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Coregulation of nitrous oxide emissions by nitrogen and temperature in China's third largest freshwater lake (Lake Taihu)

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Abstract

Nitrous oxide (N_2O) is a potent greenhouse gas and contributes to the loss of stratospheric ozone. However, the role of inland waterbodies in the dynamics of atmospheric N_2O is poorly understood. We investigated N_2O fluxes and their controlling factors in Lake Taihu, a large and shallow (2400 km^2, 1.9 m depth) eutrophic lake in eastern China. Long-term measurements (2011–2016) revealed spatial and temporal variations in the lake surface N_2O fluxes. The mean N_2O flux from the lake was 3.5 ± 1.8 (mean ± SD) μmol m^{-2} d^{-1}, with an annual N_2O budget of 134.4 ± 69.8 Mg (10^6 g) yr^{-1}. The highest N_2O fluxes occurred in the eutrophic zone with significant anthropogenic N inputs, and the lowest fluxes occurred in the noneutrophic zone with no external N inflow. A seasonal pattern in N_2O fluxes was observed only in the noneutrophic zone and was strongly correlated with water temperature. No seasonality in the N_2O fluxes was observed in the eutrophic zone with high N concentrations in the water, indicating that N concentrations play a dominant role in regulating N_2O fluxes compared to water temperature. The average N_2O emission factor in Lake Taihu was 0.18%, with temporal and spatial variations negatively associated with N concentration but positively associated with the mass ratio of dissolved organic carbon to dissolved inorganic nitrogen. Our results suggest that anthropogenic activities strongly affect N_2O fluxes in freshwater lakes.

Nitrous oxide (N_2O) is a potent greenhouse gas with a radiative forcing of 0.17 W m^{-2}, ranking third among the long-lived greenhouse gases in the atmosphere (IPCC 2013). At the same time, N_2O plays a major role in ozone depletion in the stratosphere (Ravishankara et al. 2009). There has been a steady increase in atmospheric N_2O at a rate of 0.7–0.8 ppb yr^{-1} over the past three decades (Davidson 2009; Saikawa et al. 2014). The growing atmospheric concentration of N_2O and its biological sources from the biosphere have received considerable attention, and a number of studies has been carried out to investigate the terrestrial sources of N_2O, such as natural soils (Chapuis-Lardy et al. 2007; Xu et al. 2008), synthetic fertilizers (Davidson 2009; Saikawa et al. 2014; Griffis et al. 2017), and wastewater treatment (Kampschreur et al. 2009; Foley et al. 2010).

Inland freshwater systems remain understudied in terms of their N_2O budget due to their high heterogeneity (Ivens et al. 2011; Borges et al. 2015). Inland waters can receive large N loadings from human activities, depending particularly on farming practices (Seitzinger and Kroeze 1998; Ivens et al. 2011; McCrackin and Elser 2011; Borges et al. 2018). N_2O is an intermediate product of denitrification, the microbial reduction of nitrate (NO_3^--N) to nitrite (NO_2^--N), nitric oxide (NO), N_2O, and N_2, and is a byproduct of nitrification, the microbial oxidation of ammonium (NH_4^+-N) to NO_2^--N and NO_3^--N. These microbial processes can significantly occur in lakes (Seitzinger et al. 2006), making lakes a strong source of atmospheric N_2O. The relatively long-residence time of lakes makes these waterbodies more effective than rivers in N removal and associated N_2O production (Mulholland et al. 2008; Harrison et al. 2009). Thus, a more accurate estimation of N_2O emissions from inland lakes is fundamental for quantifying an N_2O budget at a regional scale.

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Additional Supporting Information may be found in the online version of this article.

Previous studies on N_2O dynamics in freshwater lakes largely focus on boreal lakes (Mengis et al. 1996; Huttunen et al. 2003; Miettinen et al. 2015; Soued et al. 2016; Klaus et al. 2017). Also, lake eutrophication and the associated decrease in the biomass of submerged macrophytes are widespread (Sinha et al. 2017; Zhang et al. 2017), but the effects of eutrophication and submerged macrophyte coverage on N_2O fluxes remain uncertain. An increasing trend of N_2O emissions from oligotrophic to eutrophic lakes was found as a result of increasing external N loading (Mengis et al. 1997; Huttunen et al. 2003; Wang et al. 2007; Whitfield et al. 2011). This finding is in contrast to an ecosystem-scale experiment which reported a neutral response of N_2O fluxes to N enrichment in unproductive boreal lakes (Klaus et al. 2017). It has been reported that submerged macrophytes influenced lake N cycling and were significantly correlated with variability in N_2O fluxes (Davidson et al. 2015; Zhu et al. 2015). However, a number of studies have reported that submerged macrophytes do not enhance lake N removal and the associated N_2O production (Liu et al. 2017; Yao et al. 2017). Considering these contrasting results, large uncertainties remain on how eutrophication and macrophytes affect N_2O emissions from freshwater lakes.

The Intergovernmental Panel on Climate Change (IPCC) guidelines on national greenhouse gas inventories recognize that a fraction of N is converted to N_2O from N fertilizer leaching and runoff from agriculture soils. Such substance-based N_2O fluxes are defined as indirect emissions. The indirect N_2O emission factor (EF) allows for the estimation of N_2O emissions based on information of the external N inputs or background N concentrations in a system (De Klein et al. 2006). Global and regional N_2O emissions from a waterbody can be approximated by multiplying the nitrogen fertilizer input or relevant anthropogenic N loading with a predetermined EF (De Klein et al. 2006; Ivens et al. 2011; McCrackin and Elser 2011; Hu et al. 2016; Hama-Aziz et al. 2017; Fu et al. 2018). However, it should be noted that the unitless EF value can vary by an order of magnitude (Beaulieu et al. 2011; McCrackin and Elser 2011; Turner et al. 2015; Hama-Aziz et al. 2017). For example, the current default EF for rivers is 0.0025 (De Klein et al. 2006), but studies suggest that this value can be either underestimated (Beaulieu et al. 2011; Yu et al. 2013; Turner et al. 2015; Fu et al. 2018) or overestimated (Outram and Hiscock 2012; Hama-Aziz et al. 2017). In general, the EF estimates for lakes are poorly understood due to the limited number of studies in these complex ecosystems (McCrackin and Elser 2011; Outram and Hiscock 2012). More EF measurements are needed to better estimate indirect N_2O budgets (McCrackin and Elser 2011).

In this study, we report the results of field experiments on N_2O fluxes from Lake Taihu, a typical subtropical eutrophic lake in the Yangtze River Delta, China. The lake has a complex river network with 117 rivers or channels draining into the lake (Xu et al. 2010). The northern and western portions of the lake experience severe eutrophication due to pollution discharge by inflow rivers (Qin et al. 2007). Submerged vegetation dominates much of the eastern portion of the lake, with an average macrophyte biomass of 3.8 kg m^{-2} (Qin et al. 2007; Luo et al. 2016). The effects of anthropogenic N inputs and the presence of macrophytes on the lake water quality have been well documented (Qin et al. 2007; Duan et al. 2009; Xu et al. 2010); however, their roles in N_2O emissions remain poorly investigated.

The objectives of this study are as follows: (1) to characterize the spatiotemporal variations in N_2O fluxes across the entire Lake Taihu; (2) to investigate the control of biological, chemical, and physical processes on the N_2O fluxes; and (3) to quantify the N_2O contribution from Lake Taihu to the national N_2O budget.

Materials and methods

Study area

Measurements were made in Lake Taihu (30°05′–32°08′N, 119°08′–121°55′E) in eastern China. Lake Taihu is the third largest freshwater lake in China, with a surface area of 2400 km^2 and a mean depth of 1.9 m. The lake catchment covers approximately 36,500 km^2, with a complicated river network surrounded by several large cities. Inflow comes from the northern and western sides of the lake, and outflow occurs on the eastern side of the lake (Fig. 1). The local climate is characterized by high-water temperatures (monthly mean: 31°C) and rain (monthly mean: 170 mm) in the summer and low temperatures and rain in the winter (monthly mean water temperature: 3.9°C; monthly mean rain: 60 mm; Xu et al. 2010; Xiao et al. 2017). The annual precipitation is approximately 1100 mm (Lee et al. 2014; Wang et al. 2014). The anthropogenic N inputs through inflow were 2–4 times higher than the N loss via river outflow (Supporting Information Table S1 and Fig. S1). High concentrations of dissolved organic carbon (DOC) occur in the eutrophic zone due to anthropogenic inputs via river discharge and in the macrophyte-dominated zone due to carbon accumulation via primary production (Lee et al. 2014). Major pollutant sources, including industrial activities, domestic sewage, and agricultural fertilizers, are present in the catchment (Qin et al. 2007; Duan et al. 2009).

We divided Lake Taihu into three zones according to the geographic differences in vegetation and nutrient concentrations (Fig. 1). Zone 1 is the most eutrophic part of the lake, with elevated nutrient loads from river inputs, including the hypereutrophic northwest zone and Meiliang Bay. Zone 3 is the main outlet of the lake and is characterized by dense submerged vegetation (mainly *Hydrilla verticillata* and *Potamogeton malaianus*). Zone 2 has lower eutrophication and represents a transitional area (Fig. 1). This is seen in the typical mean dissolved oxygen (DO) concentrations of 6.94 mg L^{-1}, 10.03 mg L^{-1}, and 10.72 mg L^{-1} for zone 1, zone 2, and zone 3, respectively (Xiao et al. 2017), and in the average macrophyte coverage of 3.8 kg m^{-2} in zone 3 and close to zero in the other zones (Qin et al. 2007; Luo et al. 2016). The spatial contrast in

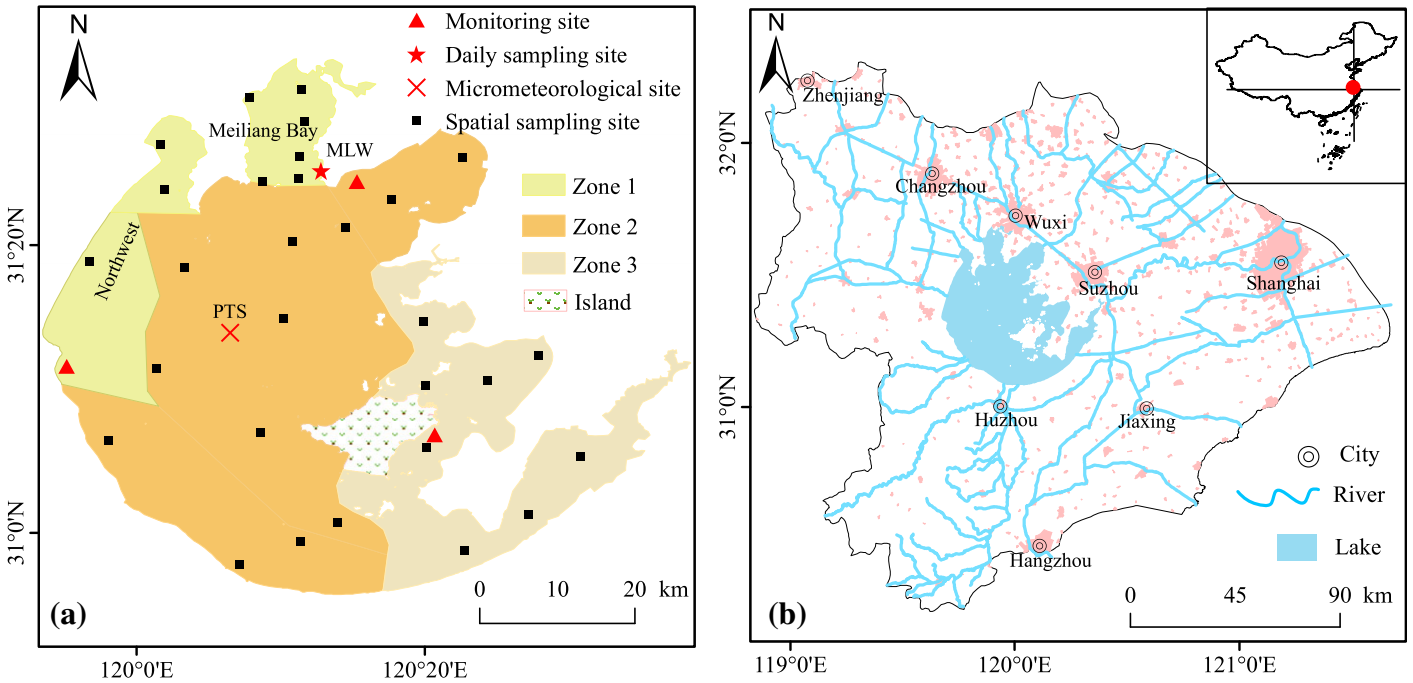

Fig. 1. Geographic location of Lake Taihu and the sampling sites; (**a**) The location of the spatial sampling sites and three biological zones at Lake Taihu: black squares, spatial sampling sites; red star, daily sampling site; red cross, PTS (Pingtaishan) lake micrometeorological site; red triangles, in situ high-frequency measurements by the China National Environmental Monitoring Center (http://www.zhb.gov.cn/hjzl/shj/dbszdczb/). Zone 1: eutrophic zone, including the Northwest Zone and Meiliang Bay; Zone 2: transition zone; Zone 3: Submerged macrophyte zone. (**b**) Lake Taihu watershed and surrounding rivers. Water flows from the north and west to the east and south.

the total nitrogen (TN) concentrations of zone 1, zone 2, and zone 3, with values of 2.58 mg L^{-1}, 1.89 mg L^{-1}, and 1.07 mg L^{-1}, respectively (Xiao et al. 2017), also supports this finding.

