Tellus (2003), 55B, 737–750
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TELLUS
ISSN 0280–6509

Nitrous oxide exchanges with the atmosphere of a constructed wetland treating wastewater

Parameters and implications for emission factors

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(Manuscript Received 27 December 2001; in final form 17 October 2002)

ABSTRACT

Static chamber measurements of N_2O fluxes were taken during the 1998 and 1999 growth seasons in a Swedish constructed wetland receiving wastewater. The dominating plant species in different parts of the wetland were *Lemna minor* L., *Typha latifolia* L., *Spirogyra* sp. and *Glyceria maxima* (Hartm.) and *Phalaris arundinacea* (L.), respectively. There were large temporal and spatial variations in N_2O fluxes, which ranged from consumption at –350 to emissions at 1791 μg N_2O m^{-2} h^{-1}. The largest positive flux occurred in October 1999 and the lowest in the middle of July 1999. The average N_2O flux for the two years was 130 μg N_2O m^{-2} h^{-1} (SD = 220). No significant differences in N_2O fluxes were found between the years, even though the two growing seasons differed considerably with respect to both air temperature and precipitation. 15% of the fluxes were negative, showing a consumption of N_2O. Consumption occurred on a few occasions at most measurement sites and ranged from 1–350 μg N_2O m^{-2} h^{-1}. 13–43% of the variation in N_2O fluxes was explained by multiple linear regression analysis including principal components. Emission factors were calculated according to IPCC methods from the N_2O fluxes in the constructed wetland. The calculated emission factors were always lower (0.02–0.27%) compared to the default factor provided by the IPCC (0.75%). Thus, direct application of the IPCC default factor may lead to overestimation of N_2O fluxes from constructed wastewater-treating wetlands.

1. Introduction

Nitrous oxide (N_2O) is an important greenhouse gas and its concentration in the atmosphere has increased continuously since the pre-industrial era, from about 275 ppbv to current levels of approximately 311 ppbv (Houghton et al., 2001). N_2O has a long atmospheric lifetime (120 yr) and 320 times stronger global warming potential compared to CO_2 (100 yr estimate; Houghton et al., 1995). The increase in its atmospheric concentration is attributed to increased anthropogenic N_2O emissions due to increases in the production and use of nitrogen fertilizers, conversion of forest to agricultural land, especially in the tropics, increased biomass-burning and various other factors. (Smith, 1997). The annual global emission of N_2O from soils has been estimated to be 9.5 ± 4.5 Tg N, corresponding to about 65% of all emissions (Houghton et al., 2001). This is, therefore, the main source of N_2O in the atmosphere. Another 5.1 ± 4.5 Tg N yr^{-1} of the N_2O emissions are accounted for as of oceanic origin, while anthropogenic sources such as biomass burning, wastewater treatment in municipal treatment plants, traffic and fertilizer production are estimated

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to contribute another 3–8 Tg N yr^{-1} (Houghton et al., 2001).

N_2O is derived from microbiological processes, i.e. nitrification and denitrification, and it may be emitted directly from agricultural fields, animal pens, waste disposal plants or pastoral systems (Houghton et al., 1996). The leaching and transport of nitrogenous compounds (organic and inorganic) from sources such as agricultural and sewage treatment facilities, waste disposal systems and pastures can cause eutrophication of waters. Many wastewater plants are equipped with denitrification systems to reduce the nitrogen burden on the recipients (Kimochi et al., 1998), but constructed wetlands can also be used for this purpose (Fey et al., 1999). Wetlands to be used in this way are constructed with various levels of control to reduce the impact of nutrients and concomitant eutrophication of lakes, rivers and coastal areas in both developed as well as developing countries (Haberl, 1999). Many full-scale wetland systems for treating wastewater are in use and the area covered by such wetlands is increasing globally (IWA, 2000). The constructions often include features designed to promote nitrogen mineralisation and nitrification of the ammonium-rich effluents transported to the wetland. Strong denitrification capacity is necessary to ensure the release of nitrate- and nitrite-N, originating from either direct inputs or via mineralisation and nitrification, in the form of N_2. Thus, both main microbial processes giving rise to N_2O formation play important roles in the desired nutrient reduction. Studies on such wetlands have shown that they can generate substantial emissions of N_2O as well as methane (Fey et al., 1999; Gui et al., 2000).

In 1995, the IPCC developed a methodology for evaluating N_2O fluxes (Houghton et al., 1996). In this technique, literature data on nitrogen leaching and nitrous oxide emissions were used to calculate different default factors as tools to predict direct and indirect N_2O fluxes. Three important sources of N_2O fluxes were distinguished: direct fluxes from agricultural soils; direct fluxes from animal production; and fluxes indirectly induced by agricultural activity and municipal sewage treatment. The indirect N_2O fluxes include those caused by leaching and runoff from ground water, surface drainage, rivers and estuaries, fluxes generated in municipal sewage treatment plants, and those from rivers and watersheds where sewage water is discharged. The indirect default factor derived by the IPCC for these type of sources is 7.5 g N_2O-N kg^{-1} N_{sewage}.

The IPCC methodology was developed to provide a means for estimating global N_2O source strengths. However, it is based on a limited data set. For example, only 25 locations in urban, agricultural and woodland areas in Japan, Israel and the USA were used for estimating indirect sources of N_2O flux from groundwater and surface drainage fluxes (Houghton et al., 1996). The data set also clearly has a limited basis with respect to N_2O fluxes in constructed wetlands treating wastewater, prompting questions about its general applicability for assessing whether N_2O is formed and released from such systems and, if so, how much.

The aims of the present study were: to estimate the N_2O fluxes from a constructed wetland treating wastewater during the vegetative growth season; to determine the effects of environmental parameters on such N_2O fluxes; and to compare the system's N_2O flux to nitrogen supply ratios with the default factor derived using IPCC figures (Houghton et al., 1996).

2. Methodology

2.1. Terminology

In this paper the term N_2O flux is used for both negative and positive N_2O exchanges, while the word N_2O emission is used only for positive fluxes. Consumption is used to describe negative N_2O fluxes.

