

Invasive *Spartina alterniflora* can mitigate N₂O emission in coastal salt marshes



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ABSTRACT

Although there are studies on nitrous oxide (N₂O) fluxes in coastal salt marshes, temporal and spatial variations of this greenhouse gas are still uncertain. Especially salt marshes of the East China Sea coast covered by invasive *Spartina alterniflora* have shown controversial results. To analyse seasonal patterns of N₂O fluxes and their relationship with environmental factors, three plots dominated by *S. alterniflora*, and differing in sediment salinity and vegetation history (P1, P2, P3), and one bare mudflat (P0) in a salt marsh of Nanhui shore in the southern fringe of Yangtze River estuary have been established. Monthly studies from March 2017 to January 2018 using a chamber technique showed that average N₂O fluxes from all four plots ranged from -41.9 to $39.3 \mu\text{g N}_2\text{O}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$, whereas average flux ($4.2 \mu\text{g N}_2\text{O}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$) in P1, P2 and P3 was not significantly different from that measured in P0 ($1.3 \mu\text{g N}_2\text{O}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$). There was a clear seasonal difference: in spring and summer, all the sites showed slight emission while consumption prevailed in autumn and winter. In vegetated sites this trend was more remarkable than in the bare mudflat. N₂O flux showed positive correlation ($p < .05$) with air and sediment temperature, and plant development (height of vegetation). Nitrate was not the limiting factor of N₂O emission in the Yangtze estuary. In the salt marsh where vegetation community was mature, higher sediment salinity reduced N₂O emission (P1 < P2) by influencing other environmental factors such as total carbon (TC), total nitrogen (TN) content and sediment texture. In comparison with other tidal macrophytes *S. alterniflora* showed relatively low N₂O emission. Therefore, it can be considered as a species for tidal zone stabilisation.

1. Introduction

Nitrous oxide (N₂O) is a potential trace greenhouse gas. The concentration of N₂O was 0.275 ppm before the industrial revolution, and is now 0.324 ppm, an increase of about 18% (Chapuis-Lardy et al., 2007). Although concentration of N₂O in the atmosphere is very low, its greenhouse effect is very high. The enhanced greenhouse effect of N₂O is about 296 times that of CO₂, and its contribution to climate warming is about 6% (IPCC, 2013).

In the mid-latitudes of northern hemisphere, N₂O emissions of rivers, estuaries and continental shelves account for 35% of aquatic emissions, of which estuaries are the main source (Seitzinger et al., 2000). Annual N₂O emission from natural wetlands is $0.97 \text{ Tg}\cdot\text{yr}^{-1}$ (Tian et al., 2015), however drainage can increase it up to $1.5 \text{ Tg}\cdot\text{yr}^{-1}$. The highest emission comes from tropical and subtropical drained organic soils (Pärn et al., 2018). In recent decades, the high-intensity human activities have led to increasing nitrogen (N) input in estuarine

areas acting as a natural barrier for purifying terrestrial pollutants and reducing their flux into the sea, estuarine wetlands play a very important role in controlling the nutrient status of estuary and offshore waters and the global N cycle (Nedwell, 1996). On the other hand, increased nitrogen loading such as fertilization may turn coastal and estuarine wetlands from sinks to sources of N₂O (Zhang et al., 2013; Chmura et al., 2016; Martin et al., 2018; Mou et al., 2019). High N input stimulates microbial life leading to significant increase in denitrification intensity and, thus, N₂O emission (Moseman-Valtierra et al., 2011).

Denitrification is the main N₂O source in the estuary sediments, with NO₃⁻ successively converted into NO₂⁻, NO, N₂O and N₂ (Jorgensen et al., 1984; Law et al., 1992). It is affected by environmental factors such as soil temperature (Barnes and Owens, 1998), soil moisture content (Bai et al., 2017), salinity (Zhang et al., 2018), content of total carbon (TC), total nitrogen (TN), and nitrate (Dong et al., 2000) in sediments.

