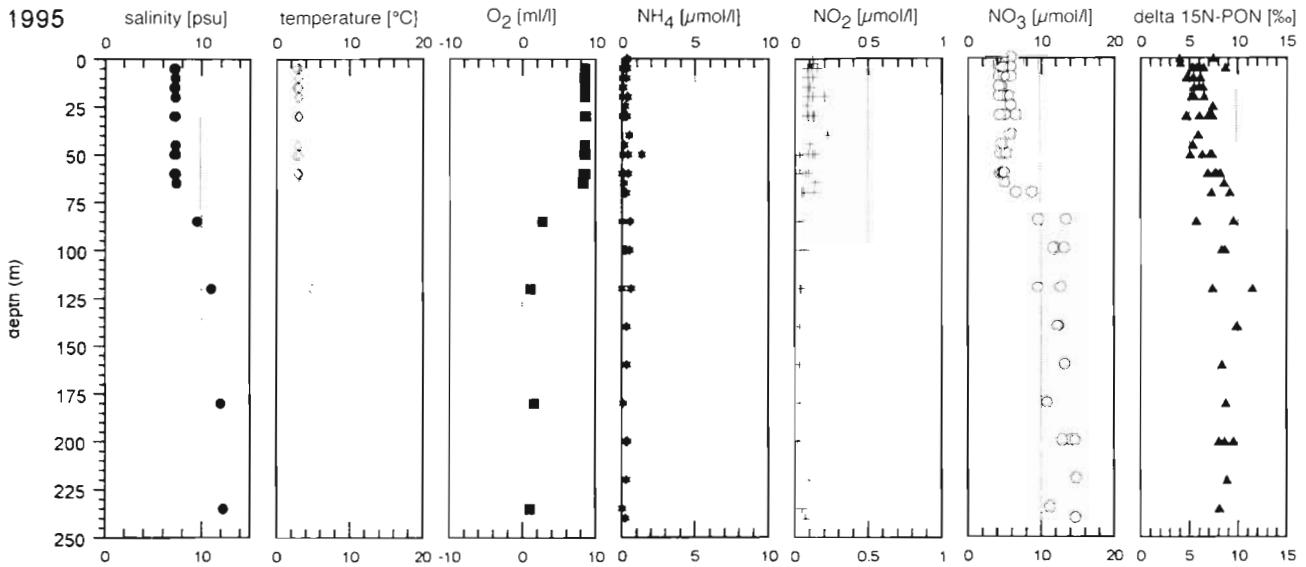

Fig. 6. Depth profiles of salinity, temperature, oxygen, NH_4^+, NO_2^- and NO_3^- and $\delta^{15}N$ of PON from Febuary 1995

tion in the layer between 200 and 240 m depth from 1994 to 1995 could be due to oxic degradation of PON sedimented from the surface and degraded during sinking. By the following year this bacterial breakdown of PON had drastically depleted the pool of oxygen (Table 2). The samples of PON obtained in the surface layer in 1995 and 1996 were not depleted in ^{15}N relative to the rest of the water column. In summer 1996, increased concentrations of NH_4^+ (5 µmol l^{-1}) and NO_2^- (0.3 µmol l^{-1}) were observed at 240 m depth, a sign that anoxic conditions were beginning to invade the water column from the sediment. The overall isotopic composition of suspended PON in deep water did not change markedly during 1995 and 1996, remaining high in both years (Figs. 6 & 7) with only a small decrease at the bottom (240 m).

DISCUSSION

The large quantities of oxygenated water that entered the Gotland Basin between 1993 and 1994

