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Year-round N_2O production by benthic NO_x reduction in a monomictic south-alpine lake

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Abstract. Nitrous oxide (N_2O) is a potent greenhouse gas, generated through microbial nitrogen (N) turnover processes, such as nitrification, nitrifier denitrification, and denitrification. Previous studies quantifying natural sources have mainly focused on soils and the ocean, but the potential role of terrestrial water bodies in the global N_2O budget has been widely neglected. Furthermore, the biogeochemical controls on the production rates and the microbial pathways that produce benthic N_2O in lakes are essentially unknown. In this study, benthic N_2O fluxes and the contributions of the microbial pathways that produce N_2O were assessed using ^{15}N label flow-through sediment incubations in the eutrophic, monomictic south basin of Lake Lugano in Switzerland. The sediments were a significant source of N_2O throughout the year, with production rates ranging between 140 and 2605 nmol $N_2O\,h^{-1}\,m^{-2}$, and the highest observed rates coinciding with periods of water column stratification and stably anoxic conditions in the overlying bottom water. Nitrate (NO_3^-) reduction via denitrification was found to be the major N_2O production pathway in the sediments under both oxygen-depleted and oxygen-replete conditions in the overlying water, while ammonium oxidation did not contribute significantly to the benthic N_2O flux. A marked portion (up to 15 %) of the total NO_3^- consumed by denitrification was reduced only to N_2O, without complete denitrification to N_2. These fluxes were highest when the bottom water had stabilized to a low-oxygen state, in contrast with the notion that stable anoxia is particularly conducive to complete denitrification without accumulation of N_2O. This study provides evidence that lake sediments are a significant source of N_2O to the overlying water and may produce large N_2O fluxes to the atmosphere during seasonal mixing events.

1 Introduction

Nitrous oxide (N_2O) is a potent greenhouse gas with a global warming potential that is ∼300 times higher than that of CO_2 over a 100 yr time horizon (Forster et al., 2007). Furthermore, N_2O is the most important stratospheric ozone-depleting substance currently being emitted to the atmosphere (Ravishankara et al., 2009). The atmospheric concentration has increased from 270 ppb in 1750 (Forster et al., 2007) to 323 ppb in 2011 (AGAGE, 2012), but there are still large uncertainties with regard to the relative contributions of the major sources and sinks of N_2O (Forster et al., 2007). Microbiological processes in soils and the ocean are the most important natural N_2O sources (Forster et al., 2007). However, the recent increase in atmospheric N_2O concentration is largely due to human intervention in the nitrogen (N) cycle, in particular through the agricultural use of synthetic N-based fertilizers (Codispoti et al., 2001; Bouwman et al., 2002; Mosier et al., 1998). In aquatic systems, anthropogenic fixed nitrogen loading can have multiple detrimental environmental effects, such as eutrophication, acidification, and the reduction of biodiversity (Galloway et al., 2003). In these environments, microbial processes taking place in redox transition zones play an important role in removing fixed N. Denitrification, for example, can be an important mechanism for removing fixed N along the land–ocean continuum by reducing it back to N_2. N_2O is a free intermediate in this process that may be released to the environment under certain conditions. N_2O is also produced during other N transformation reactions (Galloway et al., 2003) such as nitrification (specifically, ammonia oxidation) and nitrifier denitrification (Fig. 1).

Distinguishing the relative contributions of each of these major N_2O production and consumption pathways to the total N_2O flux is often challenging in aquatic systems because different types of microorganisms perform these pathways under overlapping environmental conditions. Furthermore, certain microbes carry out more than one pathway in response to changes in biogeochemical conditions. N_2O is produced during the aerobic oxidation of ammonium (NH_4^+) to nitrite (NO_2^-) when hydroxylamine (NH_2OH), an intermediate in the reaction, decomposes (Stein, 2011). Rates of ammonia oxidation depend primarily on substrate (NH_4^+) and oxygen (O_2) availability (Ward, 2008). However, in sediments, aerobic NH_4^+ oxidation and NO_2^- oxidation to nitrate (NO_3^-) can be closely coupled to anaerobic NO_3^- reduction in the redox transition zone (coupled nitrification–denitrification; Ward, 2008). Ammonia oxidizers also produce N_2O through a second mechanism known as nitrifier denitrification, an enzymatic pathway that sequentially reduces NO_2^- to nitric oxide (NO), and then N_2O (Wrage et al., 2001). The importance of nitrifier denitrification as a N_2O source appears to be higher under low-O_2 conditions (Ritchie and Nicholas, 1972; Poth and Focht, 1985). However, unlike denitrification, O_2 does not seem to inhibit nitrifier denitrification to the same extent or through the same mechanisms (Kool et al., 2011). Denitrification is the reduction of NO_3^- to N_2 via the gaseous intermediates NO and N_2O (Knowles, 1982) under anoxic or suboxic conditions (i.e., $[O_2] < 2$–$5\ \mu mol\ L^{-1}$; Devol, 2008; Codispoti et al., 2001). Denitrification can both produce and consume dissolved N_2O, releasing N_2O under conditions that suppress the activity of the N_2O reductase enzyme, for example, under low O_2 concentrations (Firestone et al., 1979; Otte et al., 1996). Furthermore, rapid transitions between oxic and suboxic conditions may cause "stop-and-go" denitrification, which causes N_2O accumulation in aquatic environments (Naqvi et al., 2000; Codispoti et al., 2001).

In lacustrine sediments, microbial activity consumes O_2 rapidly in the topmost millimeters, leading to suboxic or anoxic conditions in deeper sediment horizons, where denitrification becomes an important redox process (Hunting and van der Geest, 2011). The O_2 penetration depth is closely related to the O_2 concentration in the overlying water and the sediment reactivity (Lehmann et al., 2009; Thibodeau et al., 2010). A decrease in bottom water O_2 concentration is reflected in a narrower oxygenated zone in the sediment (Rasmussen and Jørgensen, 1992). Narrow redox zonation leads thus to an equally narrow succession of microbial processes (Stockdale et al., 2009). Changes in the redox zonation may have profound consequences on N_2O production (Otte et al., 1996). Seasonal cycles of water column mixing and stagnation can influence the oxidation state of surface sediments and modulate the penetration of redox boundaries into the sediments, potentially changing the redox environments of nitrifiers and denitrifiers (Rasmussen and Jørgensen, 1992). The few studies that have quantified N_2O fluxes from freshwater sediments indicate that lake sediments can be a significant source of N_2O. They also highlight that factors influencing N_2O production pathways in the benthic environment are still not clearly identified, particularly with regard to the relative importance of nitrification, nitrifier denitrification, and denitrification (Mengis et al., 1996; Liikanen et al., 2003b; Liikanen and Martikainen, 2003; McCrackin and Elser, 2010).

In this study, N_2O production pathways in lacustrine sediments were studied using ex situ steady state flow-through incubations with intact sediment cores (Lavrentyev et al., 2000; McCarthy et al., 2007; Liikanen et al., 2002a–c, 2003a, b; Liikanen and Martikainen, 2003) in combination with substrate ^{15}N labeling to assess benthic N_2O production rates and pathways. The experiments were conducted with sediments from a eutrophic, monomictic lake in southern Switzerland, the south basin of Lake Lugano. Monomixis and the resulting intermittent anoxia and suboxia of the bottom waters makes the south basin an ideal study site for testing the effects of variable bottom water oxygenation on the benthic N_2O production in a lake. Furthermore, previous measurements (Wenk, 2013) indicate high bottom water N_2O accumulation in the deep hypolimnion during thermal stratification in summer and fall, begging the question as to what causes N_2O accumulation in near-bottom waters. Upon water column overturn in winter, N_2O-laden bottom waters may be advected to the surface, enhancing N_2O fluxes into the atmosphere.