2.2. Field site

The study was conducted at the wastewater-treating wetland at Nykvarn in Linköping, Sweden (58°26′N, 15°36′E). This wetland was designed to treat secondary, treated municipal wastewater at a rate of 10–12 m^{-3} h^{-1}. However, during the measurement campaigns the water flow rates were 7–8 m^{-3} h^{-1}. The wetland was established in 1993 as a pilot, experimental facility designed to reduce water nitrogen levels. The wetland consists of two discontinuous series, each of three ponds joined by underground pipes (Fig. 1). After treatment in the constructed wetland, the wastewater is released to the river Stångån. One of the wetland series (A1–A3, Fig. 1) was chosen for the flux measurements in this study. Thus, a water flow of ca. 4 m^3 h^{-1} passed these ponds. The size of each pond is approximately 1000 m^2, but they vary in shape, depth and vegetation. The first pond (A1) in the series is ca. 20 × 50 m^2 in area and has a water depth of 10–30 cm. A2 and A3 are both about 30 × 30 m^2 in area and ca. 60–70 cm deep.

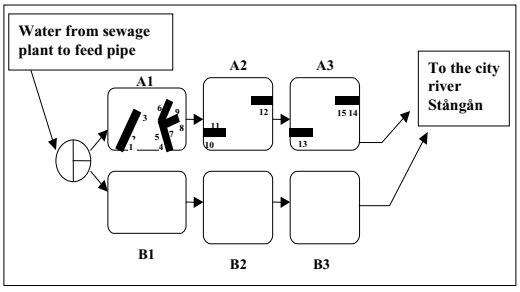

Fig. 1. Schematic view of the constructed wetland at Nykvarn, in Linköping, Sweden. Arrows indicate water flow. Boardwalks are shown in ponds A1-A3 with figures indicating the 15 different measurement locations.

Typha latifolia L. and *Lemna minor* L. dominate in A1, but there are also open areas close to the inlet and small zones with *Phalaris arundinacea* L. A2 is characterised by open areas and small stands of *T. latifolia*. In the middle of the vegetation season, *Elodea canadensis* Michx., *Spirogyra sp*. and *L. minor* also grow in this pond. About half of A3 was covered by *Glyceria maxima* Hartm. and the other half by *Spirogyra* sp. Since the nutrient concentrations in the water should decrease along the series of wetland ponds A1–A3, if they are functioning as planned, we decided to explore this presumed gradient. Thus, two boardwalks were built across each pond in the spring 1998, to give access to the different vegetation types in the respective ponds, including areas with no plants. Six wooden frames were fixed to randomly chosen points along the boardwalk in A1, and three frames to the boardwalks in both A2 and A3, into which aluminium collars (area 0.25 m^2) were inserted. The collars reached into the water, but did not touch the sediment surface to avoid disturbing the sediment structure and rhizomes. Because the vegetation was accidentally cut along the A1 boardwalks in August 1998, three new frames were established in the A1 area in early spring 1999 in order to compensate for this loss of habitat. As a consequence, 12 frames were used for the emission studies in 1998 and 15 in 1999.

2.3. Gas sampling

The gas exchange measurements (N_2O and CH_4) were taken using a static chamber technique adapted for emergent macrophytes as described by Chanton et al. (1993). The chambers were aluminium frames with transparent polycarbonate walls of 0.87 × 0.47 × 0.47 m^3 made gas-tight by silicon glue and equipped with a removable lid. To enable flux measurements to be taken from frames with growing vegetation, extending chamber sections were constructed. The lid and frames fitted into slots which were filled with water to ensure gas-tight connections. In this way the chamber volumes used ranged from 211 to 431 dm^3 over the season. The chambers were equipped with a cooling system to maintain ambient air temperature inside them. The device consisted of two fans (Multifan 4312 L, ELFA No. 54-105-84, Järfälla, Sweden) connected to a motorcycle radiator placed inside the chambers. The radiator was connected via a pump (Johnson Pump, model L650, Örebro, Sweden) to a container with ice water located outside the chamber. Thus, water was pumped from the water container through the radiator in the chamber, so that the fans could ventilate the chambers to maintain the desired temperature and ensure thorough gas mixing. The cooling was regulated by thermistors (Young Co., no. 41342 LC, Traverse City, MI, USA) registering the temperature difference between the air in the chamber and ambient air at 1 min resolution. The cooling device was operated by a data logger (Campbell Scientific Ltd., model CR10, Town, Leicestershire, UK) equipped with support software (PC 208W; version 2.3.). Humidity inside and outside the chambers was measured with hygrometers (Fischer Hygrometer model 111, www.Fisheredu.com), which were shaded from direct sunlight. Temperature sensors (Tsuruga sensors, TDS 3527, Tokyo, Japan) placed 10 cm below and 10 cm above the sediment, registered sediment and water temperatures. PAR was measured at every flux-sampling occasion (Broadband radiometer, model 1998-IL-1400, Newburyport and Skye Instruments Ltd., model SKP 200, Llandrindod Wells, Powys, UK).

In 1998 the field campaign lasted from May to early August, and in 1999 it started in April and ended in October. Flux measurements of N_2O were made every second week during 1998 and 1999, but once a month in September and October. Seven gas samples were taken over a period of 1–3 h (depending on the chamber volume).

Before sampling, the chambers were gently placed into the fixed aluminium frames, ensuring that the vegetation and the sediment were not disturbed. To facilitate pressure equilibrium during use of the chambers, a small hole was left open when the lid was placed on top of the chamber, and then sealed with a rubber septum. Gas samples were withdrawn using a sampling pump connected with gas-tight plastic tubings to the

chamber. A 22 mL gas-tight glass flask equipped with a rubber septum (Brown Chromatography Supplies, no. 151290, Wertheim, Germany) and sealed with an aluminium cap (Varian, 01-9000 36-08, Palo Alto, CA, USA) at the other end of the pump was circulated with chamber air for 30 s. Water and sediment temperatures, PAR and humidity were registered at every sampling occasion. Surface water was sampled inside each frame and immediately placed on ice for transport to the laboratory, where the samples were stored at −20 °C. Finally, the number of different plants and the plant cover were determined within each frame at every sampling occasion.

2.4. Nitrous oxide analysis and calculations

All gas samples from the chambers were analysed by gas chromatography (Varian, 3400) as described by Klemedtsson et al. (1997). The instrument was calibrated by injecting certified gas mixtures of N_2O in air purchased from AGA (AGA, code 0807).

The N_2O flux rates were calculated from the linear regression slopes of the accumulated gas concentrations in the chambers over time. The significance of regressions were evaluated according to Granberg et al. (2001), where the standard error of the estimate is compared with the total uncertainty (E_{tot}) of the flux sampling. In other words, the total uncertainty (E_{tot}) determines whether the regression is significant or not, according to the formula:

$$E_{tot} = E_a + E_b y \quad (1)$$

where E_a is the constant measurement error of the gas chromatographic analysis, E_b is the error relative to the sampled gas concentration and y is the mean value of the gas samples in each individual time series. The term E_b represents the error accumulated during the sampling procedure. In the present study, E_a varied between 0.004 and 0.017 (from May 1998 through October 1999) and E_b was estimated to be 0.01 (Klemedtsson et al., 1997). The first three measurements of each regression slope were used for estimating the N_2O fluxes to avoid underestimation of fluxes due to possible time-dependent disturbance effects of the static chamber techniques. In total, 92% of the N_2O flux measurements were accepted.