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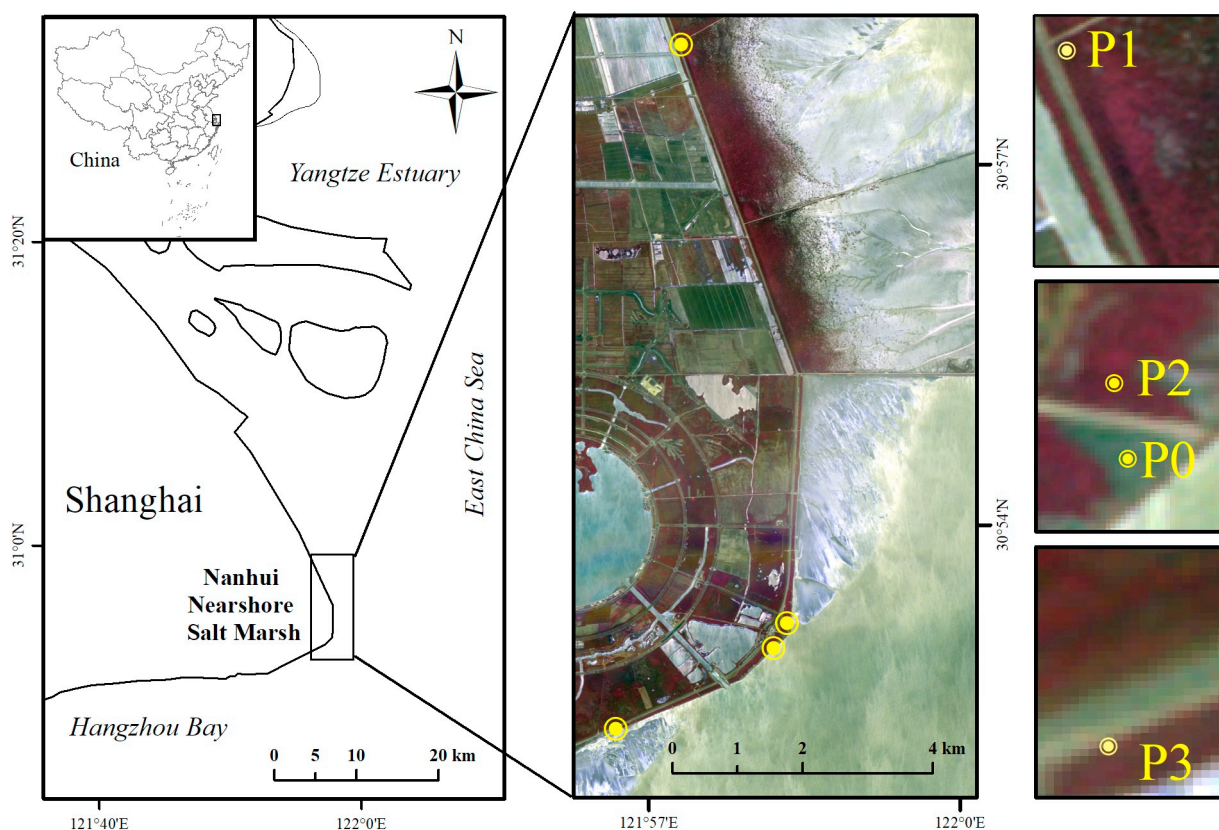


Fig. 1. Location of the four sampling plots in the Nanhui shore salt marsh of the Yangtze estuary, Shanghai, China (Photo by RapidEye Satellite).

At present, the impact of salinity on the sediment N_2O emission of estuary wetland is inconsistently known. Salinity affects microbial and plant activity and accelerates sediment C and N mineralization (Tejada et al., 2006). Many studies show that microbial activity is limited by excessive salt through osmotic stress (Smith et al., 2003; Zhang et al., 2018). On the other hand, some studies show no significant correlation between the sediment salinity and N_2O fluxes (Amouroux et al., 2002; Kontopoulou et al., 2015).

Aquatic macrophytes are important regulators of gaseous fluxes in coastal marshes whereas the impact varies among species (Wang et al., 2007; Yin et al., 2015; Yuan et al., 2015). The role of *Spartina alterniflora*, an invasive species in Chinese coastal marshes, has been reported in several papers considering N_2O emissions (Wang et al., 2007; Yin et al., 2015; Yuan et al., 2015; Xu et al., 2017; Song et al., 2018; D. Z. Gao et al., 2019a; G. F. Gao et al., 2019b; Mou et al., 2019; Zhang et al., 2019a), however, the results are by and large contrasting. Thus, Yuan et al. (2015) found that annual N_2O emissions in *S. alterniflora* and *Phragmites australis* marshes were -0.51 and -0.25 $\text{kg N}_2\text{O}\cdot\text{ha}^{-1}$, respectively while open water, a bare tidal flat and a *Suaeda salsa* marsh emitted 0.24 – 0.56 $\text{kg N}_2\text{O}\cdot\text{ha}^{-1}$. Decrease in N_2O emissions by *Spartina* invasion was reported by Mou et al. (2019) who demonstrated that *S. alterniflora* invasion did not significantly influence N_2O emissions from the natural *Cyperus malaccensis* marsh. After N addition, the invasion of *S. alterniflora* decreased N_2O emissions, owing to its stronger N uptake capacity. In contrast, Zhang et al., 2019a found that N_2O emission rate in several coastal marshes increased with *Spartina* invasion. Similarly, Gao et al., 2019b demonstrated that *S. alterniflora* invasion significantly increased soil N_2O emissions in mangrove wetlands. On the other hand, Gao et al., 2019a found that *Spartina* invasion enhanced both production and consumption of N_2O in coastal marshes: potential gross N_2O production and consumption rates were higher in *S. alterniflora* and *P. australis* stands compared to a *Scirpus mariqueter* stand and a bare mudflat. Thus, *Spartina*'s impact on N_2O fluxes is still unclear.

Affected by sewage, the water of the Yangtze River estuary and Hangzhou bay coastal waters suffers from serious eutrophication. An over 1 $\text{mg}\cdot\text{L}^{-1}$ average concentration of dissolved inorganic nitrogen (DIN) has been recorded (Chen et al., 2005), higher than the national standard for water quality (0.5 $\text{mg}\cdot\text{L}^{-1}$). Several studies have demonstrated that increased N loading increases N_2O emissions from coastal and estuarine wetlands (Moseman-Valtierra et al., 2011; Zhang et al., 2013; Chmura et al., 2016; Martin et al., 2018; Mou et al., 2019). The Nanhui coast and Yangtze estuary are located at the intersection of fresh and brackish water. Thus, physical and chemical characteristics of the tidal sediment have a complex spatial-temporal pattern which, among other factors, determines the soil microbial community structure and N_2O emission.