The study's objectives were (1) to estimate N_2O fluxes from the sediments to the overlying water column and to assess seasonal variations in these fluxes, (2) to identify the dominant benthic N_2O-producing processes in the lake, and (3) to study the possible impact of variable redox conditions of bottom waters during the seasonal cycle on N_2O production rates and pathways.

2 Sampling and methods

2.1 Site description

Lake Lugano is located in southern Switzerland/northern Italy at an altitude of 271 m above sea level (Fig. 2). It is divided by a natural dam into two main basins: the northern and southern basin (Barbieri and Polli, 1992; Lehmann et al., 2004a). Due to the limited water exchange, the basins are characterized by a distinct limnology, so that the northern and southern basins can be regarded as two separate lake systems that are connected by a narrow opening at Melide. A detailed overview of the lake's limnology can be found in Barbieri and Polli (1992) and Barbieri and Simona (2001).

This study focuses on the southern basin. It has an area of $20.3\ km^2$, a volume of $1.14\ km^3$ and a maximum depth of 95 m (Barbieri and Polli, 1992). Lazzaretti and Hanselmann (1992) and Lehmann et al. (2004a, b) described in detail

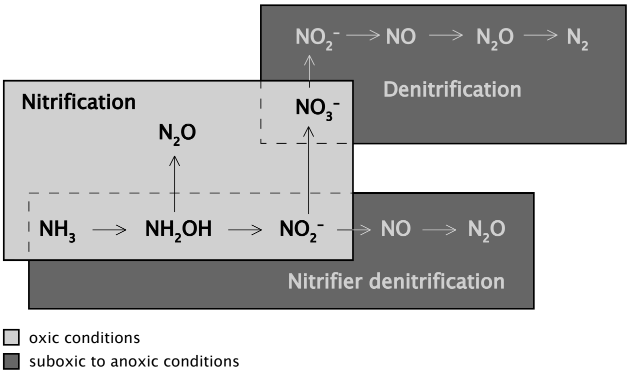

Fig. 1. N$_2$O production pathways in redox transition zones. Modified from Wrage et al. (2001).

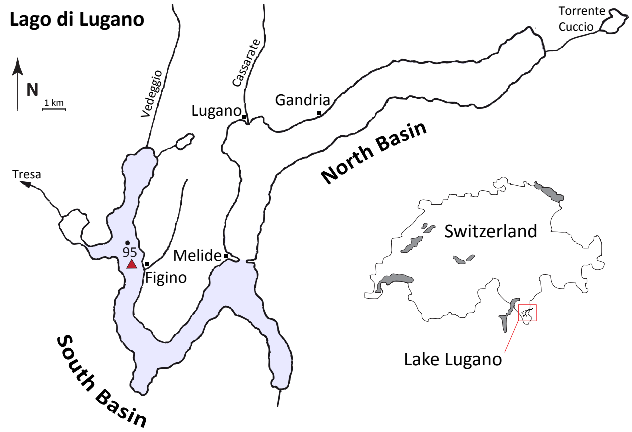

Fig. 2. Location and map of Lake Lugano. The sampling station (red triangle) is located in the south basin, west of the village of Figino, close to the point of maximum depth (modified from Barbieri and Polli, 1992).

the changes in seasonal redox conditions in the southern basin. During the mixing period (January/February to April), the whole water column becomes oxygenated and oxic conditions are found at the sediment/water interface until late spring. With the onset of thermo-stratification, generally in April, together with the increased phytoplankton production in surface waters and organic matter export to the hypolimnion, oxygen concentrations in the deep hypolimnion decrease, and by June/July, the redox transition zone has migrated from within the sediments into the water column by several meters. Complete anoxia prevails in the bottom waters until the water column turns over again in winter. Previous work on phosphorus (P) accumulation in the sediments of the south basin has shown spatial changes of the mineral content of the sediments within the south basin, and in turn the distribution of adsorbed P, as a result of the heterogeneity of the catchment geology (Veronesi, 1999). As for other sediment characteristics (porosity, organic N, organic C) the different sites within the south basin are comparable (Veronesi, 1999). The sampling site in this study is located west of the village of Figino (45°57′ N, 8°54′ E; Fig. 2) close to the deepest spot in the south basin (95 m). With regard to the above-mentioned sediment parameters, the chosen site can be considered most representative for the studied lake basin.

2.2 Sampling

Six ~50 cm sediment cores with 20 cm overlying bottom water were taken with a 5.7 cm diameter gravity corer in 2010 (April, August, October) and 2011 (January, May). The cores were stored upright and in the dark during transport to the home laboratory on the day of sampling. In addition, 3 × 20 L of bottom water were sampled using 10 L Niskin bottles. Bottom waters were stored in open 20 L plastic containers with headspace for samples taken during oxic conditions (April 2010, May 2011). During thermal stratification and anoxia in the deep hypolimnion, bottom waters were filled into gastight 25 L bags (Tedlar gas sampling bags, CEL Scientific Corporation, ITP-25) without headspace. In order to minimize the risk of O$_2$ contamination, only the lower three quarters of the Niskin bottle contents were used, and in October 2010 and January 2011, the bags were additionally stored underwater. In situ oxygen concentration of the bottom water was measured with a CTD profiler (Idronaut Ocean Seven 316Plus, Idronaut).

2.3 Steady state flow-through experiments

Steady state flow-through experiments were set up according to Gardner et al. (1991) and Lavretyev et al. (2000) (Fig. 3) within approx. 6 h after sampling in a cold room at near-in situ temperature (6.5 °C). The top caps on the liners were removed and replaced with gastight, O-ring sealed PVC plungers containing two holes. The plungers were lowered into the liners until all headspace air was released through the holes. Subsequently, the inlet water reservoir was connected to the core with gastight tubes (FEP, 0.8 mm inner diameter). A second tube connected the core with the sampling vial. A constant flow of ~1 mL min^{-1} was established with a peristaltic pump. For each sampling campaign, three duplicate flow-through experiments were set up, where two core incubations were supplied with water from one inlet water reservoir, respectively. One of the three inlet water reservoirs was amended with ^{15}N-NH$_4^+$ (ammonium chloride, > 99 % ^{15}N atom, Spectra Stable Isotopes), one with ^{15}N-NO$_3^-$ (potassium nitrate, > 99 % ^{15}N atom, Spectra Stable Isotopes) and one was left unamended as a control. The labeled substrates were added so that the in situ concentrations were doubled (final ^{15}N content ~50 %), except in the aerobic ambient O$_2$ incubations, where the background NH$_4^+$ concentration was < 1 µmol L^{-1}, and 10 µmol L^{-1} of ^{15}N-NH$_4^+$ label was added (final ^{15}N content > 90 %). In a recent study on the ammonia oxidation kinetics during archaeal ammonia oxidation in a natural marine environment (Horak