2.5. Calculations of vegetation volume

The presence and development of plants in the chambers reduced the gas volume in the chambers during the vegetation season. To account for this the plant volume was estimated using the vegetation volume–length relationships determined in the laboratory. The result of the volume estimations showed a difference of less than 5% between the N_2O fluxes derived with and without adjustments in 92% of the measurements. For the observations for which greater than 5% adjustments were made, 11 were between 5 and 10% and two were between 10 and 15%. There was no seasonal or spatial distribution pattern among these observations.

2.6. Nitrogen and phosphorus analysis

Two different sets of nitrogen data were used for this study, total nitrogen data collected weekly and mineralised nitrogen ($NH_4^+ + NO_3^- + NO_2^-$) collected at every measurement occasion. The total nitrogen data were based on wastewater samples taken from the inlet and the outlet of the constructed wetland. Total nitrogen was calculated as the sum of NO_3^-, NO_2^- and Kjeldahl nitrogen and analysed by spectrophotometric methods (TrAAcs, Bran+Luebbe, Norderstedt, in accordance with Swedish standards ISO 13395, ISO 5663). Samples for the determination of mineralised nitrogen were taken from each frame. The frozen water samples were gently thawed in the dark, and particles were allowed to settle. The supernatant was used for nitrogen analysis. All water samples were analysed by spectrophotometric methods (Autoanalyser 2, Bran Luebbe, Norderstedt, Sweden?); methods no. G-171-96 for NH_4^+ and G-172-96 for NO_3^- and NO_2^-. A cadmium reductor column was used to reduce NO_3^- to NO_2^- (Bran Luebbe 165-0301-01).

In order to include the water sample particles in the analysis for total phosphorus, the flasks were shaken before samples were pipetted. After this all samples were prepared in accordance with Swedish standard SS 028127, except that 31 mL glass flasks, closed with butyl rubber stoppers and sealed with aluminium caps, were used for the oxidation procedure to make sure that no volume was lost when the samples were oxidized during autoclaving (120 °C, 30 min). After this step spectrophotometric method no. G-175-96 was used (Bran Luebbe Autoanalyzer Applications).

2.7. Statistical methods

All statistical analyses were performed with SPSS version 9.0 software (SPSS, Inc., Chicago). The difference between the calculated N_2O flux means and medians indicated skewed distributions, which was

confirmed with probability plots. These plots showed that the data distributions were right-skewed, so the data set was normalised by log-transformation. Wilcoxon and Mann–Whitney *U*-tests were used to evaluate the significance of differences between frames, boardwalks and ponds. The significance of all statistical data was tested at the 5% probability level, unless otherwise stated. Kendall's tau was used to analyse autocorrelations between variables, and mutual relationships among them were then investigated by principal component analysis using the SPSS 'Factor' procedure. Stepwise multiple linear regression analysis was used to correlate the N_2O fluxes with the principal components. The alpha value was set to 0.05. All residuals were investigated for independence and normality. Three data sets were evaluated by separate PCA and regression analyses: the 1998 measurements, the 1999 measurements, and a combined data set for 1998 plus the 1999 data for the part of the year covered by the 1998 measurements. Thus, the spring and autumn data from 1999 were not included in the combined analysis. Values higher than 0.5 in the PC analysis were interpreted as contributing to the PCs.

2.8. *Calculations of emission factors according to the IPCC*

The N_2O fluxes were estimated according to the methodology presented by the IPCC (Houghton et al. 1996). In this procedure, emission factors from Nykarn were calculated according to the following formula:

$$N_2O\text{-}N_{\text{from fresh water system}} = N_{\text{leached to fresh water system}} \times (EF_{6\text{-}r}) \quad (2)$$

where $N_{\text{leached to fresh water system}}$ is nitrogen lost through leaching and runoff and $EF_{6\text{-}r}$ is the emission factor. Total nitrogen was used for calculating the emission factors. The suggested default factor derived by the IPCC from fresh water systems loaded with nitrogen is 0.75% (Houghton et al., 1996).

3. Results

3.1. *Nitrous oxide fluxes during 1998 and 1999*

An overview of the daily mean nitrous oxide exchange rates for the two growing seasons studied (1998 and 1999) is given in Fig. 2 and Table 1. The large temporal and spatial variation in nitrous oxide fluxes covered a range of –350 to 1791 μg N_2O m^{-2} h^{-1}, i.e.

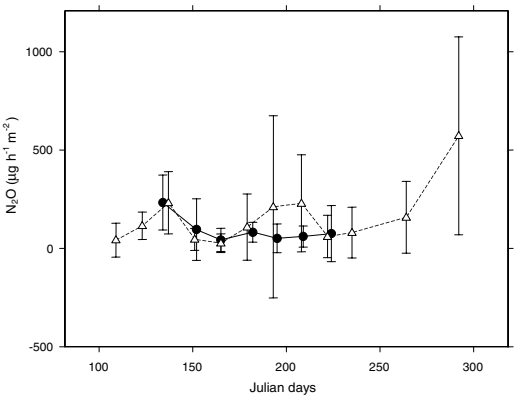

Fig. 2. Average nitrous oxide fluxes for the sampling dates at the constructed wetland at Nykvarn during 1998 (•) and 1999 (Δ).

both consumption and emission occurred. There was no statistical difference in rates ($p = 0.51$) between the two years analysed for the periods corresponding to the 1998 campaign. Consumption of atmospheric N_2O was observed for about 15% of all measurements, as shown in the daily means presented in Fig. 3. The highest emission rates were observed in October 1999 and the highest consumption rate in the middle of July 1998 (Figs. 2 and 3). The average N_2O flux for the two years, including all frames, was 130 μg N_2O m^{-2} h^{-1} (SD = 220). However, since the measuring period in 1998 was shorter and covered the summer mainly, the average of 1999 (147 μg N_2O m^{-2} h^{-1}) is likely a better descriptor for the growing season. The 1999 average for the period corresponding to the 1998 data was 114 μg N_2O m^{-2} h^{-1} and did not differ from the 1998 average rate.