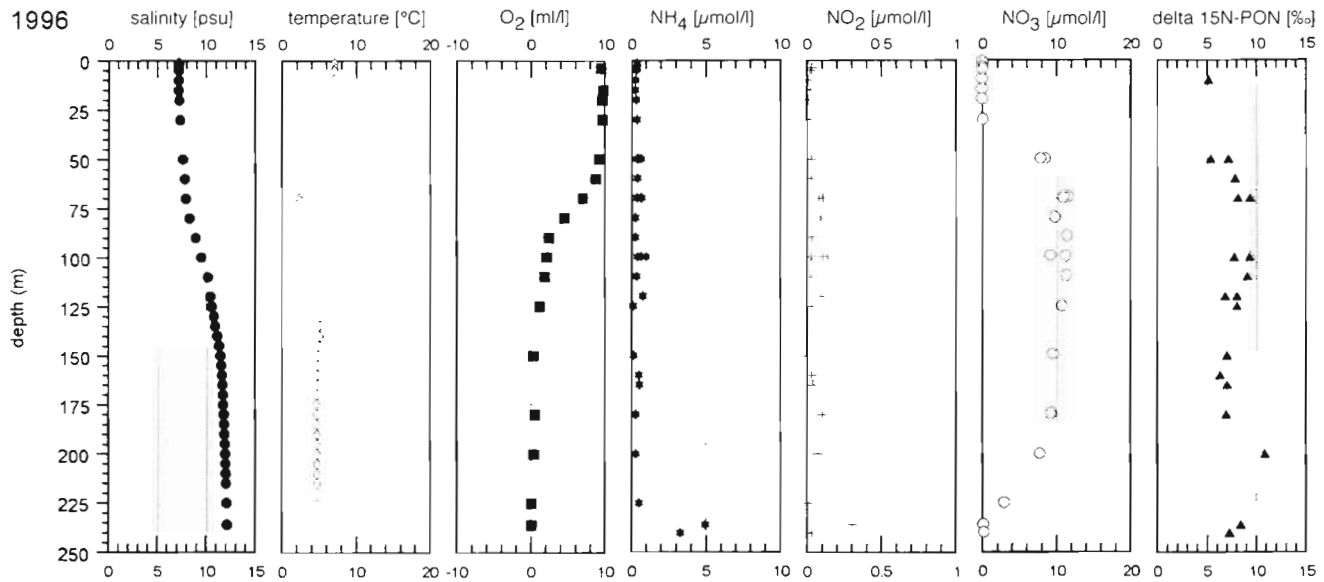

Fig. 7. Depth profiles of salinity, temperature, oxygen, NH_4^+, NO_2^- and NO_3^- and $\delta^{15}N$ of PON from July 1996

Table 2. Total amount of NH_4^+, NO_3^- + NO_2^- and O_2 below 120 m water depth towards the bottom. Values are mmol m^{-2} for the water column below 120 m

	1992	1993	1994	1995	1996
NO_3^- + NO_2^-	20	71	112	127	90
O_2	−11	7	35	17	5
NH_4^+	18	95	2	0.6	8

greatly altered the distribution of nitrogen species within the water column. Initially, this change simply reflected the physical replacement of anoxic waters with high NH_4^+ and low NO_3^- concentrations by oxygenated waters containing abundant NO_3^- and very little NH_4^+. Table 1 summarizes the bulk chemical properties of 3 distinct segments of the deep water column: (1) the winter water layer extending from the base of the thermocline to 120 m depth; (2) a layer extending from the halocline to 200 m which was characterized by low O_2 concentrations even after the first inflow event in the winter of 1992/1993; and (3) the deepest layer, with high concentrations of oxygen and nitrate extending from 200 to 240 m.

The change in chemical properties between 1994 and 1996 reflects the evolution of the system back towards the anoxic conditions typical of the basin, and allows us to estimate the magnitude of the organic inputs that are consuming O_2 and driving the system toward anoxia. The decrease of oxygen from 3 ml l^{-1} in summer 1994 to only 0.15 ml l^{-1} in 1996 is consistent with respiration of 127 µmol O_2 l^{-1} equivalent to 95.33 µmol C l^{-1} of organic carbon with a C:N ratio of 11.4 and a respiratory quotient (CO_2:O_2) of 0.75 (Kähler 1990). Although this rough calculation ignores the potential role of H_2S availability in regulating nitrate respiration (Brettar & Rheinheimer 1992), we can use it to estimate an average areal consumption of 1.9 mol C m^{-2} yr^{-1} (i.e. 23 g C m^{-2} yr^{-1}) in the bottom water over the 2 yr of sampling. Primary production in the surface layer could easily support the flux of organic carbon into the bottom water required to support this rate of respiration. Inorganic carbon budgets for the euphotic zone of the Gotland Basin imply an average rate of new production of 36 g C m^{-2} yr^{-1} (Thomas 1996), roughly half of which is supported by N_2-fixation. The consumption of oxygen in the water column could thus be explained by surface production and degradation. Roughly 65% of the organic matter from the surface layer appears to be consumed in the water column. However, the Gotland Basin is characterized by very high rates of sedimentary accumulation, on the order of 30 g C m^{-2} yr^{-1}, which also comprises laterally transported material (U. Struck et al. unpubl.).