N_2O lake surveys

Lake water samples were collected in 300 mL glass bottles to analyze the spatial-temporal variability in N_2O concentrations. The sampling procedures have previously been reported in detail (Xiao et al. 2017). Briefly, bubble-free lake water was collected at the desired depth below the lake surface using a custom-made sampler sealed with a rubber cork. The cork was removed from the opening by remote control when the sampler reached the desired depth, allowing water to enter the sampler. The water was then poured quickly into a 300 mL glass bottle, and the bottle was immediately capped without headspace using a butyl rubber stopper and sealed with a sealing membrane. Both the glass bottle and sampler were washed with local bubble-free lake water before collection.

Surface lake water samples were collected daily at a depth of 20 cm at midday from 01 August 2011 to 31 December 2016 at a fixed sample site 200 m from the shore in Meiliang Bay at the northern part of the lake (the MLW (Meiliangwan) site, 31°24′N, 120°13′E, Fig. 1). A total of 1980 water samples were collected at the MLW site to explore the temporal variation in N_2O. Diel sampling was also conducted at the MLW site to investigate diel variations in the N_2O fluxes. Surface-water samples were collected every 3 h for three consecutive days in each month from August 2011 to July 2013. Water samples were also collected at depths of 20 cm, 50 cm, 100 cm, and 150 cm at 00:00 h and 12:00 h every day during diel sampling to explore the vertical profiles of dissolved N_2O concentrations.

Whole-lake surveys were conducted seasonally from 2012 to 2016 in mid-February, mid-May, mid-August, and mid-November. In each whole-lake survey, surface-water samples were collected at a depth of 20 cm from each of the 29 sampling locations (Fig. 1). Each of these included 9 sampling sites in zone 1, 12 in zone 2, and 8 in zone 3. The sampling sites in each zone were evenly spatially distributed. Over the 5-yr sampling period, a total of 580 water samples were collected from zone 1 (n = 180), zone 2 (n = 240) and zone 3 (n = 160) to explore the spatial variability in N_2O concentrations and fluxes. Each whole-lake survey was completed in two consecutive days to reduce the biases caused by day-to-day variations and by one person to reduce systems bias.

Dissolved N_2O concentrations in the collected water samples were analyzed using the headspace equilibration method (Mengis et al. 1996; Davidson et al. 2015). Ultrahigh purity N_2 gas (99.999%) was injected into the glass bottle to create a 100-mL headspace. The glass bottle was then shaken vigorously for 5 min to allow the dissolved N_2O gas to reach equilibrium within the headspace. A small air sample (20 mL) was drawn from the headspace using a syringe with a three-way stopcock and was injected into a gas chromatograph (Model

Agilent GC7890B, Agilent, California, U.S.A.) equipped with an electronic capture detector for N_2O detection.

N_2O flux calculation

The N_2O fluxes (F_n, μmol m^{-2} d^{-1}; a positive value indicates N_2O emission from the water to atmosphere) at the lake–air interface were calculated using the transfer coefficient method based on the bulk diffusion model (Eq. 1; Cole and Caraco 1998):

$$F_n = k \times (C_w\text{-}C_{eq}) \quad (1)$$

where C_w is the dissolved N_2O concentration (μmol m^{-3}) in the surface water (top 20 cm), as measured by gas chromatography, and C_{eq} is the N_2O concentration in water in equilibrium with the atmosphere at the in situ temperature. k is the gas transfer coefficient (m d^{-1}) and was normalized to a Schmidt number of 600 (Eq. 2):

$$k/k_{600} = (S_c/S_{c600})^{-n} \quad (2)$$

where S_c is the Schmidt number of a given gas at a given temperature and water density, S_{c600} is the Schmidt number 600 at a temperature of 20°C, n is a constant determined by measured wind speed, and k_{600} is the gas transfer coefficient adjusted to Schmidt number 600 calculated by wind speed (Cole and Caraco 1998):

$$k_{600} = 2.07 + 0.215 \times U_{10}^{1.7} \quad (3)$$

where U_{10} is the wind speed at a 10-m height. The hourly wind speed measured at the PTS site, a micrometeorological site located in the center of Lake Taihu (Fig. 1; Lee et al. 2014), was used to calculate the gas transfer coefficient. Although there are a number of formulations considering both wind shear and waterside convection reported for the k_{600} calculation (e.g., MacIntyre et al. 2010), our previous study confirmed that the gas transfer coefficient in the large lake was mostly driven by wind speed (Xiao et al. 2017), which is consistent with Read et al. (2012). There were no significant differences between k values calculated by the wind-dependent model described by Cole and Caraco (1998) and those calculated by models considering both wind shear and waterside convection in the lake (Xiao et al. 2017).

N_2O emission factor calculation

EF was calculated with two different approaches (Outram and Hiscock 2012; Hama-Aziz et al. 2017). The first method, herein known as EF_a for the whole lake, was computed by dividing the annual N_2O fluxes from the lake by the total amount of N loading in 1 yr, following the IPCC approach for calculating national N_2O inventories. The second method, defined as EF_b, was calculated by using N_2O-N/NO_3^--N mass ratios (McCrackin and Elser 2011; Outram and Hiscock 2012; Turner et al. 2015; Hama-Aziz et al. 2017; Fu et al. 2018). The EF_a was calculated for the whole lake, without information on the spatiotemporal variation in N processes. Most reported EFs for aquatic systems are computed with the second method (Outram and Hiscock 2012; Turner et al. 2015; Hama-Aziz et al. 2017).

Auxiliary data

Hourly micrometeorological measurements at the PTS site included wind speed, wind direction, water temperature, air temperature, relative humidity, and four-way net radiation components (Lee et al. 2014; Xiao et al. 2017). Triplicate water samples were collected at the micrometeorological site for N_2O measurements during every site visit. During each survey, the physical, biological, and chemical properties (water temperature, pH, specific conductance [Spc], oxidation reduction potential [ORP], and DO concentration) were measured in situ with a multiparameter probe (YSI 650MDS, YSI, Yellow Springs, Ohio, U.S.A.) at a depth of 20 cm to be consistent with the water sampling. The YSI parameters were calibrated before each survey to reduce the uncertainty. It should be noted that water temperature profile measurements were conducted hourly at the MLW site (Lee et al. 2014).

The concentrations of nutrients (TN, total phosphorus [TP], ammonium, nitrate, and nitrite), DOC, and chlorophyll *a* (Chl *a*) were analyzed at each of the 29 sampling locations during the whole-lake surveys. Surface-water samples were collected from each spatial sampling site using an organic glass hydrophore and stored in acid-washed plastic bottles. Samples were preserved in ice-chilled coolers and transported to the laboratory for filtration and measurements. The concentrations of TN and TP were measured using combined persulfate digestion (Ebina et al. 1983). Ammonium (NH_4^+-N) was determined by the indophenol blue method with a spectrophotometer at 640 nm, and nitrate (NO_3^--N) and nitrite (NO_2^--N) were determined with the cadmium reduction method with a spectrophotometer at 543 nm (American Public Health Association 1995). The DOC concentrations were analyzed after filtration through precombusted 47 mm GF/F filters (porosity 0.7 μm) with a Shimadzu TOC-5000A analyzer (Duan et al. 2014). The Chl *a* was extracted with 90% hot ethanol and concentrations were determined spectrophotometrically at 665 nm and 750 nm using a Shimadzu spectrophotometer.

Data analysis

Simple linear regression and multilinear stepwise regression were carried out to find correlations between the environmental variables and N_2O fluxes. The zonal mean N_2O fluxes and environmental variables were used for temporal analysis. A zonal mean was calculated for each lake survey using all measurements within the corresponding zone. In total, there were 20 data sets for temporal correlation analysis. The flux of each lake zone was computed as the mean of all calculated fluxes in

the same zone; then, the whole-lake N_2O fluxes were computed as an area-weighted zonal flux.

A least significant difference post hoc test was conducted to examine the differences among measured variables using SPSS (version 18.0). Differences at the $p < 0.05$ level were deemed statistically significant.

Results

Environmental condition

There was no substantial spatial variation in water temperature across Lake Taihu (Xiao et al. 2017); the temperature variation was < 0.6°C between different zones (Wang et al. 2014). The water temperature also showed no vertical profile variation from the surface to the bottom of Lake Taihu (Supporting Information Fig. S2a,b), indicating that the lake was well mixed due to its shallow depth. There was a clear seasonality; the annual mean water temperature at a 20-cm depth was 17.9°C, with the highest water temperature in August (37.3°C) and the lowest in January (0.1°C). The annual mean wind speed was 4.5 m s^{-1}, with no significant spatial variation across the open water (Xiao et al. 2017).

Lake Taihu showed strong spatial variation in water chemistry (Table 1; Fig. 2). The highest concentrations of NH_4^+-N (1.75 mg L^{-1}), NO_3^--N (1.24 mg L^{-1}), NO_2^--N (0.19 mg L^{-1}), TN (5.25 mg L^{-1}), and TP (0.29 mg L^{-1}) occurred in zone 1 and were associated with discharge from inflowing rivers. Zone 1 had significantly ($p < 0.05$) higher nutrient concentrations than zone 2 and zone 3. In contrast, the lowest DO and pH were generally found in zone 1 (Fig. 2), and there were no significant differences among the three zones ($p > 0.05$). The spatial variations of various forms of N were highly intercorrelated (Fig. 2); for example, the NO_2^--N concentration was highly correlated with NO_3^--N ($r = 0.88$, $p < 0.01$) and NH_4^+-N ($r = 0.99$, $p < 0.01$) across the lake. The mean Chl *a* concentration in zone 1 (35.74 μg L^{-1}) was also significantly ($p < 0.05$) higher than that in zones 2 and 3.

Some environmental variables showed seasonal variations among the three zones (Fig. 3). The DO varied seasonally, with the peak occurring in winter (Fig. 3b). The seasonal variation in Chl *a* was consistent with water temperature (Fig. 3a,c), with significantly ($p < 0.05$) higher concentrations in summer than in winter for the three zones. In contrast, NH_4^+-N and NO_3^--N concentrations were higher in winter (Fig. 3d,e). It should be noted that zone 1 had the highest Chl *a* and nutrient concentrations over the study period.

Spatial and temporal variations in N_2O concentrations and fluxes

Sampling at different water depths at MLW from September 2011 to July 2013 indicated that, in general, dissolved N_2O concentrations were remarkably uniform on the vertical profiles (Supporting Information Fig. S2c,d). On the contrary, N_2O concentrations in the surface waters varied dramatically based on the total 20 whole-lake surveys from 2012 to 2016 (Supporting Information Fig. S3). Surface dissolved N_2O concentrations varied within wide ranges of 6.8–129.9 nmol L^{-1} in spring, 6.0–91.6 nmol L^{-1} in summer, 4.5–133.8 nmol L^{-1} in autumn, and 5.9–99.7 nmol L^{-1} in winter. The average seasonal N_2O concentrations in the lake were 15.5 nmol L^{-1}, 12.1 nmol L^{-1}, 14.8 nmol L^{-1}, and 17.4 nmol L^{-1}, in spring, summer, fall, and winter, respectively.