The main aim of this study was to analyse N_2O fluxes in coastal salt marshes covered by invasive *S. alterniflora* for following aspects: (a) temporal and spatial variation of N_2O fluxes and (b) plants and physical factors of the N_2O fluxes.

2. Materials and methods

2.1. Study site

The study was conducted in an intertidal salt marsh in the Nanhui coastal zone which is located in Shanghai, China ($30^{\circ}50'$ – $31^{\circ}10'$ N, $121^{\circ}50'$ – $122^{\circ}10'$ E) and interacts with waters of the Yangtze estuary, Hangzhou Bay and East China Sea. The tides have moderate intensity and an irregular semi-diurnal cycle. The plant community is dominated by the exotic Atlantic cordgrass (*Spartina alterniflora*) species. The Nanhui nearshore wetland has a Far East monsoon climate with an average annual temperature of 15.9 $^{\circ}\text{C}$ and annual precipitation of 1222 mm. Four plots: P1, P2, P3 and P0 (Bare flat) with highest salinity at P1 and different vegetation history were selected for the study. The plant community at P1 was dominated by *S. alterniflora*, and mixed

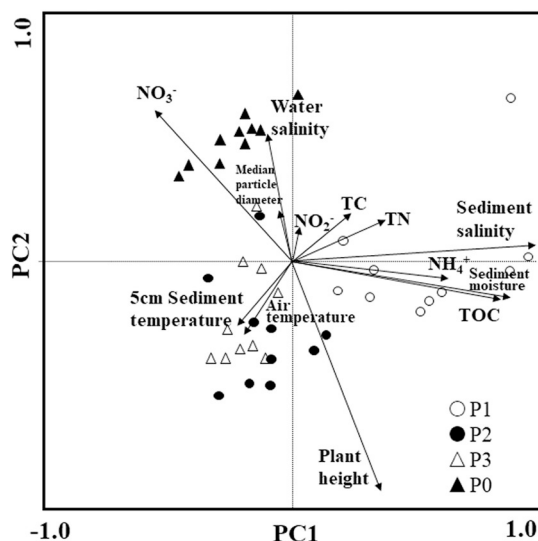


Fig. 2. Principal component analysis ordination plot showing grouping of soil samples according to environmental parameters. The first principal component (PC1) explained 50.5% of total variation, and the second principal component (PC2) explained an additional 22.2%. Abbreviations: total carbon (TC), total organic carbon (TOC), total nitrogen (TN).

Scirpus mariqueter; P2 was dominated by *S. alterniflora*, P3 was located at a newly developed *S. alterniflora* bed, and P0 was a bare mudflat (Fig. 1). P1 and P2 were occupied by vegetation already in 2007, whereas the grass in P3 was only two years old. Henceforth, P2 and P3 are referred to as old and young *Spartina* site, respectively.

During last decades this particular seashore area has experienced significant changes. Due to the dramatic sediment decline after the construction of the Three Gorges Dam and other dams in the catchment of Yangtze River, accretion of the delta has slowed down while the outer delta front has eroded (Xu et al., 2006). On the other hand, sea-level rise threatens the coastal zone. To protect the coastline and newly reclaimed areas a seawall was constructed in the Nanhui area in 2002. Recently, during the post-reclamation period, efforts for protection and maintenance of vegetation outside the seawall have been made for safety inside the seawall (Li et al., 2013, 2014).

2.2. Experimental design and measurement of gas fluxes

The N₂O fluxes were measured with a gas chromatography technique, using transparent polymethyl methacrylate chambers (28 cm diameter, 80 cm height) and collars (30 cm diameter, 20 cm height) with a 5 cm groove, inserted into the soil 15 cm deep. Inside each chamber, an electric fan was used to homogenize the chamber air, and a thermometer was fixed to monitor temperature. A triple valve was used to collect the gas samples. During the observation, the chamber was placed into the groove filled with water to prevent gas exchange with the atmosphere. Plants were included in chambers P1, P2 and P3, with five chambers in each plot.

The gas samples were taken at 8:00 am during the low tides once a month from March 2017 to January 2018, using 100 mL syringes to collect 100 mL of chamber gas into pre-evacuated gas sampling bags at 0, 10, 20 and 30 min during a 30 min period. Concentrations of N₂O in the samples were analysed within 36 h using gas chromatography with an electron capture detector, advanced flow controller, packed column, and N₂ as carrier gas (GC 2014, Shimadzu, Japan).