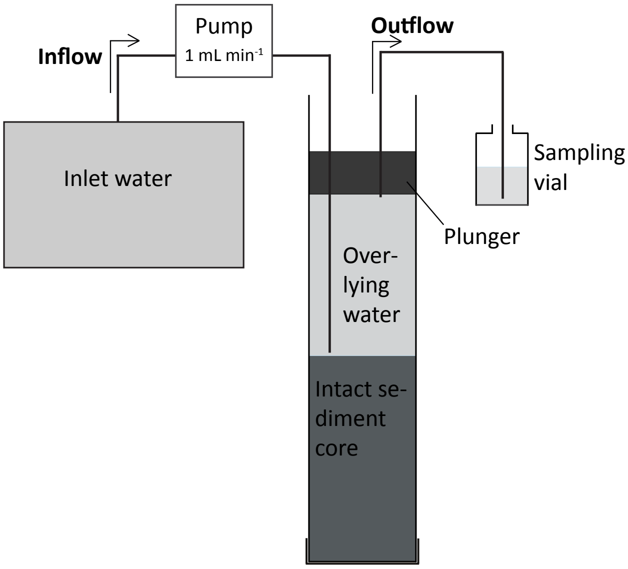

Fig. 3. Flow-through incubation setup.

et al., 2013), the ammonia oxidation half-saturation constant was less than 0.1 µmol L^{-1} (i.e., significantly lower than the ambient [NH_4^+] in this study). After a conditioning period of > 24 h (Gardner and McCarthy, 2009), the in- and outflows were sampled daily. Results are presented as the average of two (August 2010) or three incubation days (October 2010, January 2011, May 2011). In April 2010, samples for N_2O analysis were only taken on the last day of the experiment. Oxygen concentrations in the inlet water reservoirs and the outflow were measured daily with an optical sensor system (PreSens dipping probe; detection limit 0.5 µM, analytical error at suboxic oxygen levels 0.2 µM).

For N_2O analyses, glass vials (21 mL) were filled from bottom to top, and allowed to overflow for at least two bottle volumes to minimize N_2O exchange with air. The vials were capped with aluminium crimp caps with silicone septa (CS-Chromatographie Service GmbH, art. no. 300227). Subsequently, a 10 mL He headspace was added in exchange with water. The samples were sterilized with 0.2 mL of 10 mol L^{-1} NaOH to prevent further microbial activity (Sigman et al., 2001). Sample treatment was done within 1 h after sampling. After NaOH addition, the samples were analyzed within 3 days.

2.4 Determination of N_2O concentrations and benthic fluxes

N_2O concentrations and stable isotope ratios were determined using an isotope ratio mass spectrometer (IRMS, Thermo Finnigan Deltaplus XP), coupled to an automated purge and trap system (Thermo Finnigan GasBench II). N_2O concentration standards were produced using the denitrifier method (Sigman et al., 2001) to reduce NO_3^- to N_2O. Six KNO_3 solutions were produced to yield the following N_2O standard concentrations: 0.03, 0.1, 0.2, 0.5, 1.0, and 1.5 nmol N_2O mL^{-1}. Detector-sensitivity corrected IRMS peak areas of N_2O standards were compared to the respective concentrations in a regression analysis, and the resulting transfer function (Fig. 4) was used to calculate the N_2O concentrations (in nmol L^{-1}) based on detector signals of the masses 44, 45 and 46. These masses correspond to the [$^{14}N^{14}N^{16}O$], [$^{14}N^{15}N^{16}O + {}^{15}N^{14}N^{16}O + {}^{14}N^{14}N^{17}O$], and [$^{15}N^{15}N^{16}O + {}^{14}N^{14}N^{18}O + {}^{14}N^{15}N^{17}O + {}^{15}N^{14}N^{17}O$] isotopologues, respectively. For the ^{15}N-amended incubations, [$^{14}N^{15}N^{16}O + {}^{15}N^{14}N^{16}O$] $\gg$ [$^{14}N^{14}N^{17}O$], and [$^{15}N^{15}N^{16}O$] $\gg$ [$^{14}N^{14}N^{18}O + {}^{14}N^{15}N^{17}O + {}^{15}N^{14}N^{17}O$], and we therefore assume that any changes in the contributions of the natural abundance ^{18}O and ^{17}O isotopologues were negligible.

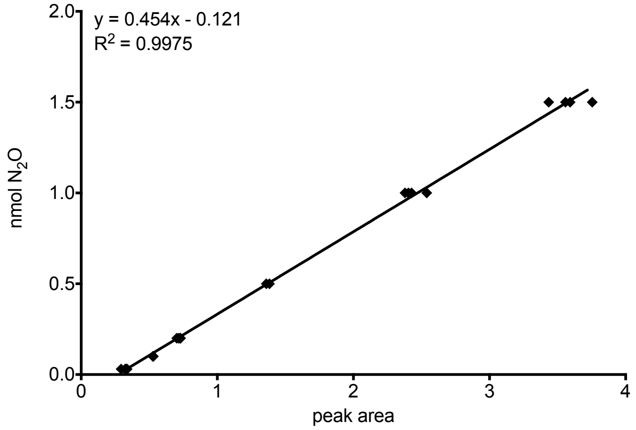

Fig. 4. Calibration curve and transfer function produced using the denitrifier method to measure dissolved N_2O concentrations. Data from January 2011.

Fluxes of N_2O of masses 44, 45 and 46 [nmol h^{-1} m^{-2}] from the sediment to the water column were then calculated from the concentration changes in the in- and out-flowing water:

$$\text{Flux}_{N_2O} = ([N_2O_{\text{out}}] - [N_2O_{\text{in}}]) \times \frac{Q}{A}, \tag{1}$$

where [$N_2O_{\text{out/in}}$] are the measured N_2O concentrations (nmol L^{-1}), Q the average flow rate (L h^{-1}) and A the sediment core surface area (m^2). Positive flux values indicate a net increase in N_2O concentration between the in- and outflowing water, and thus fluxes out of the sediments. Statistical analysis was done with the program Prism 6. Non-parametric tests (Kruskal–Wallis test and Mann–Whitney test, confidence level 95 %) were used to test for any statistical difference in N_2O fluxes between different experimental treatments, and between experiments under oxic vs. anoxic conditions.

Table 1. In situ temperature and concentrations of dissolved O_2, NH_4^+, NO_3^- and NO_2^- in bottom waters. Dissolved inorganic N concentrations were measured in the inlet water reservoirs of the control experiments (Wenk, 2013).

	Temp.	O_2	NH_4^+	NO_3^-	NO_2^-
	[°C]	[µmol L^{-1}]	[µmol L^{-1}]	[µmol L^{-1}]	[µmol L^{-1}]
April 2010	5.5	224	0.3	83.7	0.1
August 2010	5.7	0	7.1	75.7	1.7
October 2010	5.7	0	44.5	50.3	2.4
January 2011	5.9	11	13.2	69.6	2.3
May 2011	5.5	190	0.7	83.1	0

3 Results

3.1 Water column characteristics

The physical and chemical bottom water parameters at the five sampling dates are presented in Table 1. In April 2010, the water column was well mixed so that bottom waters were fully oxygenated, the NH_4^+ concentration was low (0.3 µmol L^{-1}) and the NO_3^- concentration was comparatively high (83.7 µmol L^{-1}). By August 2010, the lake was stratified, a 2 m thick anoxic near-bottom layer had developed, and the NH_4^+ concentration in the bottom water had increased (7.1 µmol L^{-1}). In October 2010, with ongoing stratification and organic matter decomposition, the anoxic bottom layer expanded by another 6 to 7 m into the water column. NO_2^- (2.4 µmol L^{-1}) as well as NH_4^+ (44.5 µmol L^{-1}) accumulated in the bottom water, but NO_3^- concentrations (50.3 µmol L^{-1}) decreased. In January 2011, samples were collected right at the beginning of the winter overturn. The O_2 gradient started to collapse, and suboxic to hypoxic concentrations of O_2 (11 µmol L^{-1}) were measured in bottom waters. In May 2011, the water column was fully oxygenated again.