The N_2O fluxes showed large spatial variations across the lake (Figs. 4, 5). The N_2O fluxes in zone 1, with an annual mean value of 11.4 μmol m^{-2} d^{-1}, were significantly ($p < 0.01$) higher than those in zones 2 and 3 (~1.3 μmol m^{-2} d^{-1}), while there was no significant difference ($p = 0.87$) in these fluxes between zone 2 and zone 3. The spatial N_2O fluxes ranged between 0.9 μmol m^{-2} d^{-1} and 58.7 μmol m^{-2} d^{-1} in spring, −0.1 μmol m^{-2} d^{-1} and 62.5 μmol m^{-2} d^{-1} in summer, −1.5 μmol m^{-2} d^{-1} and 55.6 μmol m^{-2} d^{-1} in autumn, and −1.7 μmol m^{-2} d^{-1} and 39.1 μmol m^{-2} d^{-1} in winter, showing nonsignificant ($p > 0.05$) differences among the seasons. The amplitude (maximum minus minimum) of the N_2O fluxes across the whole lake in summer (62.6 μmol m^{-2} d^{-1}) was larger than that in winter (40.8 μmol m^{-2} d^{-1}).

The N_2O fluxes at the water–air interface showed strong temporal variation over the whole lake (Fig. 5) and at the MLW site (Fig. 6). During whole-lake sampling, the highest N_2O flux was found in summer and the lowest in winter, but the differences between seasons were nonsignificant ($p > 0.05$)

Table 1. Annual mean nutrient concentration and Chl *a* concentration across three zones in Lake Taihu from 2012 to 2015.

Zone	Type	Statistics	NH_4^+-N (μmol L^{-1})	NO_3^--N (μmol L^{-1})	NO_2^--N (μmol L^{-1})	TN (μmol L^{-1})	TP (mg L^{-1})	DOC (mg L^{-1})	Chl *a* (μg L^{-1})	DOC : DIN
Zone 1	Eutrophic	Mean ± SD	54 ± 31	53 ± 31	4.95 ± 2.00	232 ± 54	0.17 ± 0.06	4.74 ± 0.75	35.74 ± 28.75	6.36 ± 4.19
		Range	50–122	16–101	3.01–11.05	161–319	0.12–0.32	3.77–5.88	7.96–109.98	1.68–14.17
Zone 2	Transitional	Mean ± SD	24 ± 11	39 ± 24	1.17 ± 0.66	152 ± 45	0.10 ± 0.02	3.84 ± 0.84	20.19 ± 11.11	7.03 ± 6.88
		Range	8–48	11–77	0.38–2.71	99–206	0.07–0.14	2.51–5.33	8.14–46.45	2.01–12.61
Zone 3	Macrophytes	Mean ± SD	18 ± 9	19 ± 16	0.54 ± 0.26	92 ± 32	0.05 ± 0.02	3.67 ± 0.81	8.88 ± 3.29	9.64 ± 4.42
		Range	6–38	4–49	0.18–0.99	55–148	0.03–0.10	2.52–5.20	4.56–16.50	3.31–16.72

Chl *a*, chlorophyll *a* concentration; DOC, dissolved organic carbon; DOC : DIN, the mass ratio of DOC (mg L^{-1}) to DIN (dissolved inorganic nitrogen, mg L^{-1}); TN, total nitrogen concentration; TP, total phosphorus concentration.

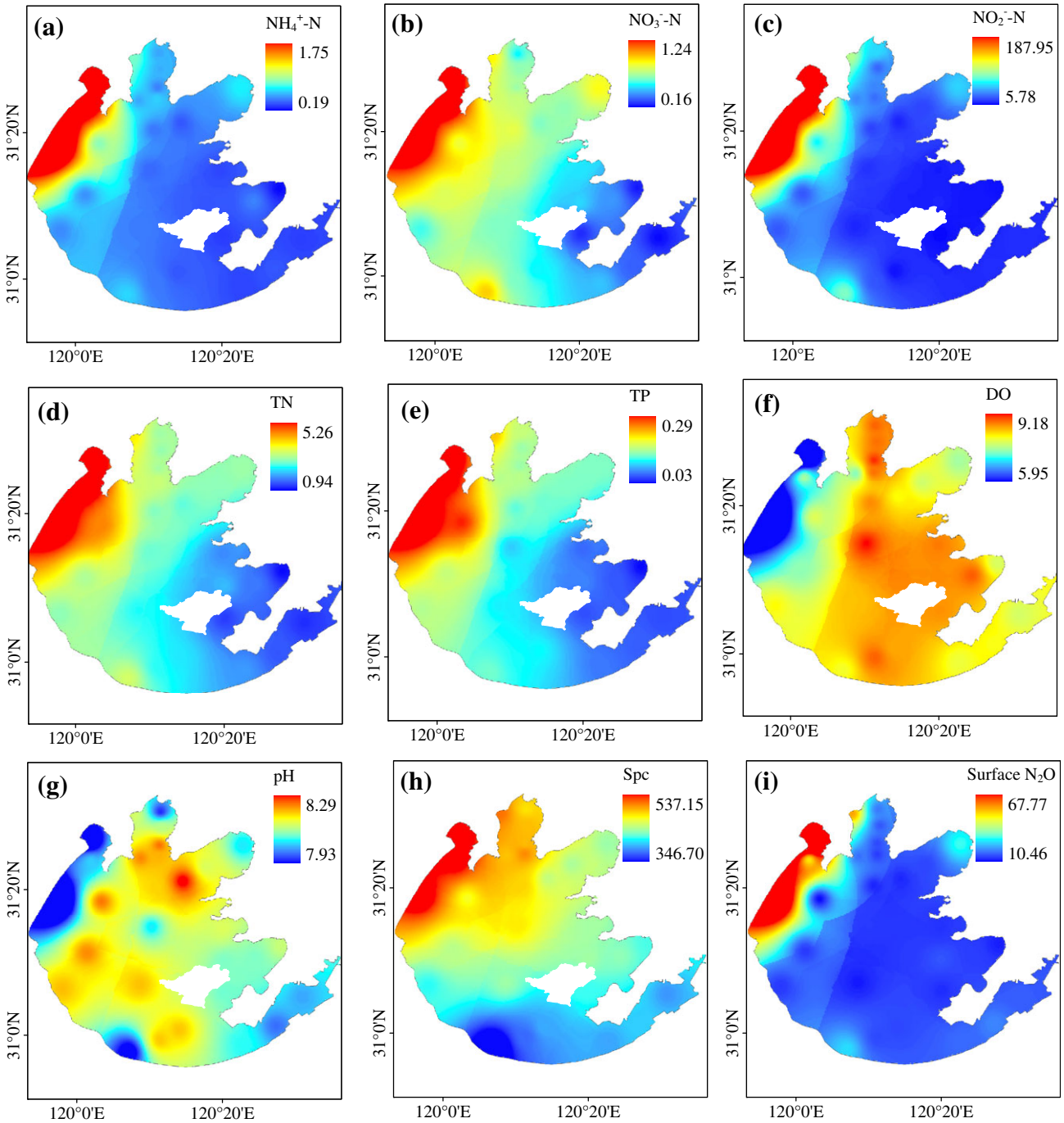

Fig. 2. Spatial variation in N loading, water quality indices, and dissolved N_2O concentration across Lake Taihu during the sampling period: (**a**) NH_4^+-N (mg L^{-1}), (**b**) NO_3^--N (mg L^{-1}), (**c**) NO_2^--N (μg L^{-1}), (**d**) TN (mg L^{-1}), (**e**) TP (mg L^{-1}), (**f**) DO (mg L^{-1}), (**g**) pH, (**h**) specific conductance (Spc, μs cm^{-1}), and (**i**) surface dissolved N_2O concentration (nmol L^{-1}).

due to high-temporal variability. From MLW daily sampling, the average winter (December, January, and February) flux was 0.4 μmol m^{-2} d^{-1}, approximately 14 times lower than the summer (June, July, and August) flux (5.1 μmol m^{-2} d^{-1}), with a statistically significant difference ($p < 0.01$). Approximately 30% of the sampled water at the MLW site was undersaturated in terms of N_2O, most often occurring in winter.

Multiple-year measurements showed a substantial interannual variability in the whole-lake N_2O fluxes. The largest N_2O fluxes occurred in 2012, with a mean value of 5.5 μmol m^{-2} d^{-1}, and the lowest N_2O fluxes occurred in 2016, with a mean value of 1.1 μmol m^{-2} d^{-1}. The most substantial interannual variability in N_2O fluxes was observed in zone 1, which showed a sharp decrease in annual fluxes from 18.1 μmol

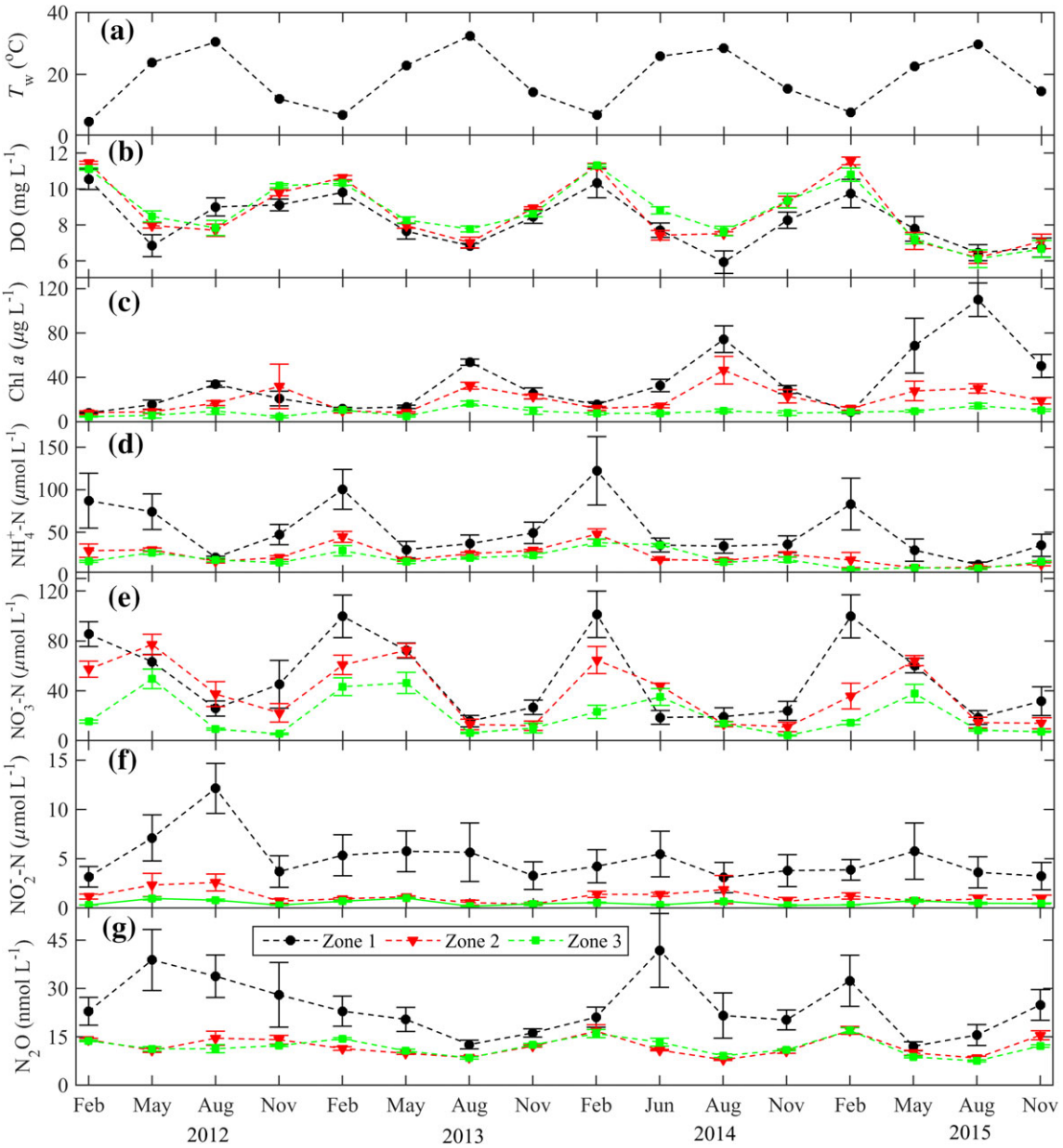

Fig. 3. Seasonal variation in (**a**) water temperature (T_w), (**b**) DO, (**c**) Chl *a* concentration, (**d**) NH_4^+-N, (**e**) NO_3^--N, (**f**) NO_2^--N, and (**g**) surface dissolved N_2O concentration in the three zones. Note that the water temperature was the monthly mean value obtained from the MLW site (Lee et al. 2014). Error bars indicate one standard error.

m^{-2} d^{-1} in 2012 to 3.6 μmol m^{-2} d^{-1} in 2016 (Fig. 4; Table 2). The N_2O fluxes in zones 2 and 3 also showed substantial interannual variability (Table 2) and decreased from 2.4 μmol m^{-2} d^{-1} in 2012 to 0.4 μmol m^{-2} d^{-1} in 2016 (zone 2) and from 1.3 μmol m^{-2} d^{-1} in 2012 to 0.6 μmol m^{-2} d^{-1} in 2016 (zone 3). However, as a whole, these interannual variability trends were nonsignificant over the study period (zone 1: $p = 0.13$; zone 2: $p = 0.37$; zone 3: $p = 0.22$; whole lake: $p = 0.13$). The annual mean N_2O flux was 3.5 ± 1.8 μmol m^{-2} d^{-1} based on long-term measurements (Table 2), with an annual N_2O budget of 134.4 ± 69.8 Mg (10^6 g) yr^{-1}.