2.3. Environmental data

Soil temperature at 5 cm depth was measured three times in each collar during each sampling session. Soil and water samples were taken

three times as replicates at each plot. Soil samples were dried for gravimetric soil moisture content and sediment texture analyses with a laser particle size analyser (LS13 320, Beckman Coulter, America). An element analyser (Vario EL III, Elementar, Germany) was used to measure total carbon (TC), total nitrogen (TN) and total organic carbon (TOC) content. Soil samples were leached with 0.1 M HCl to remove carbonate for the measurement of TOC. Soil salinity expressed by electrical conductivity was measured in the soaked-out soil with a 5:1 water: soil ratio. Seawater salinity was measured with a salinity meter (SX836, SanXin, China) and NO₃⁻-N, NO₂⁻-N and NH₄⁺-N concentration in water was determined using a continuous-flow nutrient analyser (SAN plus, Skalar, the Netherlands) according to standard methods (APHA-AWWA-WEF, 2005).

2.4. Statistical analysis

Statistical analysis was conducted with SPSS 20.0, while the figures were plotted with the Origin 9.0 software. One-way analysis of variance (ANOVA) and Least Significant Difference (LSD) post hoc tests were applied to evaluate the significance of the differences between plots in environmental factors and N₂O emission values. Logarithmic transformations were applied as appropriate to ensure that ANOVA assumptions of normality and/or homoscedasticity were met. Principal component analysis (PCA) was used to display and compare the patterns of environmental factors among different studied plots. Pearson and Spearman correlation analyses were used to characterize relationships between N₂O flux and various environmental parameters. The *p* < .05 level of significance was accepted in all cases.

3. Results

3.1. Temporal and spatial variation of environmental factors

The environmental parameters varied between the studied plots (Fig. 2 and Supplementary Table S1). The first two principal components described 72.7% of total variance. PC1 was mostly determined by the sediment characteristics (salinity, temperature, moisture, TC and TN) and NH₄⁺ and NO₃⁻ content in sediment water, whereas PC2 was explained by plant height and water salinity. According to the PCA, P0 differed from other plots mainly in absence of plants, higher NO₃⁻ and water salinity. P1 was characterised by higher sediment salinity, sediment moisture, TOC and NH₄⁺ values, and lower sediment temperature compared to the other plots. P2 and P3 were clustered close to each other. They had higher sediment temperature values and smaller TC and TN concentrations compared to the other plots.

Air and sediment temperature showed similar variations in the four plots, with the highest temperature in July and the lowest in January (Fig. 3). Selected environment factors measured in the four plots are shown in Table 1. Physical and chemical characteristics of sediment

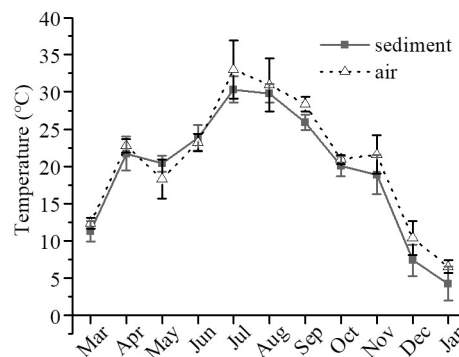


Fig. 3. Monthly mean and standard deviation of air and sediment temperature in the four plots from March 2017 to January 2018.

Table 1
Mean ± standard deviation values of sediment characteristics in the four plots.

Plot ID	Salinity	Moisture	MPD ¹	TOC	TC	TN	TC/TN
	(mS/cm)	(%)	(µm)	(g/kg)	(g/kg)	(g/kg)	
P1	3.51 ± 1.29 ^a	106.7 ± 39.6 ^a	6.3 ± 1.4 ^a	9.37 ± 1.70 ^a	18.33 ± 0.66 ^a	1.31 ± 0.18 ^a	14.2 ± 1.48 ^a
P2	1.25 ± 0.38 ^b	50.6 ± 5.9 ^b	19.5 ± 2.7 ^b	2.80 ± 0.89 ^b	12.68 ± 1.05 ^b	0.47 ± 0.08 ^b	27.6 ± 2.83 ^b
P3	1.05 ± 0.19 ^b	46.7 ± 5.7 ^b	28.7 ± 6.1 ^c	2.55 ± 0.66 ^c	12.36 ± 1.12 ^b	0.37 ± 0.07 ^b	33.6 ± 3.27 ^c
P0	1.08 ± 0.15 ^b	40.3 ± 6.6 ^b	29.7 ± 10.7 ^c	1.75 ± 1.22 ^c	11.99 ± 1.30 ^b	0.37 ± 0.14 ^b	38.0 ± 8.32 ^c

1: MPD: Median particle diameter.

Different lowercase letters indicate significant differences among plots ($p < .05$) according to the LSD test.