3.2 Benthic N_2O fluxes

Net benthic N_2O fluxes were calculated for the total N_2O as well as the unlabeled ($^{14}N^{14}N^{16}O$), the singly labeled ($^{14}N^{15}N^{16}O$, $^{15}N^{14}N^{16}O$), and the doubly labeled N_2O ($^{15}N^{15}N^{16}O$) according to Eq. (1) (Fig. 5, Table 2). In all incubations, except January 2011, total N_2O fluxes were positive, indicating that sediments released N_2O to the overlying water column. N_2O flux measurements in unamended cores (core 1 and core 2), which represent N_2O fluxes under in situ NO_3^- and NH_4^+ concentration conditions, changed from 831 and 140 nmol $N_2O\,h^{-1}\,m^{-2}$ in April 2010, to 1115 and 259 nmol $N_2O\,h^{-1}\,m^{-2}$ in August 2010, to 2426 and 2605 nmol $N_2O\,h^{-1}\,m^{-2}$ in October 2010. In January and May 2011, average N_2O fluxes were again relatively low (202 and 195 nmol $N_2O\,h^{-1}\,m^{-2}$ in January 2011, and 178 and 189 nmol $N_2O\,h^{-1}\,m^{-2}$ in May 2011 for core 1 and core 2, respectively). Overall, total N_2O fluxes calculated from unamended cores showed a strong variation between duplicate cores. In April and August 2010 the difference between the N_2O fluxes of the replicate cores was most pronounced (difference ∼ 700 and ∼ 850 nmol $N_2O\,h^{-1}\,m^{-2}$, respectively) reflecting the relatively high heterogeneity of the sediments (Fig. 5). Independent of the variance between duplicate cores, inter-seasonal differences between oxic and anoxic conditions were significant (see below).

3.3 ^{15}N-N_2O recovery from the ^{15}N labeling experiments

^{15}N label was detected as ^{15}N-N_2O in all ^{15}N-NO_3^--labeled sediment core incubations (Fig. 5). Generally, ^{15}N-N_2O fluxes in the ^{15}N-NH_4^+-labeled cores did not exceed natural abundance levels. Only in January 2011 a slightly elevated ^{15}N-N_2O flux (16 nmol $N_2O\,h^{-1}\,m^{-2}$) was measured in one of the duplicate cores. Total N_2O fluxes in cores with ^{15}N-NH_4^+ and ^{15}N-NO_3^- additions were not significantly different from N_2O fluxes of unamended cores in April, August, October 2010 and May 2011 despite the two-fold increase in NO_3^- and the addition of NH_4^+. In January 2011, total N_2O fluxes in the ^{15}N-NO_3^- cores were significantly lower than in the unamended and the ^{15}N-NH_4^+-labeled cores ($p = 0.002$ and $p = 0.015$, respectively).

4 Discussion

4.1 The sediments are a net source of N_2O throughout the year

Lacustrine sediments are a known source of N_2O (McCrackin and Elser, 2010; Liikanen and Martikainen, 2003; Mengis et al., 1996). The N_2O fluxes reported here ranged between 140 nmol $N_2O\,h^{-1}\,m^{-2}$ (April 2010) and 1115 nmol $N_2O\,h^{-1}\,m^{-2}$ (August 2010), and were comparable to measurements reported previously for other eutrophic lakes. However, the October 2010 sampling stands out in this regard, with an exceptionally high production rate of 2605 nmol $N_2O\,h^{-1}\,m^{-2}$ (Fig. 5c). Mengis et al. (1996), for example, measured net benthic N_2O fluxes of 458–542 nmol

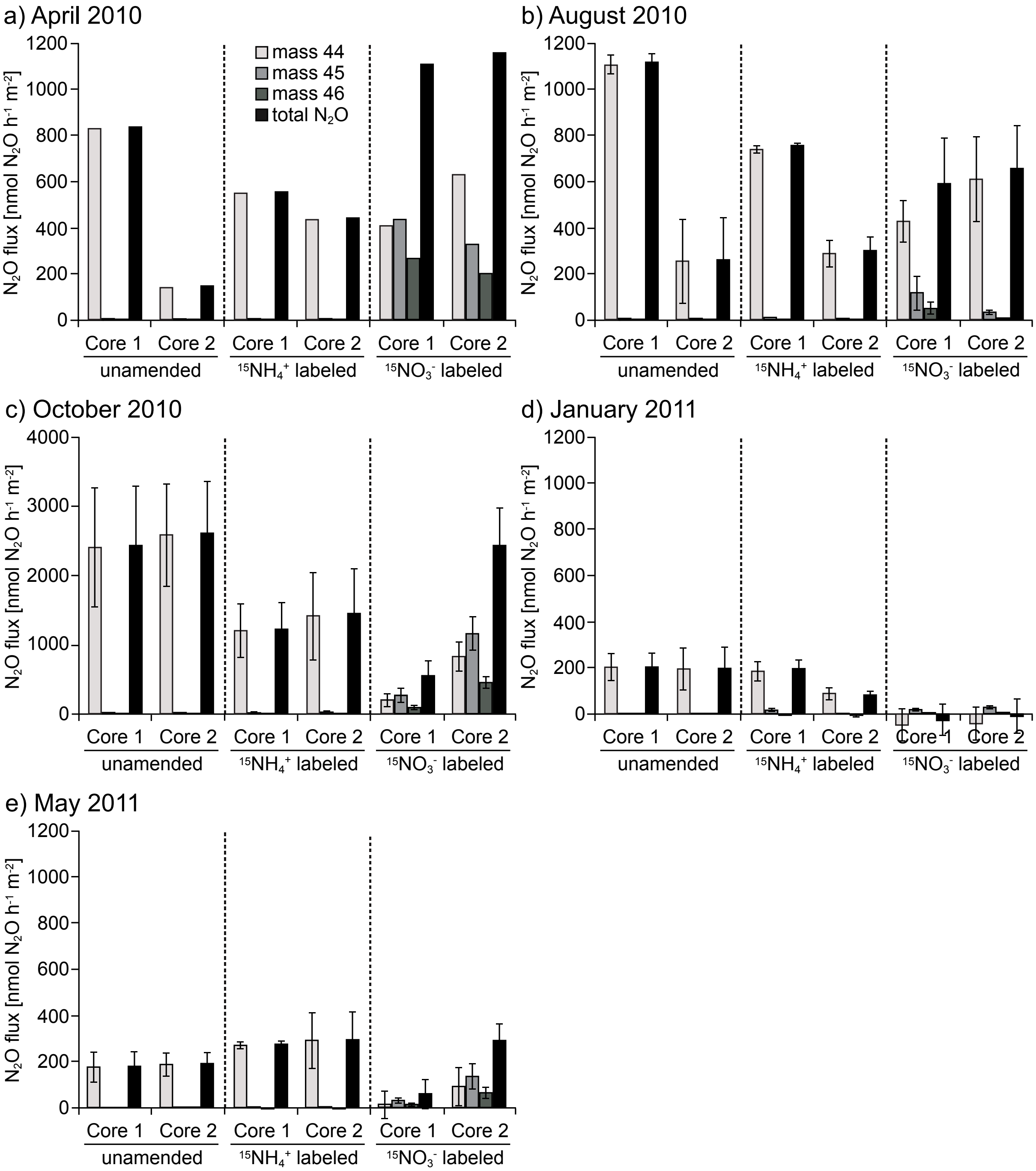

Fig. 5. N_2O fluxes from the sediment to the overlying water. Each column represents the average value of the N_2O fluxes over the experiment run time. Error bars show the standard error of the fluctuations over incubation time (April: single measurements; August: duplicate measurements; October–May: triplicate measurements). White: $^{14}N^{14}NO$ (mass 44). Light grey: $^{14}N^{15}NO$ and $^{15}N^{14}NO$ (mass 45). Dark grey: $^{15}N^{15}NO$ (mass 46). Black: total N_2O flux. Note the different scale in **(c)**.