Diurnal sampling showed statistically insignificant diel variations in N_2O fluxes (Supporting Information Fig. S4). In cold seasons, the hourly N_2O fluxes varied within relatively narrow ranges, showing nonsignificant ($p = 0.99$) differences between daytime fluxes and nighttime fluxes. In warm seasons, the hourly N_2O fluxes varied within wide ranges, for example, ranging between 5.5 μmol m^{-2} d^{-1} and 21.9 μmol m^{-2} d^{-1} in June 2012, also leading to nonsignificant ($p = 0.84$) differences between daytime fluxes and nighttime fluxes. Interestingly, the annual mean daytime flux (2.9 ± 0.2 μmol m^{-2} d^{-1}) was the same as the annual mean nighttime flux (2.9 ± 0.1 μmol

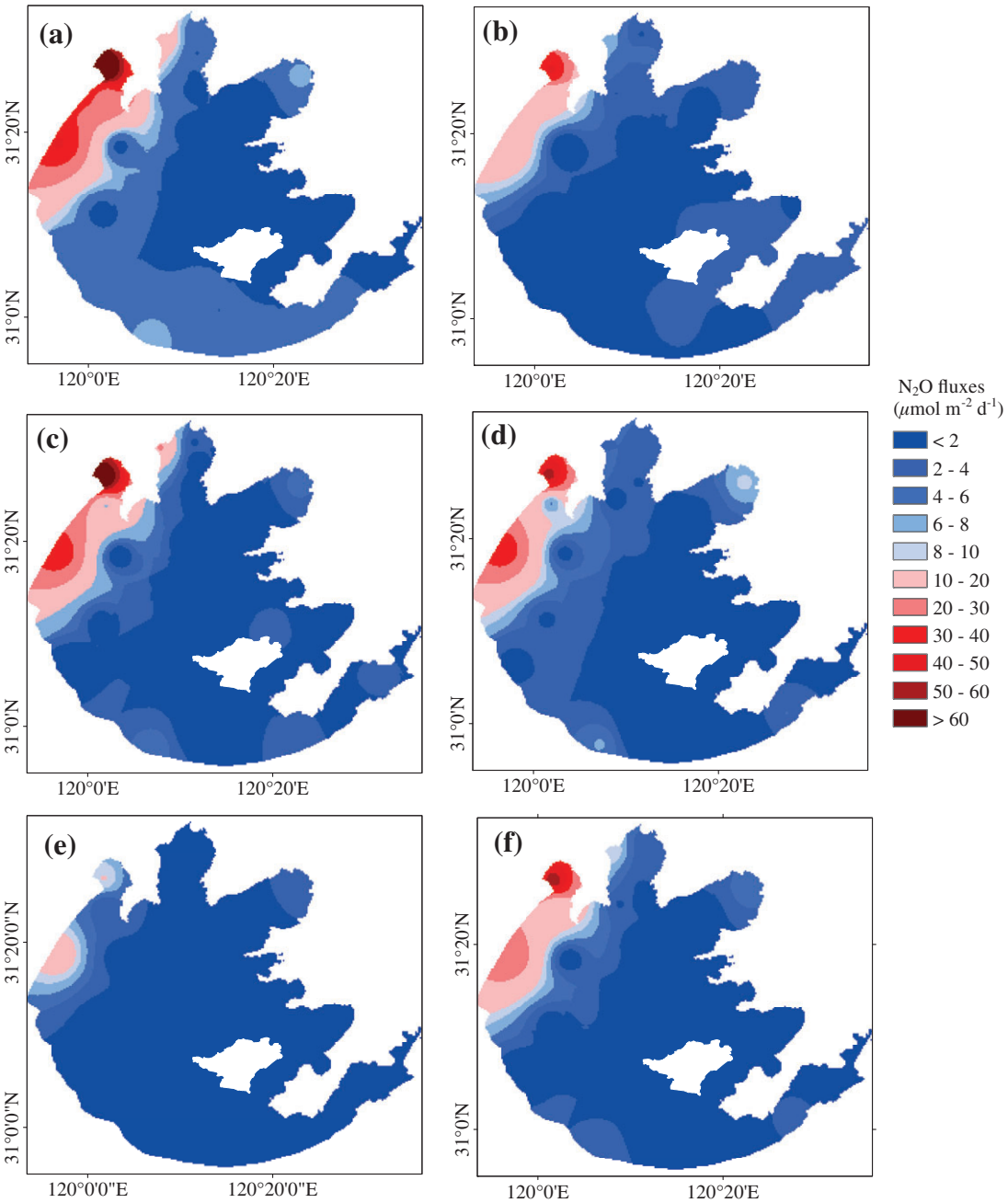

Fig. 4. Spatial variation in the annual mean N_2O fluxes during the sampling period: (**a**) 2012, (**b**) 2013, (**c**) 2014, (**d**) 2015, (**e**) 2016, and (**f**) mean value during 2012–2016.

m^{-2} d^{-1}, Supporting Information Fig. S4) during the sampling period. Hence, we did not consider diel variations when computing N_2O budgets in the following sections.

Factors influencing N_2O fluxes

There were statistically significant linear correlations between the water temperature and N_2O fluxes for zone 2 ($r = 0.49$; $p = 0.028$), zone 3 ($r = 0.60$; $p = 0.005$), and the whole lake ($p = 0.015$). However, no significant correlation was found between the water temperature and N_2O fluxes within zone 1 (Table 3). No relationship was found between the wind speed and whole-lake mean N_2O fluxes ($p = 0.80$). Daily sampling at MLW from August 2011 to December 2016 also confirmed the influence of water temperature on N_2O fluxes ($r = 0.34$, $p < 0.01$; Supporting Information Fig. S5).

The variabilities in N_2O fluxes were correlated with several environmental variables (Fig. 7; Table 4). Based on the continuous whole-lake survey over space and time across the nutrient gradient, the N_2O fluxes were positively correlated with NO_2^--N ($r = 0.68$, $p < 0.01$, $n = 460$), NO_3^--N ($r = 0.30$, $p < 0.01$,

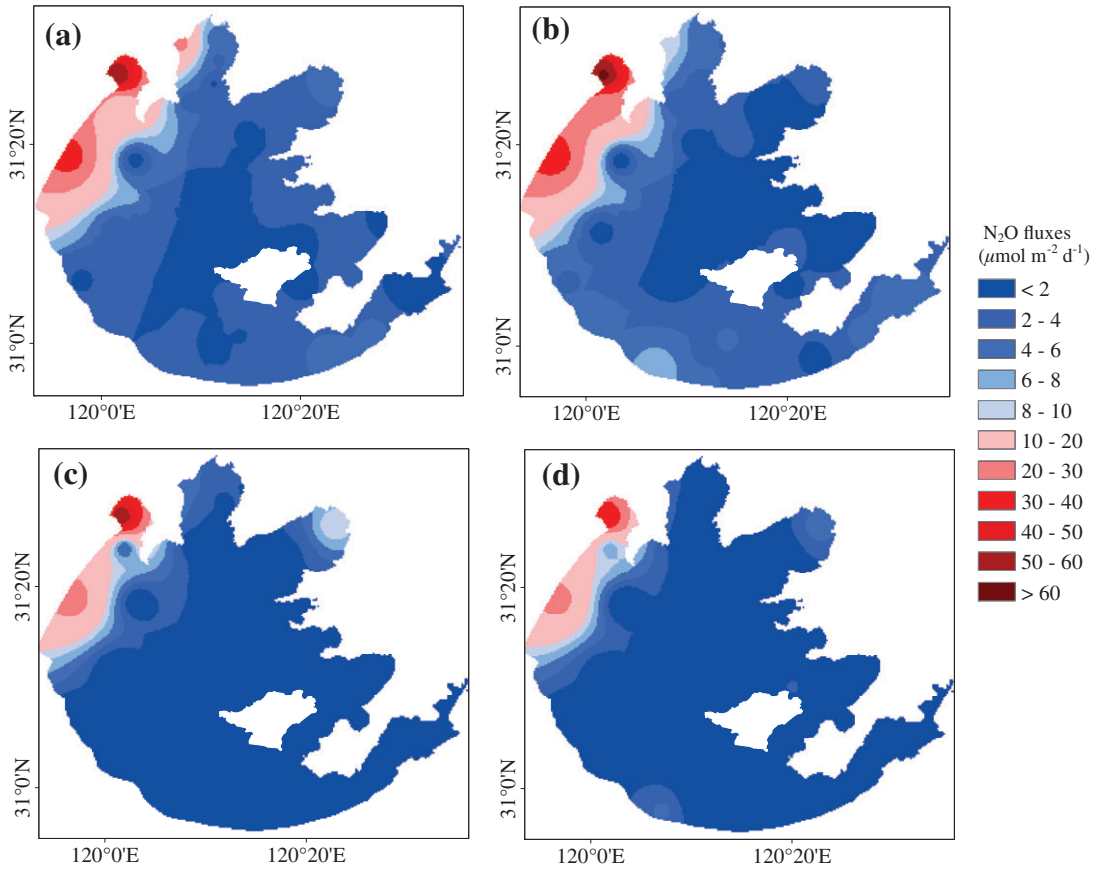

Fig. 5. Spatial variation in the N_2O fluxes in (**a**) spring (May), (**b**) summer (August), (**c**) autumn (November), and (**d**) winter (February). Data shown as the mean value during 2012–2016.

n = 460), NH_4^+-N (r = 0.45, p < 0.01, n = 460, n = 448), TN (r = 0.46, p < 0.01, n = 448), TP (r = 0.43, p < 0.01, n = 448), and DOC (r = 0.23, p < 0.01, n = 448) but negatively correlated with DO (r = –0.34, p < 0.01, n = 448) and pH (r = –0.14, p < 0.01, n = 448). No significant correlations were found between the N_2O fluxes and Chl *a* nor between the

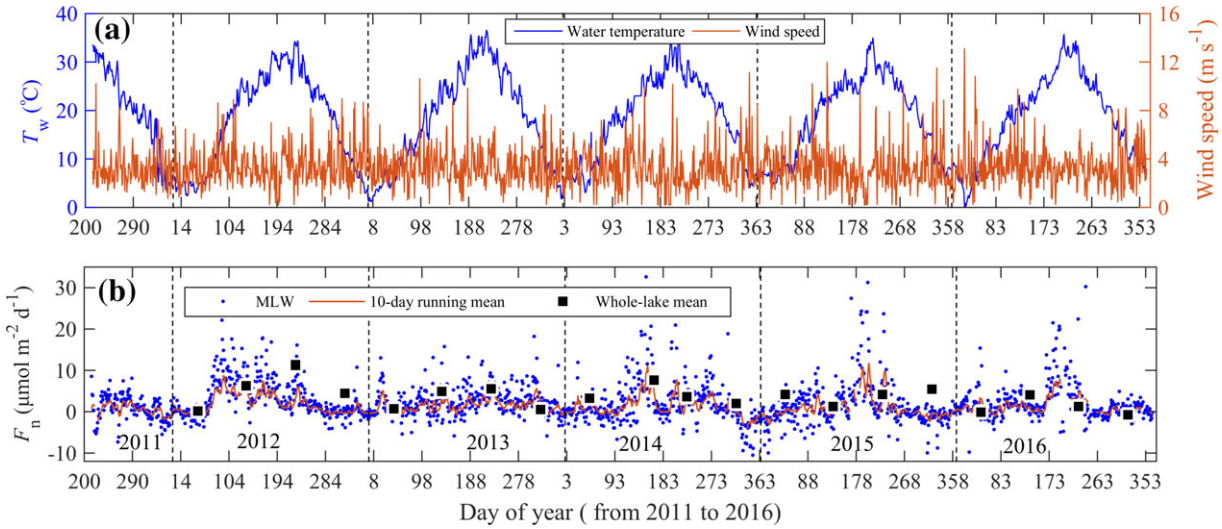

Fig. 6. Temporal variation in the meteorological factors and N_2O fluxes at the MLW site where daily water sampling took place. (**a**) Time series of water temperature (T_w,°C) and wind speed (m s^{-1}); (**b**) Temporal variation in the N_2O fluxes (F_n, μmol m^{-2} d^{-1}). Black squares indicate the whole-lake mean N_2O fluxes. Measurement locations are shown in Fig. 1.