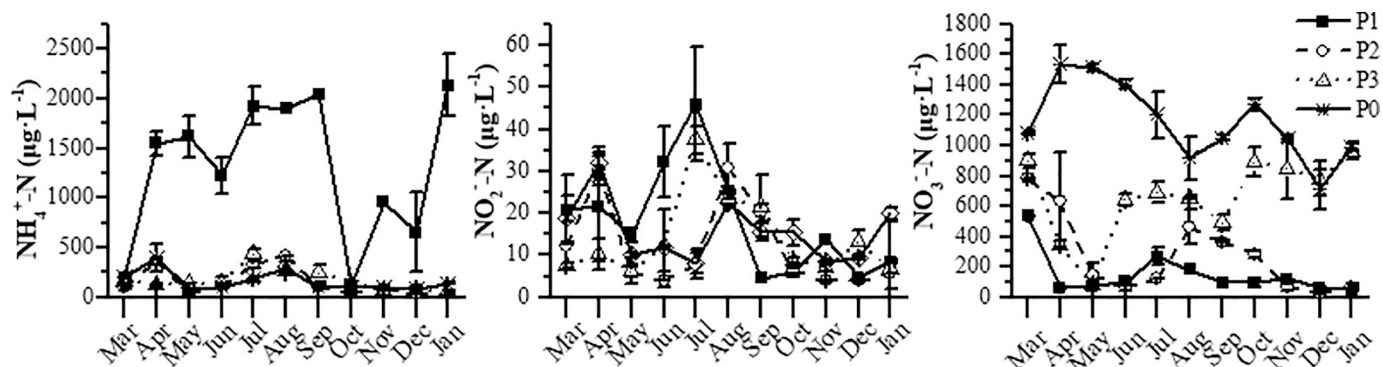


Fig. 4. Monthly mean and standard deviation of NH_4^+ , NO_2^- and NO_3^- ($\mu\text{g}\cdot\text{L}^{-1}$) in the four plots from March 2017 to January 2018.

such as salinity, moisture, median particle diameter, TC, TN and TOC at P1 were significantly different from other plots ($p < .05$). This is likely related to differences in relief, hydrology and species composition. NO_3^- -N concentration in P0 ranged from 712 to 1535 $\mu\text{g}\cdot\text{L}^{-1}$, with an average of 1172 $\mu\text{g}\cdot\text{L}^{-1}$. This was higher than those in other plots (Fig. 4). Average NO_2^- -N concentrations ranged from 3.9 to 46 $\mu\text{g}\cdot\text{L}^{-1}$. This was lower than NO_3^- and NH_4^+ concentrations and showed similar tendency among the four plots, peaking in July and August.

3.2. Temporal and spatial variation of N_2O fluxes

Temporal and spatial variation of N_2O fluxes is shown in Fig. 5. Across all sampling periods, monthly average N_2O fluxes ranged from

– 41.9 to 39.3 $\mu\text{g}\text{N}_2\text{O}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ among the four plots (Tables 2 and S2). Maximum N_2O emission was measured in July at P2, while maximum N_2O consumption was measured in November at P2. The young *Spartina* marsh (P3) showed lower average N_2O emission than the older P2 stand. In general, emission of N_2O showed a peak in spring and summer while consumption dominated in the three plots with *S. alterniflora* in autumn and winter. Significant differences were observed between sampling periods and plots ($p < .05$, Tables 2 and S2). Conversely, N_2O fluxes in the bare mudflat (P0) consumed N_2O during all sampling periods except the summer. Average annual N_2O fluxes were graded as $\text{P2} > \text{P1} > \text{P3} > \text{P0}$ (Table 2).

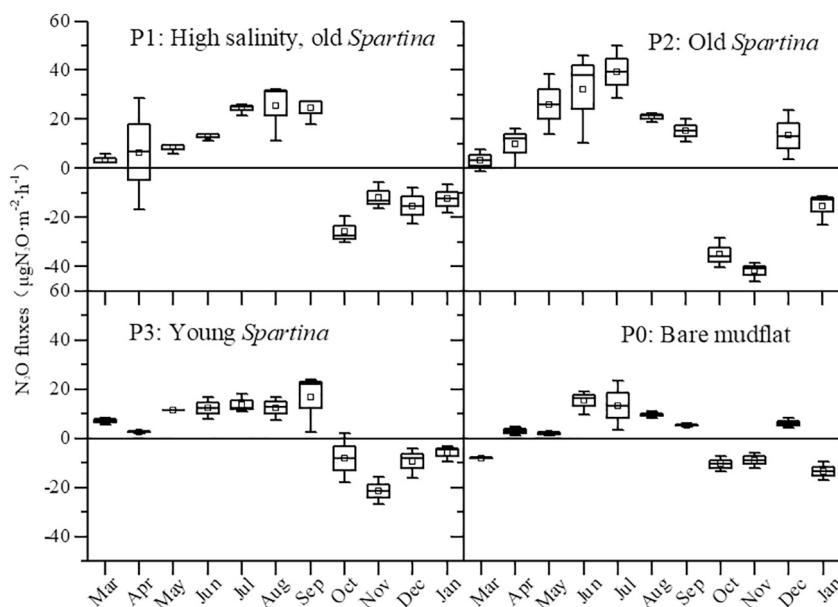


Fig. 5. Dynamics of N_2O fluxes in the study plots. The central line is the median and the dot is the mean, the edges of the box are the 25th and 75th percentiles, and the whiskers represent the 95% confidence interval.

Table 2
Seasonal N₂O fluxes (μg N₂O·m⁻²·h⁻¹) at the four plots.