Table 2. Average benthic N_2O fluxes of the masses 44 ($^{14}N^{14}NO$), 45 ($^{14}N^{15}NO$ and $^{15}N^{14}NO$) and 46 ($^{15}N^{15}NO$) and total fluxes measured in the flow-through experiments in April, August and October 2010, and January and May 2011 (SE denotes standard error; no replicate time points for April 2010).

			Mass 44 [nmol N_2O h^{-1} m^{-2}]	SE	Mass 45 [nmol N_2O h^{-1} m^{-2}]	SE	Mass 46 [nmol N_2O h^{-1} m^{-2}]	SE	Total [nmol N_2O h^{-1} m^{-2}]	SE
April 2010	Core 1	unamended	823.5		6.1		1.2		830.9	
		$^{15}N\text{-}NH_4^+$	546.6		4.9		1.2		552.7	
		$^{15}N\text{-}NO_3^-$	406.9		436.3		261.0		1104.2	
	Core 2	unamended	138.5		1.2		0.0		139.7	
		$^{15}N\text{-}NH_4^+$	432.6		4.9		1.2		438.7	
		$^{15}N\text{-}NO_3^-$	628.7		326.0		196.1		1150.7	
August 2010	Core 1	unamended	1103.7	40.6	8.9	0.4	2.5	0.4	1115.0	41.3
		$^{15}N\text{-}NH_4^+$	737.9	17.1	10.7	2.1	3.2	1.8	751.8	13.2
		$^{15}N\text{-}NO_3^-$	426.2	89.7	118.5	74.4	46.3	58.2	591.0	196.0
	Core 2	unamended	255.1	183.2	2.5	1.8	1.1	0.4	258.7	185.4
		$^{15}N\text{-}NH_4^+$	288.2	58.3	5.3	2.5	2.5	1.8	296.0	62.6
		$^{15}N\text{-}NO_3^-$	609.8	183.6	34.2	7.8	6.8	0.4	650.7	191.8
October 2010	Core 1	unamended	2401.1	857.0	19.1	6.9	5.4	1.8	2425.7	865.6
		$^{15}N\text{-}NH_4^+$	1192.4	382.8	21.6	7.8	2.9	0.8	1217.0	391.0
		$^{15}N\text{-}NO_3^-$	194.0	86.6	260.5	102.6	91.6	31.5	546.1	218.4
	Core 2	unamended	2575.8	741.6	20.4	5.8	5.5	1.8	2605.0	749.2
		$^{15}N\text{-}NH_4^+$	1409.7	633.9	28.3	13.7	2.9	1.8	1440.9	649.3
		$^{15}N\text{-}NO_3^-$	820.8	211.0	1156.2	241.4	451.2	86.9	2428.2	538.0
January 2011	Core 1	unamended	200.5	59.7	1.4	0.5	0.4	0.2	202.2	60.0
		$^{15}N\text{-}NH_4^+$	183.7	41.9	15.9	5.6	−6.3	0.9	192.9	39.7
		$^{15}N\text{-}NO_3^-$	−49.4	70.9	18.1	4.8	3.4	1.2	−27.9	67.9
	Core 2	unamended	193.5	91.0	1.4	0.7	0.6	0.3	195.1	91.9
		$^{15}N\text{-}NH_4^+$	85.9	24.4	0.9	1.3	−8.0	3.4	78.9	20.7
		$^{15}N\text{-}NO_3^-$	−43.6	71.9	27.9	5.9	4.8	1.2	−10.9	74.9
May 2011	Core 1	unamended	175.4	65.2	1.7	0.4	0.9	0.5	177.6	65.9
		$^{15}N\text{-}NH_4^+$	269.2	14.1	4.1	1.5	−0.4	0.4	272.8	15.6
		$^{15}N\text{-}NO_3^-$	13.0	59.7	31.3	11.0	14.2	5.7	59.0	63.1
	Core 2	unamended	186.4	50.6	1.8	0.5	0.5	0.5	189.1	50.5
		$^{15}N\text{-}NH_4^+$	290.1	121.9	3.2	1.3	−0.4	0.4	292.8	123.3
		$^{15}N\text{-}NO_3^-$	91.3	82.7	135.5	54.0	65.0	25.3	291.3	73.1

$N_2O\,h^{-1}\,m^{-2}$ at the bottom of the artificially oxygenated Lake Baldegg in static chamber experiments. Liikanen and Martikainen (2003) used microcosm incubation experiments with intact sediment cores to measure N_2O fluxes of up to 317 nmol $N_2O\,h^{-1}\,m^{-2}$ in a eutrophic lake in Finland. McCrackin and Elser (2010) reported an average N_2O flux of 550 nmol $N_2O\,h^{-1}\,m^{-2}$ in Norwegian lakes.

In general, comparison of the total N_2O fluxes from unamended cores (Fig. 5) to the NH_4^+- or NO_3^--amended cores shows no significant stimulation of N_2O production. In January 2011 the N_2O fluxes in the $^{15}N\text{-}NO_3^-$-labeled cores were even lower than in the unamended ones. This suggests that the in situ microbial processes were not substrate-limited so that the rates presented here are real rather than potential rates. The expectation that net N_2O fluxes vary seasonally, with potential links to water column stratification and oxygenation, was supported by our data. Incubations during fully established anoxic bottom-water conditions (August and October 2010) showed significantly ($p = 0.004$) higher N_2O fluxes than during oxic incubations (April 2010 and May 2011). Total N_2O fluxes seemed to increase during the stagnation period with increasing anoxia from August to October 2010, when maximal values were reached. Such high sedimentary N_2O production during stratification can lead to N_2O accumulation and oversaturation in the bottom water, as measured in 2009 (maximum of > 900 nmol L^{-1}; Wenk, 2013). At the beginning of the water column overturn in January 2011, together with the breakdown of the O_2 gradient, small amounts of O_2 (11 µmol L^{-1} O_2) were measured in bottom waters, and again, significantly ($p = 0.007$) lower benthic N_2O fluxes than during fully anoxic conditions were observed. The fact that more reduced conditions during stratification foster N_2O production, and that O_2 seems to hinder benthic N_2O production, implies that denitrification is the dominant N_2O production pathway (see below). It has been suggested that the rapid injection of small quantities of O_2 into O_2-deficient environments may be conducive to N_2O release by denitrifiers (Codispoti et al., 2001). However, our results indicate that total N_2O production rates were in fact very low during the initial phase of the destratification period (January 2011), the period when pulses of O_2-laden bottom waters would have been expected to cause this type of perturbation of the denitrifiers' redox environment. Rather, the reduced N_2O production in January 2011 suggests that the introduction of low concentrations of O_2 may, in fact, inhibit N_2O release.