Table 2. The annual mean N_2O fluxes (mean ± SD) for the three zones in Lake Taihu during 2012–2016.

		F_n (μmol m^{-2} d^{-1})					
Zone	Area (km^2)	2012	2013	2014	2015	2016	Mean
Zone 1	494	18.1 ± 10.2	9.0 ± 5.7	14.9 ± 8.1	11.1 ± 5.7	3.6 ± 2.5	11.4 ± 5.6
Zone 2	1397	2.4 ± 3.7	1.0 ± 2.3	1.1 ± 2.0	2.2 ± 1.4	0.4 ± 2.4	1.4 ± 0.9
Zone 3	447	1.3 ± 2.5	2.3 ± 2.1	1.7 ± 2.4	0.6 ± 0.6	0.6 ± 1.7	1.3 ± 0.7
Whole lake	2338	5.5 ± 4.8	2.9 ± 3.0	4.1 ± 3.4	3.8 ± 2.1	1.1 ± 2.3	3.5 ± 1.8

N_2O fluxes and ORP (Table 4). In addition, the seasonal N_2O fluxes were correlated with NO_2^--N in zone 1 ($r = 0.62$, $p < 0.01$, $n = 16$) and zone 2 ($r = 0.56$, $p = 0.012$, $n = 16$). In zone 3, the seasonal variation in the N_2O fluxes was moderately correlated with DOC ($r = 0.48$, $p = 0.067$, $n = 15$; Supporting Information Fig. S6a). It should be noted that the seasonal N_2O fluxes in the three zones were positively correlated with the inputs of river water from the Taihu Basin (zone 1: $r = 0.54$, $p < 0.05$; zone 2: $r = 0.67$, $p < 0.01$; zone 3: $r = 0.51$, $p < 0.05$; Supporting Information Fig. S6b).

N_2O emission factor

The multiple-year averages of EF_a and EF_b were 0.19% and 0.18%, respectively. The spatial distribution of EF_b showed an opposite pattern to that of N loadings (Figs. 2, 8a), where EF_b decreased as the N loadings increased. The spatial variation in EF_b was positively correlated with the mass ratio of DOC : DIN (Fig. 8b). Zone 3, with abundant submerged biomass, had the highest EF_b and largest mass ratio of DOC : DIN (Table 1). In addition, the seasonal variations in EF_b were also correlated with the ratio of DOC : DIN in the three zones (zone 1: $r = 0.84$, $p < 0.01$, $n = 16$; zone 2: $r = 0.88$, $p < 0.01$, $n = 16$; zone 3: $r = 0.73$, $p < 0.01$, $n = 16$; Supporting Information Table S2). The EF_b showed seasonality: autumn (mean ± SD: 0.33% ± 0.05%) > summer (0.21% ± 0.01%) > winter (0.10% ± 0.04%) > spring (0.07% ± 0.04%). There were statistically significant differences ($p < 0.01$) in EF_b among autumn, summer, and winter, while there was no significant difference ($p = 0.49$) in EF_b between winter and spring.

Table 3. Linear regression equation between N_2O fluxes (y, μmol m^{-2} d^{-1}) and water temperature (x,°C) in the three zones*.

Zone	Regression equation
Zone 1	$y = 2.16(\pm1.14)x - 27.73(\pm7.24)$; $R^2 = 0.17$; $p = 0.073$
Zone 2	$y = 0.52(\pm0.22)x - 8.06(\pm1.11)$; $R^2 = 0.24$; $p = 0.028$
Zone 3	$y = 0.35(\pm0.11)x - 4.96(\pm0.70)$; $R^2 = 0.36$; $p = 0.005$
Whole lake	$y = 0.62(\pm0.23)x - 7.63(\pm1.69)$; $R^2 = 0.29$; $p = 0.015$

*The total number of observations is 20, representing seasonal samplings between 2012 and 2016, for all the regression relations shown. The parameter bounds on the linear regression coefficients are one standard deviation.

Discussion

Consistencies with previous studies

Previous studies reported N_2O fluxes ranging from −8 μmol m^{-2} d^{-1} to 64.7 μmol m^{-2} d^{-1}, with a mean value of 2.9–3.4 μmol m^{-2} d^{-1} (Supporting Information Table S3). The N_2O fluxes reported in this study ranged from −7.1 μmol m^{-2} d^{-1} to 152.7 μmol m^{-2} d^{-1}, with a mean value of 3.5 μmol m^{-2} d^{-1}, across the whole lake. The mean N_2O emission rate from Lake Taihu was similar to that from temperate lakes (3.4 μmol m^{-2} d^{-1}) and higher than that from high latitude lakes (1.1 μmol m^{-2} d^{-1}) reported by Soued et al. (2016). For comparison, the mean N_2O flux in Lake Kivu, a large and deep-tropical lake, was 0.43 μmol m^{-2} d^{-1} (Roland et al. 2017). However, the mean N_2O flux reported in this study agrees with the global median value (3.9 μmol m^{-2} d^{-1}) estimated by Hu et al. (2016).

The diurnal pattern observed is consistent with field measurements by Yan et al. (2012), Xia et al. (2013*a*), and Yang et al. (2015) and demonstrates that a single daytime sample can be representative of the daily average N_2O flux. This finding has been widely adopted for N_2O fluxes measurements in terrestrial ecosystems (Song et al. 2009). The limited diurnal variations reported in this study may be associated with the small lake temperature variations that occur over a single day. Diurnal patterns in lake N_2O fluxes are expected when the diurnal range in temperature is > 10°C (Yang et al. 2015), while the lake water temperature differences in Lake Taihu were less than 4.2°C based on the hourly measurements at the MLW site (Supporting Information Fig. S4).

A previous study by Wang et al. (2009) reported a much larger mean flux of 11.9 μmol m^{-2} d^{-1} for Lake Taihu, which is 240% higher than the flux in the present study. This large discrepancy may have been caused by two reasons. First, Wang et al. (2009) reported N_2O emissions for only summer, when the N_2O emissions are normally high (Wang et al. 2007; Hinshaw and Dahlgren 2013; Fig. 6b). Second, the study by Wang et al. (2009) reported fluxes when the N loading in Lake Taihu was much higher than the present. Long-term monitoring indicated that whole-lake NH_4^+-N concentrations decreased from 0.39 mg L^{-1} in 2007 to 0.11 mg L^{-1} in 2016 (Supporting Information Fig. S7). The decreasing N loading in this study was consistent with previous studies that showed a

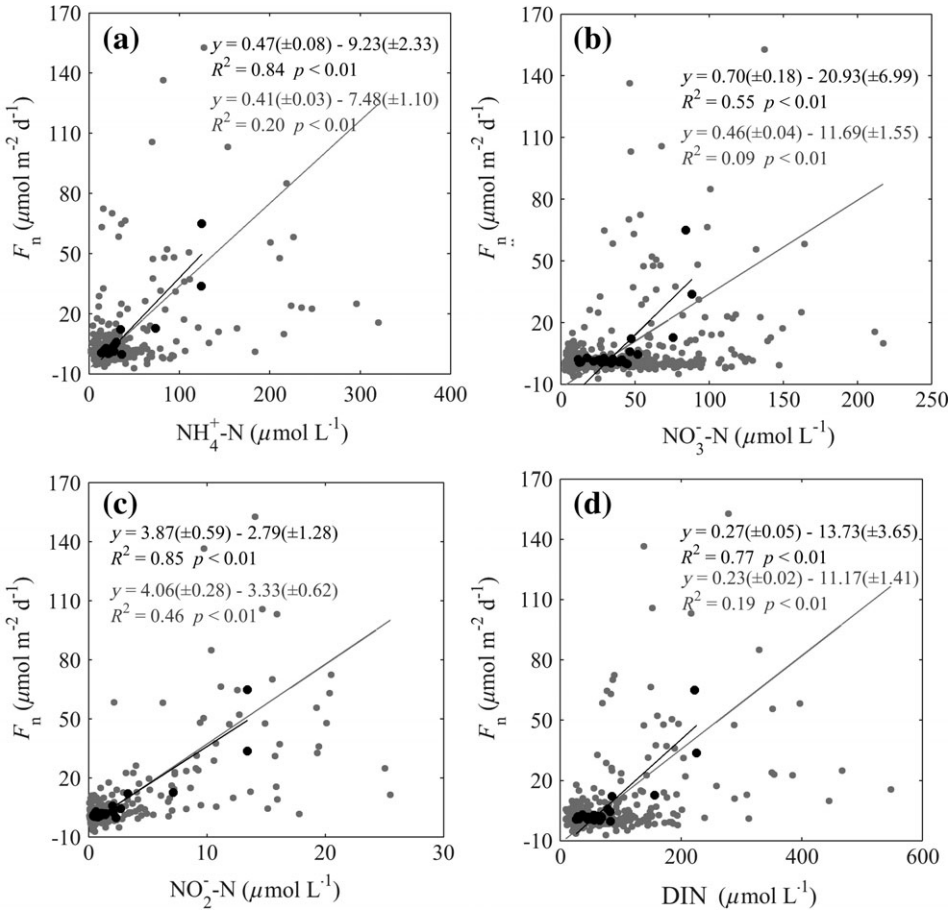

Fig. 7. Correlation of the N_2O fluxes and (**a**) NH_4^+-N, (**b**) NO_3^--N, (**c**) NO_2^--N, and (**d**) DIN across Lake Taihu. Black points represent the mean values for each spatial sampling site during 2012–2015, and gray points represent all whole-lake survey data across space and time during 2012–2015. Parameter bounds on the regression coefficients are 95% confidence limits.

decline in Chinese lake N loading due to large government investments in environmental remediation (Tong et al. 2017; Zhou et al. 2017). Frequent eutrophication events have made Lake Taihu a priority environmental topic in China, leading to major environmental protection actions (e.g., the establishment of wastewater treatment plants) to reduce river pollutant discharge and improve the lake water quality (Duan et al. 2009; Zhou et al. 2017). These actions may have directly contributed to the decrease in anthropogenic N loading input. The decreased N loading, along with the correlation shown in Fig. 7, may contribute to the moderate N_2O emissions of Lake Taihu reported here. The high-temporal variability documented in the present study emphasizes the importance of year-long monitoring sampling to achieve unbiased results. The

Table 4. Spatial Pearson correlation of the annual mean N_2O fluxes and water quality indices in different zones of the lake*.