Plot	Annual average	Max	Max	Average values for seasons			
ID	emission	consumption	consumption	Spring (March–May)	Summer (Jun. – Sep.)	Autumn (Oct. - Nov.)	Winter (Dec. – Jan.)
P1	–25.7 to 24.9	Aug.	Oct.	6.0 ^a	21.6 ^b	–18.6 ^c	–13.8 ^c
P2	–41.9 to 39.3	July	Nov.	12.9 ^a	26.7 ^a	–38.4 ^b	–1.1 ^c
P3	–21.4 to 16.3	Sept.	Sept.	7.1 ^a	13.7 ^a	–14.7 ^b	–7.7 ^c
P0	–13.4 to 15.1	June	Jan.	–1.0 ^b	10.9 ^a	–9.7 ^c	–3.6 ^{bc}

Lowercase letters indicate significant differences between seasons in the same plot (*p* < .05) according to the LSD test.

4. Discussion

4.1. Comparison with other tidal ecosystems

Our literature analysis covering 35 various tidal ecosystems showed that the *Spartina* marshes are emitting less N₂O than the bare mudflats (including the lab soil measurements), non-*Spartina* salt marshes and mangroves. The corresponding average (median) emission values were 20 (11), 103 (44), 29 (10) and 106 (13) μg N₂O·N·m⁻²·h⁻¹. Likewise, emissions were the lowest in the *Spartina* marshes (quartile range from 5 to 13 μg N₂O·N·m⁻²·h⁻¹ (Table 3).

Comparing with intensively managed grasslands on organic soils in subtropical areas, the vegetated tidal marshlands emitted up to 6 times less N₂O (IPCC, 2014). Several freshwater wetlands of subtropical and warm temperate zone emit more N₂O than *Spartina* marshes (Marín-Muñiz et al., 2015).

Salinity and nitrogen availability in sediments are likely leading factors of N₂O fluxes in *Spartina* marshes. Mesocosm studies by Zhang et al. (2018) demonstrated that N addition significantly enhanced N₂O fluxes from *Spartina* plots. Likewise, excessive N enhances N₂O emission

Table 3
N₂O emission (μg N₂O·N·m⁻²·h⁻¹) from various inter-tidal ecosystems. See Table S3 for data source.

	Mudflat or soil sample	<i>Spartina</i> salt marsh	Non- <i>Spartina</i> salt marsh	Mangroves
Sample size	5	8	8	13
Lower quartile	43	5.0	6.4	1.4
Median	44	11	10	13
Upper quartile	75	13	55	49
Average	103	20	29	106

in all tidal ecosystems (Moseman-Valtierra et al., 2011; Chmura et al., 2016). In our study, *Spartina*'s seasonal growth determined the N₂O flux showing that the plants were mediating it (Fig. 5). In the dormancy period the marshes consumed N₂O. Most likely this was due to competition between the plants and microbial communities (Burke et al., 2002; Yang et al., 2019; Zhang et al., 2019b). Interestingly, under higher salinity *Spartina* significantly decreased N₂O flux over the growth period, down to a negative (Yuan et al., 2015). One can assume that this was due to suppressed microbial processes and greater plant uptake under the saline conditions. It is also possible that the N-fixing diazotroph microflora in *Spartina* rhizosphere play a role in N supply for both plants and microbes (Gamble et al., 2010). The higher uptake by plants growing on sediments with elevated salinity is supported by laboratory experiments in which clipping of *S. alterniflora* and *Phragmites australis* significantly enhanced N₂O emission (Cheng et al., 2007; Yin et al., 2015). Fig. 6 generalises the possible relationship between N₂O fluxes and plant seasonal activities under two contrasting salinity conditions. For comparison we present N₂O emission in a *Cyperus malaccensis* freshwater marsh in Min River estuary, China (Wang et al., 2018). First of all, the fluxes were significantly higher than in the saline and brackish coastal marshes, and second, the emission fluctuated significantly during the growth period (Fig. 6). This was probably due to the pulsing effect caused by water level fluctuations during the monsoon period. Similar pulsing enhanced N₂O emissions in other freshwater wetland ecosystems (Mander et al., 2015).

4.2. Correlation between N₂O fluxes and environmental variables

For better comparison of N₂O fluxes between warmer and cooler periods the four-season-based data (Table 2) have been grouped in two – March to September and October to January (Table 4). In general, we can see that the correlation between the N₂O flux and environmental

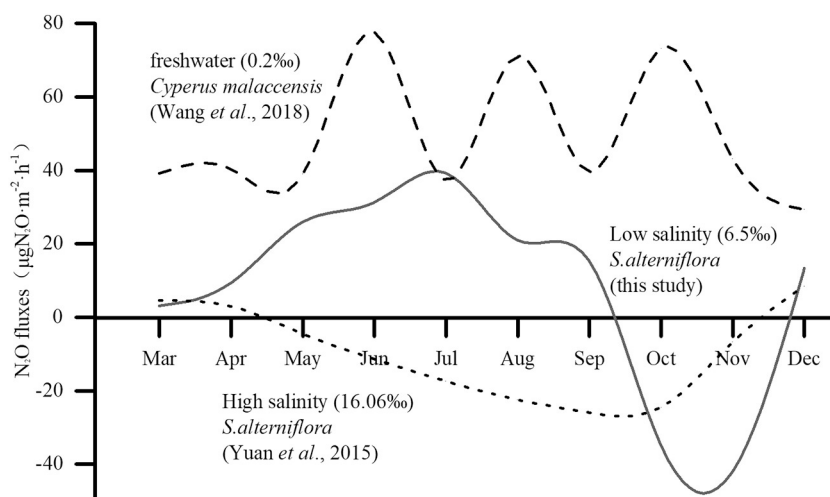


Fig. 6. Generalised annual dynamics of N₂O flux in the *Spartina alterniflora* brackish marsh of this study, the *S. alterniflora* salt marsh in Yuan et al. (2015) and a *Cyperus malaccensis* freshwater marsh in Wang et al. (2018).