Table 3. Fractionation factors for NH_4 uptake and PON degradation. ε is calculated from the slope of the regression line, where the substrate $\delta^{15}N$ values are plotted over the natural logarithm of the substrate concentration (see Eq. 3 in the text), r is the regression coefficent, and n is the number of samples

Process	Year	Depth interval (m)	ε	r	n
Ammonia uptake	1993	120–200	10.7	0.95	11
Nitrification	1993	180–240	14.3	0.95	6
PON degradation	1993	20–60	2.2	0.71	7
PON production (from NH_4 uptake)	1993	120–150	1.8	0.76	7
PON degradation	1994	20–60	2.4	0.69	27

Particulate organic nitrogen

The processes important for the understanding of stable nitrogen ratios in this investigation are nitrogen fixation, which introduces isotopically depleted material, and the degradation of PON, which results in an increase of $\delta^{15}N$. However, the fractionation within dissolved pools also affects the particulate isotope ratios, so that ammonia reduced in concentration by bacterial uptake becomes increasingly heavy in $\delta^{15}N$.

In the summers of 1993 and 1994, the vertical profiles of suspended PON show a clear surface minimum in $\delta^{15}N$. These low $\delta^{15}N$ values were associated with the presence of intense surface blooms of N_2-fixing cyanobacteria, which occur regularly in these waters in the late summer and autumn (Leppänen et al. 1988). Interestingly, the $\delta^{15}N$ of PON increased rapidly with depth below the surface mixed layer, a pattern that is commonly observed in oxic marine systems (Wada & Hattori 1976, Saino & Hattori 1980, Altabet & McCarthy 1986, Altabet 1989, Montoya et al. 1992), where it is attributed to the isotopic discrimination associated with microbial decomposition and remineralization of organic nitrogen. For our data from 1993 and 1994, we used Eq. (4) to estimate an isotopic enrichment factor of roughly 2.2‰ for the processes removing N from the pool of sinking organic matter (Table 3). These values are comparable to those derived from laboratory (Miyake & Wada 1971, Wada 1980) and field (Montoya et al. 1992) studies of organic matter decomposition under oxic conditions.

Despite the formation of ^{15}N-depleted organic matter by the N_2-fixers at the surface, the mean $\delta^{15}N$ of suspended particles in oxic portions of the underlying water column was relatively high, averaging about 8 to 10‰. As noted above, remineralization processes contributed to the elevated $\delta^{15}N$ at depth, but

denitrification may also have played a role in increasing the overall mean $\delta^{15}N$ of organic matter in the system. That is, the elevated $\delta^{15}N$ in PON at depth may partly reflect the effects of denitrification on the $\delta^{15}N$ of NO_3^-, since any residual NO_3^- will be enriched in ^{15}N as a result of the strong isotopic fractionation associated with denitrification (20 to 40‰). In view of the restriction of denitrification to the anoxic portion of the water column, deep convective mixing would be required to homogenize the isotopic signature of NO_3^- within the water column and deliver isotopically heavy nutrients to the surface waters. However, this is not expected to happen in the Baltic Sea.

Although the slowly sinking particles at depth typically had a relatively high $\delta^{15}N$ during our study, the sediments in the Gotland Basin have a low $\delta^{15}N$ (U. Struck et al. unpubl.). This suggests that the organic N entering the sediments is weighted heavily toward an isotopically depleted pool of particles. The most likely source of such low $\delta^{15}N$ particles is the surface layer, where intense blooms of N_2-fixing cyanobacteria occur during the summer and autumn. As noted above, new production in the surface layer may largly contribute to the vertical flux of organic matter to the deep water column, and roughly half of the total new production appears to be supported by N_2-fixation (Thomas 1996). Subtracting the atmospheric input which supplies 30% of new nitrogen to the central Baltic Sea on a yearly basis (HELCOM 1993) primary production from cyanobacteria is assumed to supply roughly 30% of nitrogen and carbon to the sediment. Note that under 'typical' conditions, PON within the anoxic zone also has a low $\delta^{15}N$ and could potentially contribute to the low $\delta^{15}N$ of organic matter entering the sediments, though this seems likely to be a relatively minor contribution to the total flux. In combination with sedimentary isotope measurements, our data suggest a relatively tight coupling between the isotopically depleted surface layer and the bottom. As a result, the sediments of the Gotland Basin appear to provide a rather faithful record of the isotopic composition of the uppermost water column and the activity of N_2-fixing organisms in particular.