The average N_2O emissions were significantly higher from wastewater inlet pond 1 (frames 1–9) compared to both ponds 2 and 3 (frames 10–15; $p < 0.0005$; Table 1). No statistically significant difference in emission rates was detected between ponds 2 (frames 10–12) and 3 (frames 13–15; $p = 0.14$). According to the 1999 results the fluxes increased in early spring (between Julian days 110–140) from 42 to 232 μg N_2O m^{-2} h^{-1}($p < 0.001$; Fig. 2). They then declined, paralleling changes seen in the same measurement period in 1998. The rates during the periods of decline were similar for the two seasons. The 1998 emission rates stayed at this lower level until the termination of the measurement series in August. During the 1999 summer period, the average N_2O emission

Table 1. *Nitrous oxide emissions; mean, median and range, during 1998 and 1999 in the constructed wetland at Nykvarn, Linköping, Sweden*

Pond	Frame/ Vegetation	Number of samples	Mean ± s.d.	Median	Range
			μg N_2O m^{-2} h^{-1}		
1	1–9	138	185 ± 271	99.0	−350–1791
2	10–12	53	68.2 ± 79.1	56.9	−68.7–336
3	13–15	50	41.9 ± 63.9	24.8	−122–199
All	All	241	130 ± 220	71.8	−350–1791
All	T. latifolia	134	160 ± 268	80.5	−350–1791
All	P. arundinacea	11	248 ± 225	152	27.5–703
All	Spirogyra sp.	56	63.6 ± 82.0	36.0	−123–336
All	G. maxima	32	49.5 ± 65.9	31.0	−46.4–199
All	L. minor	3	94.4 ± 76.8	94.4	40.1–149
All	Frames with no plants	5	248 ± 173	192	−6.1–499

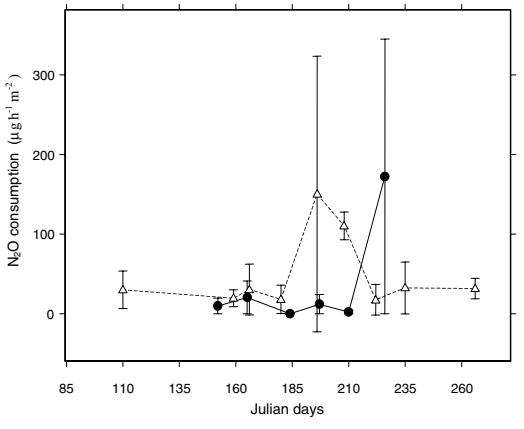

Fig. 3. Average nitrous oxide consumption rates in the constructed wetland at Nykvarn during 1998 (•) and 1999 (Δ).

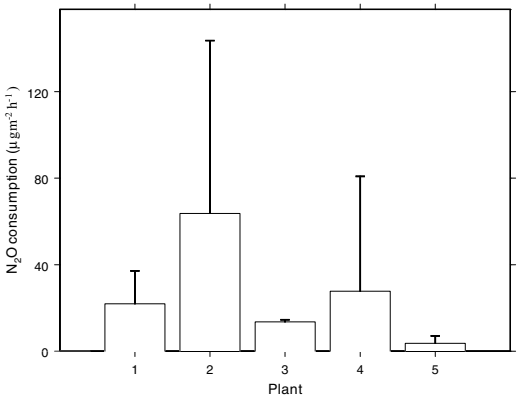

Fig. 4. N_2O consumption from areas dominated by *G. maxima* (1), *T. latifolia* (2), *P. arundinacea* (3), *Spirogyra* sp. (4) and frames with no plants (5) in the Nykvarn wetland.

rates increased to a plateau of ca. 200 μg N_2O m^{-2} h^{-1} during Julian days 190–210, but declined to the 1998 level in August. Later in the year, there was a trend of increasing fluxes at the beginning of the autumn, which grew progressively stronger to result in a statistically significant increase in N_2O emissions, from 108 to 503 μg N_2O m^{-2} h^{-1}, between Julian days 235–300. A decrease in atmospheric N_2O levels occurred in the chambers on several occasions during the flux measurements in all frames except for nos. 1 and 2. The consumption rates observed ranged from 8–350, 1–69 and 1–123 μg N_2O m^{-2} h^{-1} for A1, 2 and 3, respectively (Fig. 3). The corresponding averages were 70, 16 and 21 μg N_2O m^{-2} h^{-1}, respectively, and the average for the whole set of N_2O consumption rates was 39 μg N_2O m^{-2} h^{-1}. No statistically significant difference in the N_2O consumption rates was found between the two years (Fig. 4). However, similarly to the total emission rate patterns, there were significant differences in N_2O consumption rates among the different ponds. Thus, the N_2O consumption in A1 was significantly higher compared to the rates in A2 and A3 ($p < 0.01$). No significant differences were found between ponds A2 and A3. Figure 5 shows N_2O consumption versus the nitrate concentration. There was a negative linear correlation ($r^2 = 0.52$) between N_2O consumption and nitrate concentration, which ranged from 1–6 mg L^{-1} (Fig. 5).

The N_2O fluxes in Nykvarn were related differently to the types of plants in the frames. The strength of

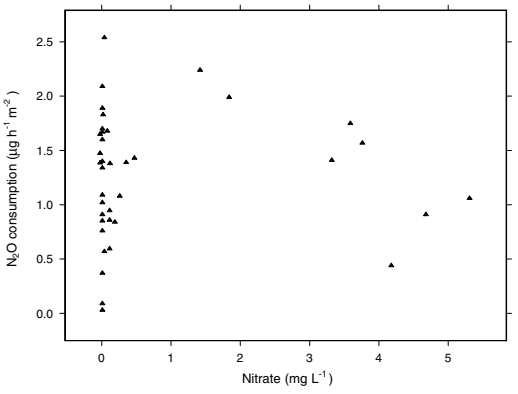

Fig. 5. N_2O consumption versus nitrate concentration measured in the constructed wetland at Nykvarn.

the fluxes (median values) associated with the different species was as follows: *P. arundinacea* > *L. minor* > *T. latifolia* > *Spirogyra* sp. > *G. maxima* (Table 1). Significant differences in N_2O fluxes were found between *P. arundiancea* and *Spirogyra* sp. and *G. maxima* ($p < 0.001$) dominated frames. There were also significant differences between frames with *T. latifolia* and *Spirogyra* sp. and *G. maxima* ($p < 0.05$). The flux rates of frames with no plants differed statistically from those with *T. latifolia*, *Spirogyra* sp. and *G. maxima* ($p < 0.01$). The frames with no plants showed N_2O flux rates in the same range as for *P. arundinacea*.