We cannot rule out the possibility that other factors besides O_2 also impacted the activity of the microbial community, and thus N_2O production. Sediment heterogeneity on small spatial scales, and the availability of reducing substrates like organic carbon or sulfide (Wenk et al., 2013a), and inorganic nitrogen compounds are, along with oxygen availability, factors that influence microbial turnover within the nitrogen cycle. Yet, the concentrations of these substrates, as well as the composition of the microbial community itself, often also depend on the seasonally changing redox conditions, so that it is difficult to fully resolve the causalities (direct or indirect) between benthic N_2O production and redox conditions.

4.2 Denitrification is the main sedimentary source of N_2O

Significant $^{15}N\text{-}N_2O$ recovery was only observed in the overlying water of the $^{15}N\text{-}NO_3^-$-labeled cores, but not in the $^{15}N\text{-}NH_4^+$-labeled cores (Fig. 5), suggesting that denitrification is the predominant N_2O production pathway in the Lake Lugano south basin sediments. Even in April 2010 and May 2011, when the sediment/water interface was fully oxygenated and nitrification was likely to occur (Ward, 2008), N_2O derived from $^{15}N\text{-}NH_4^+$ was not observed. Therefore, it is very likely that canonical anoxic denitrification is the main source of N incorporated into the N_2O produced during the incubations. Rapid oxygen consumption supported by high rates of organic matter input to the sediments can reduce the oxygen penetration depth into the sediments, producing conditions favorable to denitrification close to the sediment/water interface (Li et al., 2007). It is still not clear, however, whether canonical denitrifiers carry out all of the steps involved in reducing NO_3^- to N_2O. The process of nitrifier denitrification may also produce N_2O in the presence of NO_2^- and small amounts of O_2 (as low as 5 µmol L^{-1}; Frame and Casciotti, 2010). Although NO_3^- cannot serve as an electron acceptor instead of NO_2^- during nitrifier denitrification, we cannot rule out the possibility that denitrification in the ambient environment supplies NO_2^- to ammonia oxidizers that then convert it to N_2O during nitrifier denitrification. On the other hand, with active N_2O production through nitrifier denitrification, at least some $^{15}N\text{-}N_2O$ production in the $^{15}N\text{-}NH_4^+$-labeled cores would also be expected, but was not observed.

In general, N_2O production through NH_4^+ oxidation was not observed at the oxygenated sediment/water interface in April 2010 and May 2011 (Fig. 5a, e), even though conditions were seemingly conducive to benthic ammonium oxidation. Obviously, there was always N_2O mass 44 production from ambient substrates, which could in theory originate from the nitrification of ambient pore water NH_4^+. This would imply a dilution of the $^{15}N\text{-}NH_4^+$ tracer by rapid NH_4^+ regeneration from organic N in the sediments, as shown for soil incubations by Norton and Stark (2011). However, isotope pairing predicts that even in the case of high dilution of the labeled ammonium with ambient ammonium from N remineralization within the sediments, a small contribution of NH_4^+ oxidation to N_2O production would be indicated by a measurable efflux of $^{15}N\text{-}N_2O$ in the $^{15}N\text{-}NH_4^+$-labeled experiments. It is also possible that the added $^{15}N\text{-}NH_4^+$ was assimilated by the microbial community before it reached the nitrification zone of the sediment, and thus before nitrifiers had a chance to oxidize it. Only in January 2011 did NH_4^+ oxidation produce N_2O, when O_2 was low but not absent, an observation that agrees with previous work showing that N_2O production during nitrification is enhanced under low O_2 conditions (Stein, 2011; Goreau et al., 1980). Finally, distinguishing between N_2O produced solely by denitrification and N_2O produced by nitrification coupled to denitrification is not possible using the ^{15}N tracer approach adopted here, although, as with nitrification, some $^{15}N\text{-}N_2O$ production in the $^{15}N\text{-}NH_4^+$-labeled cores would be expected in the case of active coupling of nitrification to denitrification to N_2O.

Denitrification rates determined in a parallel study were relatively high under the truly anoxic conditions observed in October 2010 (28.2 ± 23.7 µmol N h^{-1} m^{-2}; Wenk et al., 2013b), once redox gradients and, presumably, the microbial community had stabilized. At that time and also under anoxic conditions in August 2010, up to 15 % of total NO_3^- that was reduced was released as N_2O without being completely reduced to N_2. The comparatively high net N_2O fluxes measured during these incubations indicate that N_2O production was much more efficient than N_2O consumption. Although methodological limitations prevent us from completely ruling out N_2O production during ammonia oxidation, we conclude that N_2O released from the sediment is mainly produced through nitrate reduction, and accumulates in bottom waters particularly when denitrification rates are high.

5 Conclusions

This study demonstrates that the sediments of the Lake Lugano south basin are a net source of N_2O to the water column. NO_3^- reduction by denitrification was found to be the primary source of this N_2O, while N_2O production during ammonia oxidation was minimal or not observed at all, even when oxic conditions prevailed at the sediment/water interface. Net N_2O fluxes displayed significant temporal variations, and 1–15 % of the reduced NO_3^--N accumulated as N_2O in the water column. Surprisingly, the highest fraction of NO_3^- reduced to N_2O was observed after water column stratification and the development of anoxia in the deep hypolimnion, conditions that are generally thought to favor complete denitrification to N_2.

If we extrapolate the average value of total N_2O fluxes presented in this study (879 nmol $N_2O\,h^{-1}\,m^{-2}$) in time and over the area of the Lake Lugano south basin (20.3 km^2), we can estimate that the sediments of the Lake Lugano south basin release ~ 6.9 t $N_2O\,yr^{-1}$ to the overlying water. We are aware of the fact that benthic conditions are highly variable in time and space, and that this extrapolation is likely accompanied by a large uncertainty. Capturing all spatio-temporal variability for a more robust quantitative extrapolation would require a sampling regime with higher temporal resolution and additional sampling sites. Nevertheless, our study demonstrates that considerable amounts of N_2O may be produced in the sediments of the Lake Lugano south basin, with benthic N_2O fluxes that episodically exceed 2500 nmol $N_2O\,h^{-1}\,m^{-2}$. The fate of N_2O in the water column remains uncertain. N_2O fluxes from the surface waters to the atmosphere were not addressed in this study, and the capacity of microbial processes in the water column of the lake basin to remove the N_2O before it escapes to the atmosphere has yet to be determined. We speculate, however, that significant, year-round benthic N_2O production, as observed for Lake Lugano, can lead not only to the accumulation of N_2O in the bottom waters, but ultimately also to its episodic evasion to the atmosphere during periods of destratification and water column overturn. The relevance of such lacustrine N_2O degassing events for regional or global N_2O budgets awaits further investigation.

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References

AGAGE Advanced Global Atmospheric Gases Experiment: http://agage.eas.gatech.edu/data_archive/global_mean/global_mean_md.txt (last access: 29 December 2012), 2012.