Deep particulate organic nitrogen and NH_4^+ uptake

In 1993, the $\delta^{15}N$ of PON showed a broad minimum in the region of the NH_4^+ maximum within the low-O_2 region of the water column. Similarly low $\delta^{15}N$ values have been found in other anoxic and suboxic water masses, including those in the Black Sea and the Peruvian upwelling system (Libes & Deuser 1988, Fry et al. 1991). These low ^{15}N abundances have been attributed to the isotopic fractionation that accompanies the uptake of NH_4^+ by microbes, though nitrification may be an important sink for NH_4^+ in certain portions of the water column. In the Gotland Basin, the positive relationship between $\delta^{15}NH_4^+$ and the concentration of NO_2^- is consistent with an enrichment factor (Eq. 4) of 14.3‰ for consumption of NH_4^+ in the portions of the water column with elevated concentrations of NO_2^- (Table 3). This is consistent with literature values for the fractionation that accompanies nitrification, suggesting that this process is indeed a significant sink for NH_4^+ in those portions of the water column. Nonetheless, these zones of active nitrification occur at the lower margin of the anoxic zone and cannot account for the consumption of NH_4^+ within the bulk of the anoxic water mass.

We used the $\delta^{15}N$ and concentrations of PON and NH_4^+ to provide estimates of the processes that may have contributed to the low $\delta^{15}N$ of PON in the anoxic zone. A regression of $\delta^{15}NH_4^+$ as a function of $\ln[NH_4^+]$ provides an estimate of $\varepsilon = 11$‰ for the process(es) that remove NH_4^+ from the water column. This value is similar to the enrichment factors for bacterial uptake of NH_4^+ measured in laboratory experiments by Hoch et al. (1992; $\varepsilon = 13.9$‰, $[NH_4^+] = 38$ µmol l^{-1}), and are much smaller than the 20 to 30‰ enrichment factors typically associated with nitrification (Delwiche & Steyn 1970, Miyake & Wada 1971, Mariotti et al. 1981, Yoshida et al. 1989).

For bacteria growing in the anoxic waters of the Gotland Basin, the relatively low ambient concentrations of NH_4^+ (<30 µmol l^{-1}) suggest that uptake, rather than internal assimilatory processes, is likely to be the rate limiting step in bacterial growth on NH_4^+ since ammonia uptake at these concentrations is an active process catalyzed by the enzyme glutamine synthetase. Only at millimolar concentrations does the uptake occur via diffusion, and fractionation then depends on internal assimilation processes (Hoch et al. 1992). Although the $\delta^{15}N$ of suspended particles in the winter water is relatively high (ca 8 to 10‰), the PON collected from anoxic portions of the water column has a substantially lower $\delta^{15}N$ of ca 2‰. A similar depletion in ^{15}N has also been found in anoxic waters in the Peru upwelling and the Black Sea (Libes & Deuser 1988, Fry et al. 1991) and has been attributed to isotopic fractionation during the uptake of NH_4^+ by bacteria. The active transport of NH_4^+ by bacteria is accompanied by significant isotopic fractionation ($\varepsilon = 13.9$‰), as is the passive diffusion of NH_3 ($\varepsilon = 39$‰). At low ambient NH_4^+ concentrations, 'cyclic retention' of NH_4^+ may lead to a decrease in the expression of diffusional fractionation as NH_3 diffusing out of the cell is protonated and immediately pumped back in (Hoch et al. 1992). Although we do not know the degree to which different fractionation processes act in the Gotland Basin,

our data are consistent with an overall fractionation during NH_4^+ consumption of about 11‰, which is within the range of values expected from a combination of processes including the active transport of NH_4^+ by bacteria, back diffusion of NH_3, and the enzymatic incorporation of NH_4^+ by glutamine synthetase within the cell.

Altogether, this leads to a range of ε values from −8 to −19. Our own enrichment factor falls within this range, supporting the idea that a mixture of all processes might be responsible.

Conclusions

During a 5 yr field program in the central Baltic Sea covering a period of deep water exchange and thus a change from a strongly anoxic water mass containing hydrogen sulfide to one containing significant quantities of oxygen, the stable nitrogen isotopic composition of particles and ammonia gave insight into some major nitrogen cycling processes. Such irregular intrusions of dense water masses into the deep basins of the central Baltic Sea cause a suite of changes in the N-cycle processes of the water column. The primary change in the mean isotopic composition of PON in the water column was a 7‰ shift in the $\delta^{15}N$ of suspended particles as the initially oxic waters evolved toward an anoxic condition more typical of this region. A variety of isotopic, hydrographic, and chemical evidence suggests that this change resulted from microbial activity in the water column. Although the low $\delta^{15}N$ values that characterize PON in the anoxic portions of the water column appear to result from processes occurring *in situ*, the low $\delta^{15}N$ of the underlying sediments apparently results from inputs of cyanobacterial biomass from the upper water column.

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