N_2O consumption was observed at frames dominated by *T. latifolia, Spirogyra* sp. and *G. maxima* (Table 1). The strength of the consumption (median values) associated with the different species was as follows: *Spirogyra* sp > *G. maxima* > *T. latifolia.*

T. latifolia was associated with 19 such observations, whereas consumption was observed in frames covered by *Spirogyra* sp., *G. maxima* and "frames with no plants" on ten, eight and two occasions, respectively. The two N_2O consumption observations from "frames with no plants" showed the lowest N_2O consumption. The N_2O consumption was observed only once in a frame covered by *P. arundinacea.*

3.2. Environmental parameters and derivation of principal components

Means, medians and ranges of the environmental parameters measured during every gas-sampling occasion are presented in Table 2. Mutual correlations were common among the environmental parameters (data not shown). Therefore, principal components (PCs) were derived to analyse the relationships among the parameters. In the following section PCs are

Table 2. *Environmental parameters; minimum, maximum, mean and median during 1998 and 1999 in the constructed wetland at Nykvarn, Linköping, Sweden*

Parameter	Minimum	Maximum	Mean	Median
Sediment temperature (°C)	4.6	20.5	13.9	14.6
Water temperature (°C)	5.2	27.8	15.3	15.8
Air temperature (°C)	3.7	26.5	17.1	17.9
RH in[a] (%)	36	100	72	70
RH out[a] (%)	39	100	69	69
Water level (cm)	0	28.5	14.5	16.5
CH_4 flux (mgd^{-1} m^{-2})	0	1259	169	82
NH_4^+ ($mg\ L^{-1}$)	0.007	30.7	4.0	2.0
NO_2^- ($mg\ L^{-1}$)	0.01	1.0	0.1	0.02
NO_3^- ($mg\ L^{-1}$)	0.006	24.0	3.9	1.4
N^b_{min} ($mg\ L^{-1}$)	0.01	23.9	4.0	3.1
P^c_{tot} ($mg\ L^{-1}$)	0.002	39.8	1.5	0.2
Veg. volume[d] (L)	0	40	5	3
PAR[e] ($\mu E\ m^{-2}\ s^{-1}$)	105	1697	951	964

[a]RH, relative humidity inside and outside the chamber.
[b]N_{min}, mineralized nitrogen ($NH_4^+ + NO_-^2 + NO_3^{-2}$).
[c]P_{tot}, total phosphorous.
[d]Veg. volume, vegetation volume.
[e]PAR, photosynthetically active radiation.

presented for 1998 and 1999 N_2O flux data both singly and combined.

Four PCs, explaining 71% of the total variance among the parameters, were extracted by principal component analysis (PCA) for the combined data of 1998 and 1999 (PCs 1–4; Table 3). PC 1 was positively correlated to relative humidity inside and outside the chamber and negatively correlated to both air temperature and PAR (Table 3). It was therefore considered to describe abiotic factors and interpret them as the *chamber effect*. PC 2 was positively correlated to sediment, water and air temperatures, vegetation volume and methane flux (Table 3). It was therefore considered to describe temperature effects on emissions and/or anoxia. Thus, this principal component was interpreted as the *gas activity effect*. The third PC was positively correlated to NH_4^+, water level, mineralised nitrogen (i.e. the total mineral N) and total phosphorus. This PC was considered to describe general mineralisation and accordingly interpreted as the *mineralisation effect*. PC 4 was positively correlated to nitrite, nitrate and mineralised nitrogen and was therefore regarded as describing denitrification, i.e. the *denitrification effect*.

Three PCs were derived from the PCA on the 1998 data set. They explained 69% of the total variance among the parameters (PCs 5–7; Table 3). PC 5 was positively correlated to nitrite, ammonium and sediment temperature and, thus, was considered to describe nitrification activity i.e. the *nitrification effect*. PC 6 was correlated positively to water temperature, water levels, nitrate, mineralised nitrogen and methane. Anoxia and denitrification are likely connected in this set, so this PC describes the *denitrification effect* like PC 4 (see above). PC 7 was positively correlated to water level, vegetation volume and total phosphorus and was regarded as describing vegetation distribution. Thus, PC 7 was interpreted as the *vegetation effect*.

The PCA on the 1999 data resulted in four PCs explaining 75% of the total variance among the parameters (PCs 8–11; Table 3). As for PC 7, generated from the 1998 PCA, PC 8 was positively correlated to sediment and water temperatures, and vegetation volume. Thus, it is linked to the vegetation distribution and, like PC 7, it is interpreted as the *vegetation effect*. PC 9 was negatively correlated to PAR and positively correlated to relative moisture inside and outside the chamber, so it resembles PC 1 and describes *chamber effects*. The positive correlation of PC 10 to total ammonium, total phosphorus and mineralised nitrogen led to it being considered a *mineralisation effect* in accordance with PC 3. The final PC, no. 11, like PCs 4 and 6, was positively correlated to nitrate, nitrite and water level. This PC was regarded as describing denitrification, i.e. the *denitrification effect*.

Five PCs (PCs 12–17; Table 4) were derived from the N_2O consumption data. These PCs were similar to the PCs described above (i.e. PCs 1, 3, 7 and 11) except for PC 13, which was positively correlated only to the temperature of the sediment, water and air. Therefore, PC 13 was interpreted as the *temperature effect*.

3.3. *Multiple linear regression analysis of N_2O emission*

The PCs derived from the analysis described above were used in a stepwise multiple linear regression analysis to describe the variation in the N_2O fluxes. Again, the statistical analyses were based on the three sets of data (i.e. 1998 and 1999 N_2O flux data sets, singly and combined) used for the above PCAs.

When PCs 1–4 (Table 3) were regressed against the log-transformed N_2O flux values of the combined 1998–99 data set, PCs 3 and 4 (representing the mineralisation and denitrification effects, respectively) were found to have significant influence ($p < 0.05$; Tables 3 and 5). They explained 13% of the flux variations, and 16% if the consumption rates were excluded. Subjecting the 1998 data to the same procedure showed that PCs 5 and 6 (the nitrification and denitrification effects) were also significant, explaining 17% of the N_2O fluxes, and 26% when only emission data were used. For the 1999 data set the corresponding values were 17 and 28% for the whole set and the emissions only, respectively (Table 5). As for the combined data set analyses, the mineralisation and denitrification effects (PCs 10 & 11) were significant contributors. Thus, the vegetation and chamber effects did not seem to affect the measured N_2O fluxes.