	DO	pH	NO_2^--N	NO_3^--N	NH_4^+-N	TP	TN	Spc	DOC	Chl *a*	ORP
All	−0.88†	−0.67†	0.92†	0.75†	0.92†	0.72†	0.79†	0.59†	0.56†	0.27	0.18
Zone 1	−0.90†	−0.75‡	0.91†	0.82†	0.91†	0.83†	0.88†	0.88‡	0.71‡	−0.21	0.31
Zone 2	−0.34	−0.76†	0.59‡	0.48	0.31	−0.27	−0.02	−0.54‡	−0.79†	−0.42	0.49
Zone 3	−0.34	−0.93†	0.83†	−0.01	0.26	0.29	0.17	−0.54	0.44	0.68	−0.06

*Chl *a*, chlorophyll *a* concentration; DO, dissolved oxygen concentration; DOC, dissolved organic carbon concentration; ORP, oxidation reduction potential; Spc, specific conductance; TN, total nitrogen concentration; TP, total phosphorus concentration. All, data acquired at all the spatial sampling sites (n = 29); Zone 1, data acquired at the eutrophic zone (n = 9); Zone 2, data acquired at the transitional zone (n = 12); Zone 3, data acquired at the submerged vegetation zone (n = 8).
†Correlation is significant at the 0.01 level.
‡Correlation is significant at the 0.05 level.

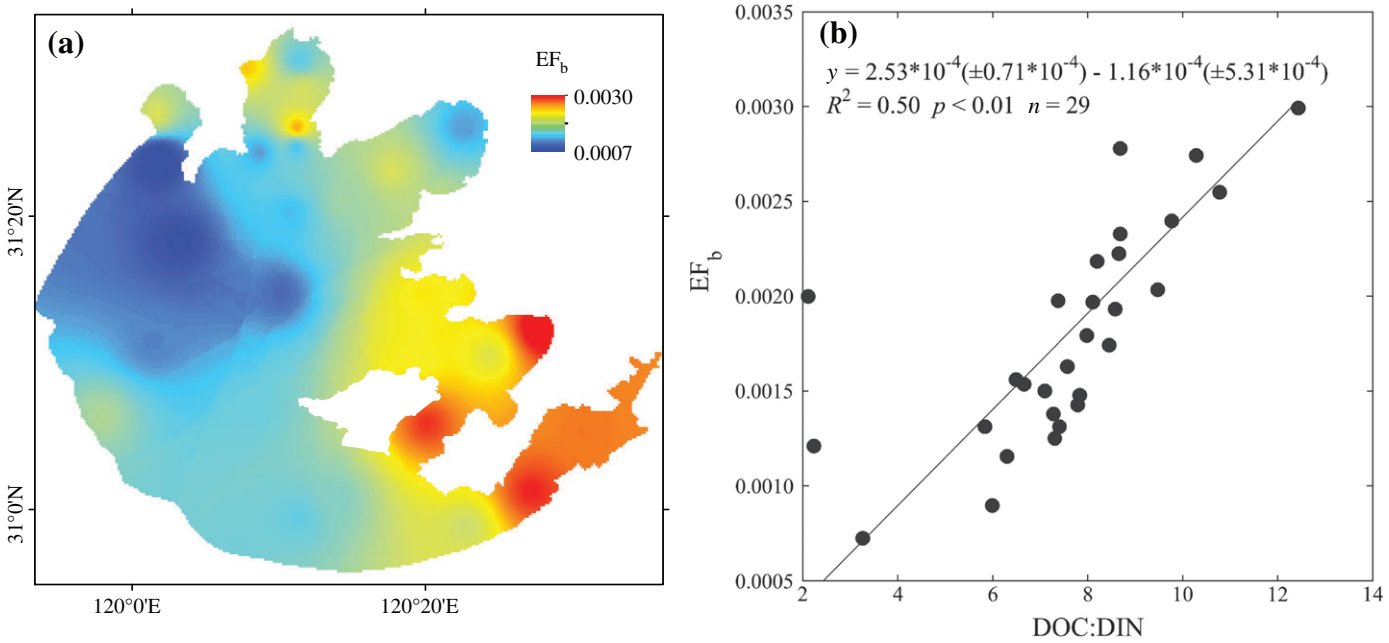

Fig. 8. (**a**) Map showing the spatial distribution of annual mean EF_b across Lake Taihu during 2012–2015 and (**b**) correlation between EF_b and the ratio of DOC concentration (mg L^{-1}) to DIN concentration (mg L^{-1}) (DOC : DIN) across Lake Taihu. Parameter bounds on the regression coefficients are 95% confidence limits.

continuity of environmental political management can also justify long-term high-frequency monitoring in lake environments (Tong et al. 2017; Zhou et al. 2017).

Given the large variability in observed fluxes across time and space, ranging from −7.1 μmol m^{-2} d^{-1} to 152.7 μmol m^{-2} d^{-1}, we extrapolated the Lake Taihu measurements to the whole basin lake (~3160 km^2) to estimate the annual budget of N_2O fluxes at the zonal base. We estimated that the whole basin lake emits approximately 113 Mg N of N_2O to the atmosphere yearly compared to the estimated basin-level riverine N_2O emissions of 1400 Mg yr^{-1} (Wang et al. 2009; Yu et al. 2013; Xia et al. 2013*a,b*). For comparison, the estimated N_2O emissions from cultivated lands in the drainage basin are 12,000 Mg yr^{-1} (Wang et al. 2009).

Coregulation of lake N_2O fluxes by N and temperature

The large differences in the N_2O fluxes between eutrophic zone 1 and zone 2 are an indication of the importance of nitrogen pollution in N_2O emissions. The mean N_2O flux in zone 1 was 11.4 μmol m^{-2} d^{-1}, compared with 1.4 μmol m^{-2} d^{-1} in zone 2. Zone 1 received a majority of the river N discharge, while there were no direct river N inputs to zone 2 and zone 3 (Supporting Information Fig. S1; Xiao et al. 2017). Anthropogenic N inputs contributed to the substantial differences in N concentrations among the different zones. For example, the zonal mean of the NH_4^+-N concentrations in zone 1 was 192% higher than that in zone 2 and zone 3.

Strong N control on N_2O fluxes can be further demonstrated by the positive correlation between N_2O fluxes and NH_4^+-N, NO_3^--N, and NO_2^--N concentrations (Table 4; Fig. 7). High N loading (NH_4^+-N, NO_3^--N, and NO_2^--N) drives microbial processes of N_2O production. Lake Taihu receives anthropogenic N via river discharge and, therefore, contains high concentrations of N (Qin et al. 2007; Duan et al. 2009) that lead to enhanced N_2O emissions. This finding is consistent with previous studies, which reported that N_2O fluxes across the water–air interface were determined by N loading in aquatic ecosystems with external N inputs (Baulch et al. 2011; Beaulieu et al. 2011; Yu et al. 2013; Hama-Aziz et al. 2017; Borges et al. 2018). Given these strong positive correlations between N_2O and N in the water and the uniformly distributed water temperature, wind speed (wind-dependent gas transfer coefficient), and solar radiation (Wang et al. 2014; Xiao et al. 2017), the spatial variability in N_2O fluxes is primarily attributed to N loading in the lake.

The control of temperature on N_2O emissions in Lake Taihu is consistent with the control of temperature on microbial processes related to N_2O production (Beaulieu et al. 2010; Hinshaw and Dahlgren 2013). The lower water temperature may have contributed to the undersaturation of N_2O in winter (Fig. 6b). However, the importance of temperature control varied among the three zones (Table 3). In particular, the positive correlation between water temperature and N_2O emissions was significant in zone 2 and zone 3 and not significant in zone 1. The weak correlation between N_2O emissions and temperature in zone 1 shows the importance of N regulation for N_2O fluxes in eutrophic waters (Wang et al. 2007; Davidson et al. 2015).

The lake N_2O fluxes and N loading exhibited opposite seasonal patterns (Figs. 3, 5). High N concentrations in inflowing rivers occurred in summer due to the heavy application of fertilizers (Ju et al. 2009; Zhao et al. 2015), corresponding to relatively low N concentrations in the lake (Fig. 3d,e). This contrasting pattern is caused by the stimulation of N biogeochemical cycling by warm water temperatures (Beaulieu et al. 2010), which contributed to the larger N removal rates in the summer. In contrast, low-water temperatures in winter reduced these microbial processes, leading to N accumulation and low N_2O emissions. Our results, together with a previous study on the weak response of N_2O fluxes to N enrichment in boreal lakes (mean temperature: 1–3°C; Klaus et al. 2017), indicate that water temperature and N loading interactively control N_2O fluxes in Lake Taihu.

Controls of the lake N_2O EFs

EF_b showed a strong variation across the lake. The strong positive spatial correlation between the EF_b and DOC : DIN mass ratio (Fig. 8b) implied that higher DOC may enhance N_2O production, as suggested in previous studies (Xia et al. 2013*a*; Hu et al. 2016). The submerged macrophytes in zone 3 are a labile carbon source for N_2O production, which contributes to the high EF_b of this zone (Fig. 8a). Consistent with a few previous studies on rivers and streams (Mulholland et al. 2008; Beaulieu et al. 2011; Hinshaw and Dahlgren 2013; Hu et al. 2016), the EF_b reported here was also inversely correlated with N loadings (Figs. 2, 8a). This finding was attributed to decreasing microbial activity with increasing N inputs due to progressive biological saturation (Mulholland et al. 2008; Hu et al. 2016; Hampel et al. 2018). Both of these findings indicated that the variability in the N_2O EF was related to a range of factors, such as anthropogenic N inputs and DOC concentrations (Hu et al. 2016; Hama-Aziz et al. 2017).

There are some EF_b hot spots in zone 1, where algae biomass is abundant (Fig. 7a). Algal blooms likely enhanced the N_2O production by enhancing organic inputs (Chen et al. 2012). Field measurements showed that seasonal variations in the EF_b in zone 1 were correlated with Chl *a* ($r = 0.50$, $p = 0.05$, $n = 16$), an indicator of algal abundance. Algae may directly influence N_2O production in freshwater lakes by enhancing carbon inputs. A study showed that the previous year's winter temperature influences the algal blooming in Lake Taihu (Duan et al. 2009). An increase in the winter temperature over time was observed based on long-term continuous measurements in Lake Taihu during the sampling period (Fig. 3a; Lee et al. 2014); for example, the winter temperature in 2014, with a mean value of 6.2°C, was significantly ($p = 0.04$) higher than that in 2011 (mean value of 4.5°C). These findings not only explained the yearly increase in the Chl *a* maximum peaks, especially in eutrophic zone 1 (Fig. 3c) but also suggested that a climate-change-related increase in water temperature can influence lake N_2O production via stimulating algal blooming.

Environmental control of N_2O fluxes

The present study showed that the absence of submerged macrophytes had no significant effect on lake N_2O fluxes (Figs. 4, 5). The mean N_2O flux and dissolved N_2O concentration in zone 3, with abundant submerged macrophytes, were 1.3 μmol m^{-2} d^{-1} and 11.8 nmol L^{-1}, respectively. For comparison, the values in zone 2 were 1.4 μmol m^{-2} d^{-1} and 12.0 nmol L^{-1}, respectively. Our results are consistent with the field measurements by Yao et al. (2017), which reported that submerged macrophytes are not a primary factor in the spatial distribution of N_2O. A previous study showed that macrophytes could indirectly influence N_2O fluxes by altering the DIN concentrations and oxygen availability in aquatic ecosystems (Murray et al. 2015). However, the concentrations of DIN and DO in Lake Taihu showed small differences between zone 2, with an absence of submerged macrophytes, and zone 3, with abundant submerged macrophytes (Fig. 2). Additionally, our field measurements showed that beyond the threshold of the DOC : DIN mass ratio (3–5), surface N_2O concentrations and N_2O fluxes variabilities in the lake had stable trends with an increasing DOC : DIN mass ratio (Supporting Information Fig. S8). The mean DOC : DIN mass ratios were 7.03 and 9.64 (Table 1) in zone 2 and zone 3, respectively. Both of these may have contributed to the observed results in the lake.

N_2O fluxes are driven by the microbial processes of nitrification and denitrification in aquatic ecosystems. The water column remains well oxygenated in the shallow Lake Taihu (Supporting Information Fig. S9; Hu et al. 2015), indicating denitrification cannot occur in the water column (Zhang et al. 2010), while the high concentrations of DO and NH_4^+-N provide favorable conditions for the occurrence of nitrification (Zhang et al. 2010; Whitfield et al. 2011; Hampel et al. 2018). The observed fluxes were highly correlated with NH_4^+-N and NO_2^--N (Fig. 7), which also confirmed that nitrification dominated N_2O production in the lake (Yu et al. 2013). A previous study showed that the nitrification rates of the water column at the eutrophic sampling site in Lake Taihu were significantly higher fueled by high NH_4^+-N concentrations (Hampel et al. 2018), and this finding may have contributed to the higher N_2O fluxes in eutrophic zone 1 (Figs. 4, 5). Some N_2O peaks occurred in the bottom water in May 2013 based on the N_2O vertical profile measurements at MLW (Supporting Information Fig. S2c,d), which suggest that denitrification could occur in the sediments and thus contribute to lake N_2O production (Xu et al. 2010; Zhang et al. 2010).