Barbieri, A. and Polli, B.: Description of Lake Lugano, Aquat. Sci., 54, 181–183, doi:10.1007/BF00878135, 1992.

Barbieri, A. and Simona, M.: Trophic evolution of Lake Lugano related to external load reduction: Changes in phosphorus and nitrogen as well as oxygen balance and biological parameters, Lakes Reserv. Res. Manage., 6, 37–47, doi:10.1046/j.1440-1770.2001.00120.x, 2001.

Bouwman, A. F., Boumans, L. J. M., and Batjes, N. H.: Modeling global annual N_2O and NO emissions from fertilized fields, Global Biogeochem. Cy., 16, 1080, doi:10.1029/2001GB001812, 2002.

Codispoti, L. A., Brandes, J. A., Christensen, J. P., Devol, A. H., Naqvi, S. W. A., Paerl, H. W., and Yoshinari, T.: The oceanic fixed nitrogen and nitrous oxide budgets: Moving targets as we enter the anthropocene?, Sci. Mar., 65, 85–105, 2001.

Devol, A. H.: Denitrification Including Anammox, in: Nitrogen in the Marine Environment, 2nd edition, Capone, D. G., Bronk, D. A., Mulholland, M. R., and Carpenter, E. J. (Eds.), Academic Press, San Diego, 199–261, 2008.

Firestone, M. K., Smith, M. S., Firestone, R. B., and Tiedje, J. M.: The influence of nitrate, nitrite, and oxygen on the composition of the gaseous products of denitrification in soil, Soil Sci. Soc. Am. J., 43, 1140–1144, doi:10.2136/sssaj1979.03615995004300060016x, 1979.

Forster, P., Ramaswamy, V., Artaxo, P., Berntsen, T., Betts, R., Fahey, D.W., Haywood, J., Lean, J., Lowe, D.C., Myhre, G., Nganga, J., Prinn, R., Raga, G., Schulz, M., and Van Dorland, R.: Changes in Atmospheric Constituents and in Radiative Forcing, in: Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, edited by: Solomon, S., Qin, D., Manning, M., Chen, Z., Marquis, M., Averyt, K. B., Tignor, M., and Miller, H. L., Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA, 130–234, 2007.

Frame, C. H. and Casciotti, K. L.: Biogeochemical controls and isotopic signatures of nitrous oxide production by a marine ammonia-oxidizing bacterium, Biogeosciences, 7, 2695–2709, doi:10.5194/bg-7-2695-2010, 2010.

Galloway, J. N., Aber, J. D., Erisman, J. W., Seitzinger, S. P., Howarth, R. W., Cowling, E. B., and Cosby, B. J.: The nitrogen cascade, Bioscience, 53, 341–356, doi:10.1641/0006-3568(2003)053[0341:tnc]2.0.co;2, 2003.

Gardner, W. S. and McCarthy, M. J.: Nitrogen dynamics at the sediment-water interface in shallow, sub-tropical Florida Bay: Why denitrification efficiency may decrease with increased eutrophication, Biogeochemistry, 95, 185–198, doi:10.1007/s10533-009-9329-5, 2009.

Gardner, W. S., Seitzinger, S. P., and Malczyk, J. M.: The effects of sea salts on the forms of nitrogen released from estuarine and freshwater sediments: Does ion pairing affect ammonium flux?, Estuaries, 14, 157–166, 1991.

Goreau, T. J., Kaplan, W. A., Wofsy, S. C., McElroy, M. B., Valois, F. W., and Watson, S. W.: Production of NO_2^- and N_2O by nitrifying bacteria at reduced concentrations of oxygen, Appl. Environ. Microb., 40, 526–532, 1980.

Horak, R. E. A., Qin, W., Schauer, A. J., Armbrust, E. V., Ingalls, A. E., Moffett, J. W., Stahl, D. A., and Devol, A. H.: Ammonia oxidation kinetics and temperature sensitivity of a natural marine community dominated by Archaea, ISME J., 7, 2023–2033, 2013.

Hunting, E. R. and van der Geest, H. G.: Predictability of bacterial activity and denitrification in aquatic sediments with continuous measurements of redox potential, Int. J. Environ. Sci. Te., 8, 553–560, 2011.

Knowles, R.: Denitrification, Microbiol. Rev., 46, 43–70, 1982.

Kool, D. M., Dolfing, J., Wrage, N., and Van Groenigen, J. W.: Nitrifier denitrification as a distinct and significant source of nitrous oxide from soil, Soil Biol. Biochem., 43, 174–178, doi:10.1016/j.soilbio.2010.09.030, 2011.

Lavrentyev, P. J., Gardner, W. S., and Yang, L.: Effects of the zebra mussel on nitrogen dynamics and the microbial community at the sediment-water interface, Aquat. Microb. Ecol., 21, 187–194, doi:10.3354/ame021187, 2000.

Lazzaretti, M. A. and Hanselmann, K. W.: The role of sediments in the phosphorus cycle in Lake Lugano. II. Seasonal and spatial variability of microbiological processes at the sediment-water interface, Aquat. Sci., 54, 285–299, doi:10.1007/BF00878142, 1992.

Lehmann, M. F., Bernasconi, S. M., McKenzie, J. A., Barbieri, A., Simona, M., and Veronesi, M.: Seasonal variation of the δ^{13}C and δ^{15}N of particulate and dissolved carbon and nitrogen in Lake Lugano: Constraints on biogeochemical cycling in a eutrophic lake, Limnol. Oceanogr., 49, 415–429, doi:10.4319/lo.2004.49.2.0415, 2004a.

Lehmann, M. F., Bernasconi, S. M., Barbieri, A., Simona, M., and McKenzie, J. A.: Interannual variation of the isotopic composition of sedimenting organic carbon and nitrogen in Lake Lugano: A long-term sediment trap study, Limnol. Oceanogr., 49, 839–849, doi:10.4319/lo.2004.49.3.0839, 2004b.

Lehmann, M. F., Barnett, B., Gelinas, Y., Gilbert, D., Maranger, R. J., Mucci, A., Sundby, B., and Thibodeau, B.: Aerobic respiration and hypoxia in the Lower St. Lawrence Estuary: Stable isotope ratios of dissolved oxygen constrain oxygen sink partitioning, Limnol. Oceanogr., 54, 2157–2169, doi:10.4319/lo.2009.54.6.2157, 2009.

Li, Q. M., Zhang, W., Wang, X. X., Zhou, Y. Y., Yang, H., and Ji, G. L.: Phosphorus in interstitial water induced by redox potential in sediment of Dianchi Lake, China, Pedosphere, 17, 739–746, doi:10.1016/S1002-0160(07)60089-7, 2007.

Liikanen, A. and Martikainen, P. J.: Effect of ammonium and oxygen on methane and nitrous oxide fluxes across sediment-water interface in a eutrophic lake, Chemosphere, 52, 1287–1293, doi:10.1016/s0045-6535(03)00224-8, 2003.

Liikanen, A., Tanskanen, H., Murtoniemi, T., and Martikainen, P. J.: A laboratory microcosm for simultaneous gas and nutrient flux measurements in sediments, Boreal Environ. Res., 7, 151–160, 2002a.

Liikanen, A., Flojt, L., and Martikainen, P.: Gas dynamics in eutrophic lake sediments affected by oxygen, nitrate, and sulfate, J. Environ. Qual., 31, 338–349, doi:10.2134/jeq2002.3380, 2002b.