A stepwise linear regression analysis based solely on N_2O consumption rates was also undertaken (Tables 2 and 4). The stepwise multiple regression analysis with PCs 12–16 showed that PC 16 (the *vegetation effect*; Table 4) could explain 43% of the variation in atmospheric N_2O consumption rates.

3.4. *Estimation of emission factors based on the Nykvarn wetland study*

The N_2O fluxes in 1998 and 1999 were used to calculate emission factors in each pond (A1–A3) of the

Table 3. *Principal components (PC) of the different variables of the N_2O flux data matrixes 1998+1999, 1998 and 1999*[a]

	1998+1999				1998			1999			
Variable	PC 1 Chamber effect	PC 2 Gas activity effect	PC 3 Mineralisation effect	PC 4 Denitrification effect	PC 5 Nitrification effect	PC 6 Denitrification effect	PC 7 Vegetation effect	PC 8 Vegetation effect	PC 9 Chamber effect	PC 10 Mineralisation effect	PC 11 Denitrification effect
Sediment temperature	0.048	0.911	−0.094	−0.057	0.638	0.213	0.279	0.905	−0.050	0.059	−0.159
Water temperature	−0.275	0.825	−0.058	0.051	−0.014	0.608	−0.050	0.829	−0.316	0.078	0.017
Air temperature (T_{air})	−0.607	0.571	−0.231	−0.276				0.631	−0.553	−0.222	−0.289
Photosynthetic radiation (PAR)	−0.790	−0.113	0.207	−0.059				−0.019	−0.769	−0.049	0.215
Relative moisture inside the chamber (rh_i)	0.886	−0.019	0.108	−0.091				−0.019	0.913	0.033	0.062
Relative moisture outside the chamber (rh_o)	0.884	−0.044	−0.026	−0.120				−0.077	0.850	0.040	−0.212
Water level	−0.114	0.051	0.653	−0.096	0.120	0.429	0.825	0.308	0.147	0.126	0.589
CH_4 flux	0.212	0.666	0.148	0.271	0.330	0.752	0.242	0.550	−0.026	0.433	−0.136
NH4	−0.031	−0.158	0.864	0.154	0.727	0.241	−0.089	−0.013	0.048	0.932	0.194
NO2	−0.094	0.130	0.026	0.866	0.865	−0.031	−0.051	−0.099	−0.383	0.140	0.769
NO3	0.019	−0.057	0.082	0.859	0.437	0.770	0.077	−0.331	−0.227	0.025	0.803
Mineralized nitrogen (N_{min})	−0.026	−0.163	0.840	0.383	0.699	0.647	0.005	−0.721	0.026	0.905	0.335
Total phosphorus (P_{tot})	0.173	0.287	0.632	−0.126	−0.098	0.149	0.743	0.217	0.059	0.789	−0.138
Vegetation volume	0.079	0.505	0.360	−0.367	0.086	−0.374	0.770	0.696	0.271	−0.046	0.208
Variance explained (% of total)	22.0	19.1	17.7	12.6	39.1	18.6	11.1	23.4	21.4	20.8	9.0

[a]Rotation method: Varimax with Kaiser normalization, rotation converged in seven iterations. Values above 0.5 are marked in bold.

Table 4. *Principal components (PC) of the different variables of the N_2O consumption data matrix 1998+1999*[a]

Variable	PC 12 Chamber effect	PC 13 Temp. effect	PC 14 Denitrification effect	PC 15 Mineralisation effect	PC 16 Vegetation effect
Sediment temperature	0.231	0.878	0.135	−0.284	0.082
Water temperature	−0.006	0.904	0.173	−0.187	0.111
Air temperature (T_{air})	−0.392	0.797	−0.273	−0.114	0.097
Photosynthetic radiation (PAR)	−0.827	−0.211	−0.324	0.010	−0.070
Relative moisture inside the chamber (rh_i)	0.908	−0.202	0.176	−0.060	0.156
Relative moisture outside the chamber (rh_o)	0.924	−0.019	−0.033	0.020	−0.105
Water level	−0.158	0.168	0.204	0.217	0.809
CH_4flux	0.343	0.338	0.670	−0.026	−0.174
NH4	−0.019	−0.236	−0.047	0.952	0.058
NO2	0.107	0.029	0.912	0.108	−0.001
NO3	0.085	−0.085	0.944	0.072	−0.067
Mineralized nitrogen (N_{min})	0.006	−0.247	0.226	0.926	0.037
Total phosphorous (P_{tot})	0.103	0.202	−0.170	0.100	0.759
Vegetation volume	0.118	−0.163	−0.219	−0.303	0.667
Variance explained (% of total)	25.5	23.6	14.8	13.5	7.3

[a]Rotation method: Varimax with Kaiser normalization, rotation converged in seven iterations. Values above 0.5 are marked in bold.

Table 5. *Seven models derived from stepwise multiple regression of N_2O fluxes data vs. derived principal components for 1998 and 1999*[a]

Model[b]/Data set	Equation	R^2_{adj}
1998+1999	$y = 3.27 + 1.01$ Denitrification effect $+ 0.72$ Mineralization effect $+\in$	0.13
only positive N_2O fluxes	$y = 1.91 + 0.11$ Denitrification effect $+ 0.19$ Mineralization effect $+\in$	0.16
1998[c]	$y = 1.45 + 0.43$ Nitrification effect $+ 0.25$ Denitrification effect $+\in$	0.17
Only positive N_2O fluxes[c]	$y = 1.78 + 0.23$ Nitrification effect $+ 0.17$ Denitrification effect $+\in$	0.26
1999	$y = 1.44 + 0.50$ Dentrification effect $+ 0.28$ Mineralization effect $+\in$	0.17
Only positive N_2O fluxes	$y = 1.93 + 0.27$ Dentrification effect $+ 0.11$ Mineralization effect $+\in$	0.28
1998+1999 N_2O consumption	$y = -1.25$–0.37 Vegetation effect $+\in$	0.43

[a]$\in$ = residuals for each model.
[b]Significance level is 5%.
[c]Relative moisture, PAR and air temperature were excluded in the model.

constructed wetland. These emission factors were calculated in two different ways. First, the ratio between N_2O fluxes and nitrogen coming into the constructed wetland was calculated and then the ratio between N_2O fluxes and nitrogen reduction (nitrogen coming into the wetland minus nitrogen going out) in the wetland was derived. Thus, the N_2O fluxes are both based on averages of N_2O fluxes in 1998 and 1999, but they have different denominators. The calculated emission factors ranged from 0. 2 to 2.7 g N_2O-N per kg N. Regardless of which of the two different methods of calculation was used, the emission factors decreased by ca. 80% from pond A1 to pond A3. There was no statistical difference between the emission factors calculated between 1998 and 1999 ($p = 0.42$), and no correlation was found between the emission factors

and mineralised nitrogen based on calculated seasonal and weekly averages.