Previous studies have reported that low pH may limit nitrification rates and reduce aquatic N_2O emissions (Beman et al. 2011; Soued et al. 2016). But negative relationship between the pH and N_2O fluxes was found in Lake Taihu (Table 4). Also, a significant positive correlation between the N_2O fluxes and DO was expected in the lake water if N_2O was primarily produced via the nitrification process (Yu et al. 2013). However, our results showed that the N_2O fluxes were negatively correlated with DO concentrations ($r = -0.34$, $p < 0.01$,

n = 448) in the lake, consistent with previous measurements in rivers with external N loading (Rosamond et al. 2012; Yu et al. 2013). These results indicated N_2O production in the lake was favored under low DO and low pH due to the high anthropogenic N inputs, which was consistent with field measurements in rivers in eastern China (Yu et al. 2013).

Conclusions, implications, and perspectives

With a 5-yr continuous measurement period of N_2O concentrations and fluxes, we quantified the budget of N_2O fluxes and investigated their spatial and temporal variations in Lake Taihu. The annual mean N_2O flux of this eutrophic lake was 3.5 ± 1.8 μmol m^{-2} d^{-1}, with an annual N_2O budget of 134.4 ± 69.8 Mg (10^6 g) yr^{-1}, suggesting that Lake Taihu is a moderate source of atmospheric N_2O compared to other inland lakes worldwide.

The highest N_2O fluxes occurred in the eutrophic zone, where anthropogenic N inputs were high. The lowest N_2O fluxes occurred in noneutrophic zones that received no river nutrient inputs. N inputs and temperature coregulated the variations in N_2O fluxes from Lake Taihu. The observed fluxes were highly correlated with NH_4^+-N and NO_2^--N concentrations, with respect to lower correlations to NO_3^--N concentrations. This indicates that nitrification is an important process regulating N_2O in the lake. The temporal variations in the observed fluxes were primarily regulated by water temperature.

Previous studies proposed that N_2O emissions from aquatic ecosystems in eastern China were high due to high anthropogenic N loading (Seitzinger and Kroeze 1998; Seitzinger et al. 2000). For example, agricultural N fertilizer use alone within the study areas has reached 550 kg N ha^{-1} yr^{-1}, and 50–80% of this anthropogenic N is lost to waters (Xing and Zhu 2002; Ju et al. 2009). For comparison, N fertilizer use in the US Corn Belt, an intensive agricultural region, was 100 kg N ha^{-1} yr^{-1} (Turner et al. 2015; Griffis et al. 2017). Given the significant positive correlations between N_2O fluxes and N loading (Table 4; Fig. 7), the reported moderate N_2O emissions from Lake Taihu from 2012 to 2016 were probably due to the decrease in N loading, especially in zone 1 (Supporting Information Figs. S7, S10). The high temporal and spatial variabilities reported here highlight the importance of long-term and spatially distributed sampling to achieve unbiased results.

Long-term field measurements of N_2O fluxes in the highly heterogeneous Lake Taihu will shed further light on the effect of basin and lake management on N_2O emissions from freshwater lakes. Additionally, eutrophication and global warming would enhance lake N_2O emissions given the coregulation of N_2O fluxes by anthropogenic N input and water temperature. This study, to the best of our knowledge, represents the first attempt to quantify lake N_2O fluxes at such a high temporal and spatial resolution. The results show that Lake Taihu is a moderate source of atmospheric N_2O and provide new insights into the biogeochemistry of Lake Taihu, the third largest freshwater lake in China. The results provide a valuable data source for model development and validation to project N_2O fluxes from fresh waterbodies under a changing environment.

The N_2O EFs reported in the lake were comparable to the global mean EFs of 0.17% from riverine ecosystems (Hu et al. 2016). The minor difference between EF_a and EF_b in this study was probably due to the thorough temporal and spatial sampling strategy (Outram and Hiscock 2012; Hama-Aziz et al. 2017). Previous studies reported EF_b ranging from 0.03% to 1% (McCrackin and Elser 2011; Outram and Hiscock 2012). In addition, different EFs for different water types within the same catchment have been reported (Outram and Hiscock 2012). Our data showed that EFs varied spatially within one lake along gradients of N enrichment and submerged macrophytes (Fig. 8a).

The annual mean net anthropogenic N input to Lake Taihu was approximately 0.03 Tg N yr^{-1} (Supporting Information Table S1), but the proportion of transported N converted to N_2O was unknown. Our results reported here show that only 0.30% of the net N input was lost as N_2O. Taking the national total N_2O emissions as 2150 Gg yr^{-1} (Zhou et al. 2014), the N_2O emissions from Lake Taihu accounted for 0.006% of the national N_2O budget across China. Considering the large N flow into Lake Taihu (Supporting Information Table S1, Fig. 2), the relatively low N_2O emissions may be attributed to the shallow depth of the lake, which may limit the space for the development of microbial communities, and to the well-oxygenated water, which prevent the occurrence of denitrification. Nitrification and denitrification rates should be measured in future studies of Lake Taihu to have a more complete quantification of N cycles.

References

American Public Health Association. 1995. Standard methods for the examination of water and wastewater, 19th ed. APHA, American Water Works Association, Water Environment Federation.

Baulch, H., S. Schiff, R. Maranger, and P. Dillon. 2011. Nitrogen enrichment and the emission of nitrous oxide from streams. Global Biogeochem. Cycles **25**: GB4013. doi: 10.1029/2011GB004047

Beaulieu, J. J., W. D. Shuster, and J. A. Rebholz. 2010. Nitrous oxide emissions from a large, impounded river: The Ohio River. Environ. Sci. Technol. **44**: 7527–7533. doi:10.1021/es1016735

Beaulieu, J. J., and others. 2011. Nitrous oxide emission from denitrification in stream and river networks. Proc. Natl. Acad. Sci. USA **108**: 214–219. doi:10.1073/pnas.1011464108

Beman, J. M., and others. 2011. Global declines in oceanic nitrification rates as a consequence of ocean acidification.

Proc. Natl. Acad. Sci. USA **108**: 208–213. doi:10.1073/pnas.1011053108

Borges, A. V., and others. 2015. Globally significant greenhouse-gas emissions from African inland waters. Nat. Geosci. **8**: 637–642. doi:10.1038/ngeo2486

Borges, A. V., and others. 2018. Effects of agricultural land use on fluvial carbon dioxide, methane and nitrous oxide concentrations in a large European river, the Meuse (Belgium). Sci. Total Environ. **610**: 342–355. doi:10.1016/j.scitotenv.2017.08.047

Chapuis-Lardy, L., N. Wrage, A. Metay, J.-L. Chotte, and M. Bernoux. 2007. Soils, a sink for N_2O?A review. Glob. Chang. Biol. **13**: 1–17. doi:10.1111/j.1365-2486.2006.01280.x

Chen, X. F., L. Y. Yang, L. Xiao, A. J. Miao, and B. D. Xi. 2012. Nitrogen removal by denitrification during cyanobacterial bloom in Lake Taihu. J. Freshw. Ecol. **27**: 243–258. doi:10.1080/02705060.2011.644405

Cole, J. J., and N. F. Caraco. 1998. Atmospheric exchange of carbon dioxide in a low-wind oligotrophic lake measured by the addition of SF_6. Limnol. Oceanogr. **43**: 647–656. doi:10.4319/lo.1998.43.4.0647

Davidson, E. A. 2009. The contribution of manure and fertilizer nitrogen to atmospheric nitrous oxide since 1860. Nat. Geosci. **2**: 659–662. doi:10.1038/ngeo608

Davidson, T. A., and others. 2015. Eutrophication effects on greenhouse gas fluxes from shallow-lake mesocosms override those of climate warming. Glob. Chang. Biol. **21**: 4449–4463. doi:10.1111/gcb.13062

De Klein, C., and others. 2006. Chapter 11: N_2O emissions from managed soils, and CO_2 emissions from lime and urea application, p. 1–54. *In* IPCC guidelines for national greenhouse gas inventories. Prepared by the National Greenhouse Gas Inventories Programme, v. **4**. Institute for Global Environmental Strategies.

Duan, H., R. Ma, S. A. Loiselle, Q. Shen, H. Yin, and Y. Zhang. 2014. Optical characterization of black water blooms in eutrophic waters. Sci. Total Environ. **482**: 174–183. doi:10.1016/j.scitotenv.2014.02.113

Duan, H. T., and others. 2009. Two-decade reconstruction of algal blooms in China's Lake Taihu. Environ. Sci. Technol. **43**: 3522–3528. doi:10.1021/es8031852

Ebina, J., T. Tsutsui, and T. Shirai. 1983. Simultaneous determination of total nitrogen and total phosphorus in water using peroxodisulfate oxidation. Water Res. **17**: 1721–1726. doi:10.1016/0043-1354(83)90192-6

Foley, J., D. de Haas, Z. Yuan, and P. Lant. 2010. Nitrous oxide generation in full-scale biological nutrient removal wastewater treatment plants. Water Res. **44**: 831–844. doi:10.1016/j.watres.2009.10.033

Fu, C., X. Lee, T. Griffis, J. Baker, and P. Turner. 2018. A modeling study of direct and indirect N_2O emissions from a representative catchment in the U.S. Corn Belt. Water Resour. Res. **54**: 3632–3653. doi:10.1029/2017WR022108

Griffis, T. J., and others. 2017. Nitrous oxide emissions are enhanced in a warmer and wetter world. Proc. Natl. Acad. Sci. USA **114**: 12081–12085. doi:10.1073/pnas.1704552114

Hama-Aziz, Z. Q., K. M. Hiscock, and R. J. Cooper. 2017. Indirect nitrous oxide emission factors for agricultural field drains and headwater streams. Environ. Sci. Technol. **51**: 301–307. doi:10.1021/acs.est.6b05094

Hampel, J. J., M. McCarthy, W. Gardner, L. Zhang, H. Xu, G. Zhu, and S. Newell. 2018. Nitrification and ammonium dynamics in Taihu Lake, China: Seasonal competition for ammonium between nitrifiers and cyanobacteria. Biogeosciences **15**: 733–748. doi:10.5194/bg-15-733-2018

Harrison, J. A., and others. 2009. The regional and global significance of nitrogen removal in lakes and reservoirs. Biogeochemistry **93**: 143–157. doi:10.1007/s10533-008-9272-x

Hinshaw, S. E., and R. A. Dahlgren. 2013. Dissolved nitrous oxide concentrations and fluxes from the eutrophic San Joaquin River, California. Environ. Sci. Technol. **47**: 1313–1322. doi:10.1021/es301373h

Hu, M., D. Chen, and R. A. Dahlgren. 2016. Modeling nitrous oxide (N_2O) emission from rivers: A global assessment. Glob. Chang. Biol. **22**: 3566–3582. doi:10.1111/gcb.13351

Hu, Z., Q. Xiao, J. Yang, W. Xiao, W. Wang, S. Liu, and X. Lee. 2015. Temporal dynamics and drivers of ecosystem metabolism in a large subtropical shallow lake (Lake Taihu). Int. J. Environ. Res. Public Health **12**: 3691–3706. doi:10.3390/ijerph120403691

Huttunen, J. T., and others. 2003. Fluxes of methane, carbon dioxide and nitrous oxide in boreal lakes and potential anthropogenic effects on the aquatic greenhouse gas emissions. Chemosphere **52**: 609–621. doi:10.1016/s0045-6535(03)00243-1

IPCC. 2013. Climate change 2013: The physical science basis, p. 53–54. *In* T. F. Stocker, D. Qin, G. K. Plattner, M. Tignor, S. K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex, and P. M. Midgley [eds.], Contribution of working group I to the fifth assessment report of the Intergovernmental Panel on Climate Change. Cambridge Univ. Press.