Table 4
Correlations between N₂O fluxes and environmental factors in different plots and time groups.

Plot ID	month	air temperature	sediment temperature	sediment moisture	sediment salinity	water salinity	Median diameter	Plant height	NO ₃ ⁻	NO ₂ ⁻	NH ₄ ⁺	TOC	TC	TN
P1	Mar.-Sep.	0.890**	0.961**	-0.137	0.079	-0.626	-0.314	0.950**	-0.217	0.774*	0.192	-0.55	0.884**	0.812*
	Oct.-Jan.	-0.454	-0.521	-0.328	0.814	0.443	0.713	-0.443	-0.108	0.727	0.558	0.838	-0.721	-0.739
P2	Mar.-Sep.	0.651	0.673	0.434	-0.113	-0.423	-0.807	0.502	-0.925**	0.094	-0.521	-0.184	0.326	0.364
	Oct.-Jan.	-0.762	-0.737	0.831	0.926	0.588	-0.802	-0.522	-0.544	-0.754	-0.296	0.904	-0.796	0.803
P3	Mar.-Sep.	0.541	0.573	0.455	-0.186	-0.429	0.955	0.813*	0.067	0.558	0.567	-0.629	0.917*	-0.162
	Oct.-Jan.	-0.731	-0.633	0.061	0.93	0.545	-0.103	-0.161	0.416	0.21	0.245	0.782	0.106	0.106
P0	Mar.-Sep.	0.840*	0.856*	-0.059	-0.777*	-0.716	-0.318	-0.156	0.036	-0.156	-0.335	-0.425	0.947*	0.596
	Oct.-Jan.	-0.266	-0.307	0.543	0.756	0.071	-0.905	0.704**	-0.791	-0.746	-0.663	-0.026	0.906	0.905
P0-P3	Mar.-Sep.	0.550**	0.612**	0.194	0.065	-0.513**	-0.327	0.704**	-0.544**	0.261	-0.08	0.063	0.327	0.255
	Oct.-Jan.	-0.505*	-0.44	-0.115	-0.013	0.409	0.129	-0.343	0.247	0.018	0.186	-0.027	-0.375	-0.346

* - significant correlation ($p < .05$), ** - highly significant correlation ($p < .01$).

factors clearly depended on the season showing higher concentration values in warmer period. That is coherent with the growth of *Spartina*, which actively regulates N₂O fluxes (Fig. 5). Another important vegetation-related factor is the age of the *Spartina* stand. In the mature P2 *S. alterniflora* stand N₂O fluxes were significantly higher than those in the younger P3 stand (Table 2), although environmental conditions in these sites did not differ significantly. Likewise, in P2 NO₃⁻ concentration in sediment water strongly determined the N₂O flux; in warm period the correlation coefficient was as high as 0.925; Table 4) whereas in all other sites no significant correlation between the N₂O flux and NO₃⁻ was found. N₂O fluxes positively correlated with air temperature in P1 ($p < .01$), P0 ($p < .05$) and P0-P3 ($p < .05$), and with sediment temperature in P1 ($p < .01$), P0 ($p < .05$) and P0-P3 ($p < .01$) (Table 4). Previous studies also showed that temperature can control N₂O flux (Rask et al., 2002). For instance, the 5 cm sediment temperature was the main factor of N₂O emission in the Yangtze River tidal flat in summer (Wang et al., 2007).

N₂O fluxes showed negative correlation with NO₃⁻ concentration in P2 ($p < .01$). The result was inconsistent with most previous studies which indicated that NO₃⁻ promotes denitrification and N₂O emission (Moseman-Valtierra et al., 2011). As other authors report (Tauchnitz et al., 2008; Sun et al., 2017), plants may use up the NO₃⁻ for growth in spring and summer. This is supported by the dynamics of NO₃⁻ content in P0 driven by tides while NO₃⁻ content in P1-P3 reflected NO₃⁻ take-up by plants.

Correlation between N₂O flux and NH₄⁺ content was insignificant among the four plots. For instance, NH₄⁺ concentration in P1 was significantly higher than that in the other plots ($p < .05$), but N₂O emission in P1 was not different from other plots. DIN in P0 ranged from 0.79 to 1.97 mg/L with an average 1.33 mg/L. That was higher than Category Four standard of sea water quality (DIN ≤ 0.5 mg/L, GB3097-1997). The Nanhui nearshore salt marsh was dominated by tidal inputs of NO₃⁻ eutrophic tide, while NH₄⁺ was not a main limiting factor and it did not directly fuel N₂O emission. Positive correlation ($p < .05$) between N₂O flux and NO₂⁻ concentration in P1 lasted from March till September.