4. Discussion

4.1. N_2O fluxes during 1998 and 1999

In Table 6 the nitrous oxide fluxes from Nykvarn are compared with fluxes from similar, nutrient-rich wetland systems and sewage treatment plants. Only two previous studies seem to have analysed N_2O fluxes in constructed wetlands treating sewage wastewater (Table 6), reporting figures in the range -4.2–470 μg N_2O m^{-2} h^{-1} (Fey et al., 1999; Gui et al., 2000). The N_2O fluxes measured at Nykvarn showed considerably higher flux rates, in the range -350–1800 μg N_2O m^{-2} h^{-1}. However, the measurements by Fey et al. (1999), taken at two constructed wetlands in Friedelhausen in Germany (each about half the size of the ponds in Nykvarn) covered the winter period from November untill March, which may explain their lower fluxes (0–470 μg N_2O m^{-2} h^{-1}). The soil temperatures during their measuring campaign ranged from below zero to 13 °C. In contrast, Gui et al. (2000) measured N_2O fluxes (-4.2–196 μg N_2O m^{-2} h^{-1}) from June to December in a full-scale system (60 000 m^3 d^{-1}) at water temperatures ranging from 5–28.5 °C. At Nykvarn the measurements were conducted at the same temperature range of the sediments and water, 4.6 and 27.8 °C, as for the study of Gui et al., but emissions were one order of magnitude higher. Thus, the higher N_2O emission and consumption in Nykvarn compared to the other two studies cannot be explained by temperature differences. It should also be noted that there were similar trends, with higher emissions during the autumn in both the Chinese (Gui et al., 2000) and Swedish studies, and in both cases consumption occurred mainly during the summer. Thus, there are indications of similarities in the regulation of nitrous oxide exchange in the three areas, even though the ranges of the fluxes

Table 6. *Literature data of N_2O flux data from wetlands with and without sewage water input and sewage treatment plants*[a]

Habitat	Location	N_2O fluxes (μg N_2O m^{-2} h^{-1})	N_2O-N/N load (%)	Number of measurements	Reference
Constructed wetlands treating sewage water	Sweden	$-3.5x10^2$– 1.8×10^3	0.02–0.27	241	This study
	Germany	$(39–4.7) \times 10^2$ [a]	0.11	152	Fey et al., 1999
	China	$(-4.2–2.0) \times 10^2$	–	7	Gui et al., 2000
Other nutrient-rich constructed wetlands	Wales	0–9.9 [b]	0.53 [f]	350	Freeman et al., 1997
	Denmark	$(9.2–4.1) \times 10^2$		120	Paludan et al., 1996
	Wales	0–33 [b]	–	14	Dowrick et al., 1999
	USA	10 [k]	–	750	Delaune et al., 1990
	Germany	$(0–5.9) \times 10^2$	–	40	Augustin et al., 1998
	Germany	$(-13–2.8) \times 10^2$	–	44	Merbach et al., 1996
Sewage treatment plants	USA	5.0×10^3– 7.5×10^4 [c]	–	29	Czepiel et al., 1995
	USA	4.2×10^2–1.7×10^3 [d]	–	15	Czepiel et al., 1995
	USA	$27–2.7 \times 10^2$ [j]	0.3–0.6 [h]	37	Smith et al., 1981
	Japan	[e]	0.01–0.08	20	Kimochi et al., 1998
	Germany	6.2×10^3 [k]	0.001	?	Sümer et al., 1995

[a]Data show the average or the range of data. Winter measurements.
[b]Recalculated from data, dry and wet communities.
[c]Aerated tanks.
[d]Non-aerated tanks.
[e](0–2.4) $\times 10^4$ μg N_2O m^{-3} h^{-1}, depth of basins unknown.
[f]NO_3^- only.
[g]Only NO_3^- measurements.
[h]Only NH_4^+ measurements, pilot experiment.
[i]Peatlands for forestry and agriculture.
[j]Unlimed.
[k]Arithmetical mean.

differed. However, neither Gui et al. (2000) nor Fey et al. (1999) reported e.g. spatial variation in the fluxes they observed, so it is difficult to assess the differences that they claim to have occurred over the measured seasons.

There have been several investigations into N_2O exchange between wetlands and the atmosphere in catchment areas, such as peatlands and marshes (Martikainen et al. 1993; Delanue et al., 1990; Augustin et al., 1998; Dowrick, 1999) and in riparian areas of ponds (Merbach et al., 1996; cf. Table 6). The rates reported for these studies range from consumption of atmospheric nitrous oxide (only in the study by Merbach et al. 1996) at 13 μg N_2O m^{-2} h^{-1} to emissions of 590 μg N_2O m^{-2} h^{-1} (Augustin et al., 1998; Table 6). Thus, both consumption and emission rates were below those observed at the Nykvarn wetland. Consumption was observed only once in the study of Merbach et al., in late August, in a series of measurements taken every second week between May and October at a weakly eutrophic pond shore where the water table was 10 cm below the soil surface.

In order to place the emissions from Nykvarn and other wetlands into a broader perspective they were compared with N_2O emissions from sewage treatment plants (Table 6). Consumption of atmospheric N_2O does not seem to have been observed in any case in such systems, but emission rates up to 75 000 μg N_2O m^{-2} h^{-1}have been measured (Czepiel et al., 1995). In summary, the positive N_2O fluxes seen in the present study were ca. three to ten times higher than in previous studies of constructed wetlands treating wastewater (although substantially lower than the highest emissions from sewage treatment plants), whereas the consumption rate was approximately 80 times higher. The N_2O fluxes from Nykvarn were three to 180 times higher compared to other wetland systems. No consumption rates for N_2O were reported for these systems.

4.2. *Influence of different parameters on the N_2O exchange patterns*

The stepwise linear regression models described 13–28% of the N_2O flux variation (Table 5). Generally, better model fits were found for emissions only. This is probably because the processes involved in production and consumption are different and, thus, their modes of regulation differ (Conrad, 1996). Three PCs were involved in the regression outcome: i.e. the Mineralisation (PCs 3 and 10), Nitrification (PC 5) and Denitrification (PCs 4, 6 and 11) effects, all of which contain components known to affect nitrous oxide formation. The mineralisation effect was strongly coupled to the ammonia content and the total concentration of mineral nitrogen in the water. The nitrification had strong relations to ammonia, nitrite and total minerlised nitrogen, while the denitrification PC were characterised by nitrite nitrate and total mineralised nitrogen. Hence, the parameters expected to influence the nitrous oxide formation by nitrification and denitrification were those governing the PCs responding in the regressions.