Liikanen, A., Murtoniemi, T., Tanskanen, H., Vaisanen, T., and Martikainen, P. J.: Effects of temperature and oxygen availability on greenhouse gas and nutrient dynamics in sediment of a eutrophic mid-boreal lake, Biogeochemistry, 59, 269–286, doi:10.1023/a:1016015526712, 2002c.

Liikanen, A., Ratilainen, E., Saarnio, S., Alm, J., Martikainen, P. J., and Silvola, J.: Greenhouse gas dynamics in boreal, littoral sediments under raised CO_2 and nitrogen supply, Freshwater Biol., 48, 500–511, doi:10.1046/j.1365-2427.2003.01023.x, 2003a.

Liikanen, A., Huttunen, J. T., Murtoniemi, T., Tanskanen, H., Vaisanen, T., Silvola, J., Alm, J., and Martikainen, P. J.: Spatial and seasonal variation in greenhouse gas and nutrient dynamics and their interactions in the sediments of a boreal eutrophic lake, Biogeochemistry, 65, 83–103, doi:10.1023/A:1026070209387, 2003b.

McCarthy, M. J., Gardner, W. S., Lavrentyev, P. J., Moats, K. M., Joehem, F. J., and Klarer, D. M.: Effects of hydrological flow regime on sediment-water interface and water column nitrogen dynamics in a great lakes coastal wetland (Old Woman Creek, Lake Erie), J. Great Lakes Res., 33, 219–231, doi:10.3394/0380-1330(2007)33[219:EOHFRO]2.0.CO;2, 2007.

McCrackin, M. L. and Elser, J. J.: Atmospheric nitrogen deposition influences denitrification and nitrous oxide production in lakes, Ecology, 91, 528–539, doi:10.1890/08-2210.1, 2010.

Mengis, M., Gachter, R., and Wehrli, B.: Nitrous oxide emissions to the atmosphere from an artificially oxygenated lake, Limnol. Oceanogr., 41, 548–553, 1996.

Mosier, A., Kroeze, C., Nevison, C., Oenema, O., Seitzinger, S., and van Cleemput, O.: Closing the global N_2O budget: Nitrous oxide emissions through the agricultural nitrogen cycle – OECD/IPCC/IEA phase II development of IPCC guidelines for national greenhouse gas inventory methodology, Nutr. Cycl. Agroecosys., 52, 225–248, doi:10.1023/A:1009740530221, 1998.

Naqvi, S. W. A., Jayakumar, D. A., Narvekar, P. V., Naik, H., Sarma, V., D'Souza, W., Joseph, S., and George, M. D.: Increased marine production of N_2O due to intensifying anoxia on the Indian continental shelf, Nature, 408, 346–349, doi:10.1038/35042551, 2000.

Norton, J. M. and Stark, J. M.: Regulation and measurement of nitrification in terrestrial systems, in: Methods in Enzymology: Research on nitrification and related processes, edited by: Part, A. and Klotz, M. G., Elsevier Academic Press INC, San Diego, USA, 486, 343–368, ISBN: 978-0-12-381294-0, doi:10.1016/s0076-6879(11)86015-5, 2011.

Otte, S., Grobben, N. G., Robertson, L. A., Jetten, M. S. M., and Kuenen, J. G.: Nitrous oxide production by *Alcaligenes faecalis* under transient and dynamic aerobic and anaerobic conditions, Appl. Environ. Microb., 62, 2421–2426, 1996.

Poth, M. and Focht, D. D.: N-15 kinetic-analysis of N_2O production by Nitrosomonas-Europaea – An examination of nitrifier denitrification, Appl. Environ. Microb., 49, 1134–1141, 1985.

Rasmussen, H. and Jørgensen, B. B.: Microelectrode studies of seasonal oxygen-uptake in a coastal sediment – Role of molecular-diffusion, Mar. Ecol.-Prog. Ser., 81, 289–303, doi:10.3354/meps081289, 1992.

Ravishankara, A. R., Daniel, J. S., and Portmann, R. W.: Nitrous Oxide (N_2O): The Dominant Ozone-Depleting Substance Emitted in the 21st Century, Science, 326, 123–125, doi:10.1126/science.1176985, 2009.

Ritchie, G. A. F. and Nicholas, D. J.: Identrification of sources of nitrous-oxide produced by oxidative and reductive processes in *Nitrosomonas europaea*, Biochem. J., 126, 1181–1191, 1972.

Sigman, D. M., Casciotti, K. L., Andreani, M., Barford, C., Galanter, M., and Böhlke, J. K.: A bacterial method for the nitrogen isotopic analysis of nitrate in seawater and freshwater, Anal. Chem., 73, 4145–4153, doi:10.1021/ac010088e, 2001.

Stein, L. Y.: Surveying N_2O-producing pathways in bacteria, in: Methods in enzymology: Research on nitrification and related processes, Part A, edited by: Klotz, M. G., Elsevier Academic Press INC, San Diego, USA, 486, 131–152, ISBN: 978-0-12-381294-0, doi:10.1016/s0076-6879(11)86006-4, 2011.

Stockdale, A., Davison, W., and Zhang, H.: Micro-scale biogeochemical heterogeneity in sediments: A review of available technology and observed evidence, Earth-Sci. Rev., 92, 81–97, doi:10.1016/j.earscirev.2008.11.003, 2009.

Thibodeau, B., Lehmann, M. F., Kowarzyk, J., Mucci, A., Gelinas, Y., Gilbert, D., Maranger, R., and Alkhati, M.: Benthic nutrient fluxes along the Laurentian Channel: Impacts on the N budget of the St. Lawrence marine system, Estuar. Coast. Shelf S., 90, 195–205, doi:10.1016/j.ecss.2010.08.015, 2010.

Veronesi, M.: Nutrient release from the sediments of the southern basin of Lake Lugano (Switzerland – Italy), Ph.D. thesis, Faculty of Science, University of Zürich, Switzerland, 1999.

Ward, B. B.: Nitrification in marine systems, in: Nitrogen in the marine environment, 2nd edition, edited by: Capone, D. G., Bronk, D. A., Mulholland, M. R., and Carpenter, E. J., Academic Press, San Diego, 199–261, ISBN: 978-0-12-372522-6, 2008.

Wenk, C. B.: Nitrogen transformation pathways, rates and isotopic signatures in Lake Lugano, Ph.D. thesis, Department of Environmental Sciences, University of Basel, Switzerland, 2013.

Wenk, C. B., Blees, J., Zopfi, J., Veronesi, M., Bourbonnais, A., Schubert, C. J., Niemann, H., and Lehmann, M. F.: Anaerobic ammonium oxidation (anammox) bacteria and sulfide-dependent denitrifiers coexist in the water column of a meromictic south-alpine lake, Limnol. Oceanogr., 58, 1–12, doi:10.4319/lo.2013.58.1.0001, 2013a.

Wenk, C. B., Zopfi, J., Gardner, W. S., McCarthy, M., Niemann, H., Veronesi, M., and Lehmann, M. F.: Partitioning between benthic and pelagic nitrate reduction in the Lake Lugano south basin, Limnol. Oceanogr., submitted, 2013b.

Wrage, N., Velthof, G. L., van Beusichem, M. L., and Oenema, O.: Role of nitrifier denitrification in the production of nitrous oxide, Soil Biol. Biochem., 33, 1723–1732, doi:10.1016/S0038-0717(01)00096-7, 2001.