Plant roots release root exudates and exfoliants as substrates for soil microbial activity (Schlesinger, 1997). Environmental characteristics of P2 such as sediment salinity, water salinity and sediment moisture content were similar to that of P3, but plant height and age of development at P2 were higher than P3. N₂O flux at P2 was higher than P3. The height of N₂O emission was also the peak period of plant growth. N₂O fluxes in P0 were low, and significantly correlated with TC ($P < .05$). This should be related to the fact that soil microorganisms only had sediment C and N for nitrification and denitrification in the bare mudflat. The N₂O was probably converted to N₂ with N₂O reductase (controlled by *nosZ* genes) under the anoxic conditions (Zumft, 1997; Kolb and Horn, 2012). The activity of plants was higher in spring and summer. Oxygen content in the soil around the plant roots was smaller in the autumn and winter. This promoted the reduction of N₂O into N₂. A sealing experiment showed that *S. alterniflora* could be used as a transport channel of N₂O (Li, 2015). Spatial variation of greenhouse gas emissions in the Yancheng coastal wetland was mainly attributed to the type of halophyte vegetation (Xu et al., 2014). All that showed that plants promote N₂O emission.

To the background of global climate change, sea level rise and saltwater invasion are serious threats to salt marshes (Williams and Rosenheim, 2015). Salinity plays an important role in the biogeochemical cycle of carbon and nitrogen in estuaries and coastal wetlands. This study found no effect of sediment salinity on temporal variations of N₂O emission, which might have been due to long-term adaptation of microorganisms to sediment salinity. However, spatial variations of N₂O emission were affected by sediment salinity. Likewise, salinity could indirectly affect N₂O emission through an impact on TOC, TC or TN levels (compare the values in Table S4). In P1 under relatively high salinity the N₂O flux was lower than that in P2, P3 and P0, although the

TC/TN ratio in P1 was significantly lower. In freshwater ecosystems TC/TN below 15–20 can rapidly enhance N₂O emission (Klemetsson et al., 2005) but in saline conditions the relations can be different. Wang et al. (2012) showed that with the increase of salinity on *Phragmites australis* marsh, sediment characteristics significantly changed in the Minjiang estuary from fresh to brackish water. The N₂O emission in this study was higher than that under high salinity with *S. alterniflora* in Yancheng, China (Yuan et al., 2015; Fig. S3). High N₂O flux has been recorded in the brackish region of the estuary (Bange et al., 1996; Law et al., 1991). Sun et al. (2017) also found that high salinity could inhibit N₂O emission in the Yellow River estuary wetland.

Comparison of N₂O fluxes from *Spartina*-covered marshes and natural grasses shows sometimes higher fluxes for *Spartina* (e.g. comparison to *S. salsa*; Yuan et al., 2016), sometimes for natural grasses (Zhang et al., 2019a). However, in most cases there are no significant differences (Mou et al., 2019; Table 3) but the fluxes from natural grasses show higher variation (Table 3). In addition to relatively low N₂O emission potential by *S. alternifolia* beds, one should point out *Spartina*'s importance in bank protection. In Nanhai and similar post-reclamation sites potentially damaged by erosion, vegetation is needed for bank stabilisation. In addition to bank protection and low N₂O emission, other potential ecosystem services of *Spartina* such as provisioning biomass for bio-fertilisers, biochar, and raw material for pharmaceuticals (Qin et al., 2019) must be pointed out. When harvested in October, *Spartina* sites can be intensively used by migratory birds (unpublished observations from the Jiangsu coastal area, China). In autumn, when photosynthesis is still active, harvesting will decrease N₂O flux and also CH₄ emissions (Cheng et al., 2007; He et al., 2013)

5. Conclusion

This study found that *Spartina* promotes N₂O emissions during the growing season and inhibits them during plant dormancy. The *Spartina* areas and the bare mudflat showed no significant difference in annual average N₂O emissions. Nevertheless, we can conclude that *Spartina* stands are not significant N₂O sources compared to other tidal macrophytes (Table S3) and thus, they can be considered as a measure to mitigate N₂O emissions from tidal ecosystems.

Air and sediment temperature, sediment water NO₃⁻ and presence of plants were the main drivers of N₂O flux dynamics. However, NO₃⁻ is not a limiting factor of N₂O flux in the nitrogen-rich Yangtze estuary. Type and age of plant communities determined the temporal and spatial variability of N₂O flux. Sediment salinity affected spatial rather than temporal variability of N₂O flux under the relatively low salinity. Seasonal plant dynamics determined N₂O emissions whereas in an analogous study in more saline conditions the development of *Spartina* during the growing season was negatively correlated with N₂O fluxes causing the N₂O consumption instead of emission. No significant correlation was found between N₂O flux and environmental characteristics such as TOC, TN or sediment texture. The lack of correlation may be affected by complex abiotic and biotic interactions. Thus, the N₂O flux shifted between a source and a sink under the complex interactions. Frequent environmental changes due to seasonal and tidal cycles in estuaries and coastal areas complicate N₂O production and consumption processes. Therefore, further studies are needed, especially to characterize those mechanisms from a microbial perspective.

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Declaration of Interest Statement

None.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.ecoleng.2020.105758>.

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