It is not possible to distinguish plant-mediated N_2O emissions from N_2O released from the sediment in the Nykvarn study, since we used chambers that covered both vegetation and sediment. However, the principal component analyses for 1998 and 1999 distinguished PCs related to vegetation parameters such as vegetation volume, PAR and air moisture, which affected the N_2O fluxes. These PCs explained 23 and 11% of the variance, respectively, but they were not recognised as parameters in the multiple linear regression analysis done using all the PCs. This may be due to the rough method of determining the amount of plant biomass in the different frames. Despite this, there were some clear trends between the nitrous oxide fluxes and the vegetation. The highest N_2O emissions came from the *P.s arundinacea* L. area, followed by areas with *L. minor*, *T. latifolia* L., *Spirogyra* sp. and *G. maxima*. The highest N_2O consumption was found from the "moderately emitting areas", especially *T. latifolia* followed by *G. maxima* and *Spirogyra* sp. areas. Thus, the plant habitats emitting N_2O at relatively high rates showed weak consumption of N_2O and vice versa. The reason for this is not known, but there is strong evidence from other studies suggesting that the types and strength of interactions between plants and gases differ substantially among different soil systems (Joabsson et al., 1999).

Another relevant result is the high uptake of nitrous oxide and that the N_2O consumption at Nykvarn took place during the summer period, when the nitrate concentrations measured were mostly low. It is not surprising that we found high N_2O consumption in this system, because there are bacteria that are adopted to use N_2O for their metabolism. N_2O is also very soluble in water (in contrast to CH_4), which makes the gas available for the bacteria. Calculations according to Fick's first law show that consumption at this rate is not limited by the diffusion of N_2O from the atmosphere in the chamber into the water column within the frame. A majority of the N_2O consumption data

(78%) occurred when nitrate concentrations were below 0.5 mg L^{-1} (Fig. 5). There was also a negative linear correlation ($r^2 = 0.52$) between N_2O consumption and nitrate concentration, ranging from 1–6 mg L^{-1} (Fig. 5). Thus, consumption may be linked to shortage of electron acceptors for the denitrifying bacteria (i.e. nitrate deficiency) and, hence, their use of N_2O as a substitute. The ability of denitrifiers to use nitrous oxide has long been recognised (Payne, 1981). The low nitrate concentration occurring during the summer period is likely due not only to plant and microbial uptake of nitrogen for growth, but also to dissimilative nitrate reduction. According to the data shown in Fig. 5, nitrate seems to affect the consumption rate when this electron acceptor reaches concentrations greater than 0.5 mg L^{-1}. Below this threshold other factors seemed to govern the rate of N_2O consumption, as shown by the large variation in rates at nitrate concentrations below the detection level (i.e. <0.010 mg L^{-1}). One explanation to the large variation in N_2O flux rates at this low nitrate concentration is that the measurement samples may reflect both aerobic and anaerobic conditions. Thus, during anaerobic conditions N_2O is consumed, while during aerobic conditions, consumption of N_2O is low because of high availability of nitrate from by nitrification of ammonia.

4.3. Emission factors from Nykvarn and other studies compared to the default factor derived by the IPCC

Table 6 shows the ratio of N_2O fluxes versus total nitrogen load. The ratio derived from the present study is much lower compared to that observed in several other cases, in a wastewater-treating wetland in Germany, for example, which is a horizontal-flow system and somewhat smaller than the Nykvarn wetland. The difference in results could be due to differences in sediment and vegetation properties or hydraulic regimes. Many studies have not provided information on incoming nitrogen levels, which makes it impossible to calculate emission ratios. The suggested default factor derived by the IPCC from fresh water systems loaded with sewage nitrogen is 0.75% (Houghton et al., 1996). In the present study, emission factors of 0.02–0.27% were calculated from the N_2O flux data, thus being considerably lower than the estimated IPCC default factor. Another relevant result is that the emission factors calculated from Nykvarn decreased from pond A1 to A3. Therefore, our data suggest that the default factor derived by the IPCC is not appropriate for this type of aquatic system. First, as described above, the default factor calculated by IPCC seems to be overestimated. Second, the default factor is static, i.e. independent of type of wetland system and important parameters in the wetland. It is worth mentioning that neither our study nor the IPCC's determination of a default factor includes any winter measurements. This may underestimate the default value as well as the emission factors from Nykvarn. However, we can draw the conclusion from this study that the emission factor has a dynamic response to the nutrient status in the water, and therefore, it is not suitable to use a static calculation. It seems inappropriate to apply a general emission factor to all constructed wetland systems. The shift of the emission factor found in this study (through the ponds) suggests that the factors should be adjusted to account for differences in important influences such as nitrogen load and vegetation type.

The constructed wetland in Nykvarn was, as described above, designed as a pilot system to reduce nitrogen coming from a sewage plant. The results from five years' operation (1993–1998) showed that a reduction of 2000 kg N ha^{-1} yr^{-1} with a nitrogen load of 3500–4000 kg N ha^{-1} $yr^{-1}$was possible (Tonderski and Hansson, 2001). These results were obtained using partially nitrified waters from the Åbylund sewage plant. In our study we show that the reduction of nitrogen partly occurred through N_2O emission to the atmosphere. Although the emission rates were high compared to other ecosystems (Table 6), their contribution to the nitrogen budget for the wetland was low. The rest of the nitrogen in the budget was either accumulated in the plants or transported to the atmosphere as gaseous nitrogen loss (N_2). Based on the average flux, ca. 80 μg N_2O–N h^{-1} m^{-2}, about 7 kg of nitrogen may be released from the Nykvarn wetland per year in this manner, corresponding to 0.4% of the estimated overall reduction. This figure is similar to the overall reduction (0.53%; Table 6) for a constructed wetland treating dairy sewage water as described by Freeman et al. (1997).

5. Acknowledgements

This work was part of a project funded by Tekniska verken i Linköping AB. We also thank Peter Doersch, Department of Soil and Water Sciences, Agricultural University of Norway, for analysing N_2O fluxes during parts of 1998 and Annika Nielsen, Department of Water and Environmental Studies, Linköping, for assistance with nitrogen and phosphorus analyses.

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