Ivens, W. P. M. F., D. J. J. Tysmans, C. Kroeze, A. J. Löhr, and J. van Wijnen. 2011. Modeling global N_2O emissions from aquatic systems. Curr. Opin. Environ. Sustain. **3**: 350–358. doi:10.1016/j.cosust.2011.07.007

Ju, X. T., and others. 2009. Reducing environmental risk by improving N management in intensive Chinese agricultural systems. Proc Natl Acad Sci USA **106**: 3041–3046. doi:10.1073/pnas.0813417106

Kampschreur, M., H. Temmink, R. Kleerebezem, M. Jetten, and M. van Loosdrecht. 2009. Nitrous oxide emission during wastewater treatment. Water Res. **43**: 4093–4103. doi:10.1016/j.watres.2009.03.001

Klaus, M., A.-K. Bergström, A. Jonsson, A. Deininger, E. Geibrink, and J. Karlsson. 2017. Weak response of greenhouse gas emissions to whole lake N enrichment. Limnol. Oceanogr. **63**: S340–S353. doi:10.1002/lno.10743

Lee, X., and others. 2014. The Taihu eddy flux network: An observational program on energy, water and greenhouse gas fluxes of a large freshwater lake. Bull. Am. Meteorol. Soc. **95**: 1583–1594. doi:10.1175/bams-d-13-00136.1

Liu, W., L. Yao, X. Jiang, L. Guo, X. Cheng, and G. Liu. 2017. Sediment denitrification in Yangtze lakes is mainly influenced by environmental conditions but not biological communities. Sci. Total Environ. **616-617**: 978–987. doi:10.1016/j.scitotenv.2017.10.221

Luo, J., and others. 2016. Applying remote sensing techniques to monitoring seasonal and interannual changes of aquatic vegetation in Taihu Lake, China. Ecol. Indic. **60**: 503–513. doi:10.1016/j.ecolind.2015.07.029

MacIntyre, S., A. Jonsson, M. Jansson, J. Aberg, D. E. Turney, and S. D. Miller. 2010. Buoyancy flux, turbulence, and the gas transfer coefficient in a stratified lake. Geophys. Res. Lett. **37**. doi:10.1029/2010gl044164

McCrackin, M. L., and J. J. Elser. 2011. Greenhouse gas dynamics in lakes receiving atmospheric nitrogen deposition. Global Biogeochem. Cycles **25**. doi:10.1029/2010gb003897

Mengis, M., R. Gachter, and B. Wehrli. 1996. Nitrous oxide emissions to the atmosphere from an artificially oxygenated lake. Limnol. Oceanogr. **41**: 548–553. doi:10.4319/lo.1996.41.3.0548

Mengis, M., R. Gächter, and B. Wehrli. 1997. Sources and sinks of nitrous oxide (N_2O) in deep lakes. Biogeochemistry **38**: 281–301. doi:10.1023/A:1005814020322

Miettinen, H., J. Pumpanen, J. Heiskanen, H. Aaltonen, I. Mammarella, A. Ojala, J. Levula, and M. Rantakari. 2015. Towards a more comprehensive understanding of lacustrine greenhouse gas dynamics—two-year measurements of concentrations and fluxes of CO_2, CH_4 and N_2O in a typical boreal lake surrounded by managed forests. Boreal Environ. Res. **20**: 75–89.

Mulholland, P. J., and others. 2008. Stream denitrification across biomes and its response to anthropogenic nitrate loading. Nature **452**: 202–205. doi:10.1038/nature06686

Murray, R. H., D. V. Erler, and B. D. Eyre. 2015. Nitrous oxide fluxes in estuarine environments: Response to global change. Glob. Chang. Biol. **21**: 3219–3245. doi:10.1111/gcb.12923

Outram, F. N., and K. M. Hiscock. 2012. Indirect nitrous oxide emissions from surface water bodies in a lowland arable catchment: A significant contribution to agricultural greenhouse gas budgets? Environ. Sci. Technol. **46**: 8156–8163. doi:10.1021/es3012244

Qin, B., P. Xu, Q. Wu, L. Luo, and Y. Zhang. 2007. Environmental issues of Lake Taihu, China. Hydrobiologia **581**: 3–14. doi:10.1007/s10750-006-0521-5

Ravishankara, A. R., J. S. Daniel, and R. W. Portmann. 2009. Nitrous oxide (N_2O): The dominant ozone-depleting substance emitted in the 21st century. Science **326**: 123–125. doi:10.1126/science.1176985

Read, J. S., and others. 2012. Lake-size dependency of wind shear and convection as controls on gas exchange. Geophys. Res. Lett. **39**: 39. doi:10.1029/2012gl051886

Roland, F. A., F. Darchambeau, C. Morana, and A. Borges. 2017. Nitrous oxide and methane seasonal variability in the epilimnion of a large tropical meromictic lake (Lake Kivu, East-Africa). Aquat. Sci. **79**: 209–218. doi:10.1007/s00027-016-0491-2.

Rosamond, M. S., S. J. Thuss, and S. L. Schiff. 2012. Dependence of riverine nitrous oxide emissions on dissolved oxygen levels. Nat. Geosci. **5**: 715–718. doi:10.1038/ngeo1556

Saikawa, E., and others. 2014. Global and regional emissions estimates for N_2O. Atmos. Chem. Phys. **14**: 4617–4641. doi:10.5194/acp-14-4617-2014

Seitzinger, S., and others. 2006. Denitrification across landscapes and waterscapes: A synthesis. Ecol. Appl. **16**: 2064–2090. doi:10.1890/1051-0761(2006)016[2064:DALAWA]2.0.CO;2

Seitzinger, S. P., and C. Kroeze. 1998. Global distribution of nitrous oxide production and N inputs in freshwater and coastal marine ecosystems. Global Biogeochem. Cycles **12**: 93–113. doi:10.1029/97gb03657

Seitzinger, S. P., C. Kroeze, and R. V. Styles. 2000. Global distribution of N_2O emissions from aquatic systems: Natural emissions and anthropogenic effects. Chemosphere Glob. Chang. Sci. **2**: 267–279.

Sinha, E., A. M. Michalak, and V. Balaji. 2017. Eutrophication will increase during the 21st century as a result of precipitation changes. Science **357**: 405–408. doi:10.1126/science.aan2409

Song, C., X. Xu, H. Tian, and Y. Wang. 2009. Ecosystem-atmosphere exchange of CH_4 and N_2O and ecosystem respiration in wetlands in the Sanjiang Plain, Northeastern China. Glob. Chang. Biol. **15**: 692–705. doi:10.1111/j.1365-2486.2008.01821.x.

Soued, C., P. A. del Giorgio, and R. Maranger. 2016. Nitrous oxide sinks and emissions in boreal aquatic networks in Québec. Nat. Geosci. **9**: 116–120. doi:10.1038/ngeo2611

Tong, Y., and others. 2017. Decline in Chinese lake phosphorus concentration accompanied by shift in sources since 2006. Nat. Geosci. **10**: 507–511. doi:10.1038/NGEO2967

Turner, P. A., T. J. Griffis, X. Lee, J. M. Baker, R. T. Venterea, and J. D. Wood. 2015. Indirect nitrous oxide emissions from streams within the US Corn Belt scale with stream order. Proc. Natl. Acad. Sci. USA **112**: 9839–9843. doi:10.1073/pnas.1503598112

Wang, H., L. Yang, W. Wang, J. Lu, and C. Yin. 2007. Nitrous oxide (N_2O) fluxes and their relationships with water-sediment characteristics in a hyper-eutrophic shallow lake, China. J. Geophys. Res. Biogeosci. **112**. doi:10.1029/2005jg000129

Wang, S. L., and others. 2009. The spatial distribution and emission of nitrous oxide (N_2O) in a large eutrophic lake in eastern China: Anthropogenic effects. Sci. Total Environ. **407**: 3330–3337. doi:10.1016/j.scitotenv.2008.10.037

Wang, W., and others. 2014. Temporal and spatial variations in radiation and energy balance across a large freshwater lake in China. J. Hydrol. **511**: 811–824. doi:10.1016/j.jhydrol.2014.02.012

Whitfield, C. J., J. Aherne, and H. M. Baulch. 2011. Controls on greenhouse gas concentrations in polymictic headwater lakes in Ireland. Sci. Total Environ. **410**: 217–225. doi:10.1016/j.scitotenv.2011.09.045

Xia, Y., Y. Li, X. Li, M. Guo, D. She, and X. Yan. 2013*a*. Diurnal pattern in nitrous oxide emissions from a sewage-enriched river. Chemosphere **92**: 421–428. doi:10.1016/j.chemosphere.2013.01.038

Xia, Y. Q., Y. F. Li, C. P. Ti, X. B. Li, Y. Q. Zhao, and X. Y. Yan. 2013*b*. Is indirect N_2O emission a significant contributor to the agricultural greenhouse gas budget? A case study of a rice paddy-dominated agricultural watershed in eastern China. Atmos. Environ. **77**: 943–950. doi:10.1016/j.atmosenv.2013.06.022

Xiao, Q., and others. 2017. Spatial variations of methane emission in a large shallow eutrophic lake in subtropical climate. J. Geophys. Res. Biogeosci. **122**: 1597–1614. doi:10.1002/2017jg003805

Xing, G. X., and Z. L. Zhu. 2002. Regional nitrogen budgets for China and its major watersheds. Biogeochemistry **57**: 405–427. doi:10.1023/a:1016508323200

Xu, H., H. W. Paerl, B. Qin, G. Zhu, and G. Gao. 2010. Nitrogen and phosphorus inputs control phytoplankton growth in eutrophic Lake Taihu, China. Limnol. Oceanogr. **55**: 420–432. doi:10.4319/lo.2010.55.1.0420

Xu, X., H. Tian, and D. Hui. 2008. Convergence in the relationship of CO_2 and N_2O exchanges between soil and atmosphere within terrestrial ecosystems. Glob. Chang. Biol. **14**: 1651–1660. doi:10.1111/j.1365-2486.2008.01595.x

Yan, W. J., L. B. Yang, F. Wang, J. N. Wang, and P. Ma. 2012. Riverine N_2O concentrations, exports to estuary and emissions to atmosphere from the Changjiang River in response to increasing nitrogen loads. Global Biogeochem. Cycles **26**: 15. doi:10.1029/2010gb003984

Yang, M., and others. 2015. N_2O fluxes from the littoral zone of a Chinese reservoir. Biogeosciences **12**: 4711–4723. doi:10.5194/bg-12-4711-2015

Yao, L., C. Chen, G. Liu, and W. Liu. 2017. Sediment nitrogen cycling rates and microbial abundance along a submerged vegetation gradient in a eutrophic lake. Sci. Total Environ. **616–617**: 899–907. doi:10.1016/j.scitotenv.2017.10.230

Yu, Z., and others. 2013. Nitrous oxide emissions in the Shanghai river network: Implications for the effects of urban sewage and IPCC methodology. Glob. Chang. Biol. **19**: 2999–3010. doi:10.1111/gcb.12290

Zhang, G., J. Zhang, S. Liu, J. Ren, and Y. Zhao. 2010. Nitrous oxide in the Changjiang (Yangtze River) Estuary and its adjacent marine area: Riverine input, sediment release and atmospheric fluxes. Biogeosciences **7**: 3505–3516. doi:10.5194/bg-7-3505-2010

Zhang, Y., and others. 2017. Global loss of aquatic vegetation in lakes. Earth Sci. Rev. **173**: 259–265. doi:10.1016/j.earscirev.2017.08.013

Zhao, Y., Y. Xia, C. Ti, J. Shan, B. Li, L. Xia, and X. Yan. 2015. Nitrogen removal capacity of the river network in a nigh nitrogen loading region. Environ. Sci. Technol. **49**: 1427–1435. doi:10.1021/es504316b

Zhou, F., and others. 2014. A new high-resolution N_2O emission inventory for China in 2008. Environ. Sci. Technol. **48**: 8538–8547. doi:10.1021/es5018027

Zhou, Y., and others. 2017. Improving water quality in China: Environmental investment pays dividends. Water Res. **118**: 152–159. doi:10.1016/j.watres.2017.04.035

Zhu, D., and others. 2015. Nitrous oxide emission from infralittoral zone and pelagic zone in a shallow lake: Implications for whole lake flux estimation and lake restoration. Ecol. Eng. **82**: 368–375. doi:10.1016/j.ecoleng.2015.05.032

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Conflict of Interest

None declared.

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