Nitrous oxide emissions could reduce the blue carbon value of marshes on eutrophic estuaries

Brittney L Roughan¹, Lisa Kellman¹, Erin Smith² and Gail L Chmura³,⁴

¹ Department of Earth Sciences, St. Francis Xavier University, Antigonish, Nova Scotia, B2G 2W5, Canada
² Agriculture and Agri-Food Canada, Science and Technology Branch, Truro, Nova Scotia, B2N 2T6, Canada
³ Department of Geography, McGill University, Montreal, Quebec, H3A 0B9, Canada
⁴ Author to whom any correspondence should be addressed.

E-mail: gail.chmura@mcgill.ca

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Abstract

The supply of nitrogen to ecosystems has surpassed the Earth’s Planetary Boundary and its input to the marine environment has caused estuarine waters to become eutrophic. Excessive supply of nitrogen to salt marshes has been associated with shifts in species’ distribution and production, as well as marsh degradation and loss. Our study of salt marshes in agriculturally intensive watersheds shows that coastal eutrophication can have an additional impact. We measured gas fluxes from marsh soils and verified emissions of nitrous oxide ($\text{N}_2\text{O}$) in nitrogen-loaded marshes while the reference marsh was a sink for this gas. Salt marsh soils are extremely efficient carbon sinks, but emissions of $\text{N}_2\text{O}$, a greenhouse gas 298 times more potent than $\text{CO}_2$, reduces the value of the carbon sink, and in some marshes, may counterbalance any value of stored carbon towards mitigation of climate change.

Although more research is merited on the nitrogen transformations and carbon storage in eutrophic marshes, the possibility of significant $\text{N}_2\text{O}$ emissions should be considered when evaluating the market value of carbon in salt marshes subject to high levels of nitrogen loading.

1. Introduction

As Rockström notes, the supply of nitrogen (N) to ecosystems has surpassed the Earth’s Planetary Boundary (Rockström et al 2009) and N input to the marine environment has caused estuarine waters to become eutrophic, resulting in major ecological shifts. Excessive supply of N to estuarine salt marshes has been associated with shifts in species’ distribution (Bertness et al 2002) and production, the latter resulting in peat degradation and marsh loss (Deegan et al 2012). Salt marshes are intertidal ecosystems that provide many ecosystem services such as storm buffering and filtering of nutrients from coastal waters (Barbier et al 2011). Salt marshes are intertidal ecosystems that provide many ecosystem services such as storm buffering and filtering of nutrients from coastal waters (Barbier et al 2011). They have been widely recognized as important carbon sinks, for instance in the North American Climate, Clean Energy, and Environment Partnership Action Plan (Government of Canada 2016). The carbon stored in their soils has been branded as blue carbon (McLeod et al 2011) and standards are available for crediting this carbon on the voluntary carbon market (Verified Carbon Standard 2015). Through photosynthesis, salt marsh vegetation sequesters carbon dioxide ($\text{CO}_2$), largely as below-ground biomass in their saturated soils where decomposition is minimal. Globally, salt marsh soils may store as much as $4.8–87.2\ \text{Tg C yr}^{-1}$, an order of magnitude higher than terrestrial forest soils (McLeod et al 2011).

Salt marshes function as ‘nutrient filters’ for coastal waters by reducing excess N through storage in organic matter or conversion to N gases, including nitrous oxide ($\text{N}_2\text{O}$) (Megonigal and Neubauer 2009). The influence of fluctuating tides on the soils of salt marshes causes them to regularly switch from anoxic to oxic conditions, which allows the production of $\text{N}_2\text{O}$ through both denitrification and nitrification (Megonigal and Neubauer 2009). A potent greenhouse gas with a global warming potential (GWP) 298 times more potent than $\text{CO}_2$, $\text{N}_2\text{O}$ has become the third largest contributor to radiative forcing after methane ($\text{CH}_4$), which has a GWP of 34 (IPCC 2013). The GWPs of $\text{N}_2\text{O}$
and CH4 reflect the forcing of gases when emitted as a pulse, but their sustained fluxes from ecosystems have even greater climate feedbacks (Neubauer and Megonigal 2015) which we recognize as we compare our measured greenhouse gas fluxes from marshes to their capacity to serve as CO2 sinks. In addition to its contribution to climate warming, N2O is expected to remain the primary substance responsible for stratospheric ozone depletion throughout the 21st century (Ravishankara et al 2009).

Research has shown that experimental fertilization can shift salt marsh soils from sinks of N2O to sources (Cartaxana and Lloyd 1999, Kenny et al 2004, Blackwell et al 2010, Moseman-Valtierra et al 2011, Zhang et al 2013, Chmura et al 2016, Pastore et al 2017) or increase those that are already sources (Smith et al 1983). Thus, cultural eutrophication of coastal waters, i.e. nutrient enrichment from sewage treatment plants and runoff from intensive agriculture or urban land use, would be expected to generate similar responses (Smith et al 1983).

Studies of greenhouse gas fluxes in salt marshes on eutrophic waters have provided comparisons of different kinds of land use or fluxes from different coastal ecosystems. Adams et al (2012) conducted a field study on the hypernutritured Blackwater Estuary in England, comparing gas fluxes from soils of natural marshes and marshes that had been previously drained for agricultural use and reflushed. They found that both types of marshes were net sources of CH4 and N2O and that the relative magnitude of fluxes varied with land use. The marshes previously used for agriculture had higher fluxes of N2O, but lower fluxes of CH4. Over four seasons Livesley and Andrusiak (2012) measured N2O fluxes along a gradient with salt marsh situated between terrestrial vegetation and mangroves. Their sites were along Westernport Bay, Victoria, Australia where coastal land use was primarily agriculture, industry and urban. Seasonal flux of N2O from the salt marsh ranged from −1.58 to 2.39 μg m⁻² h⁻¹, and overall was a net source. In their field study Ford et al (2012) compared CH4 and N2O fluxes from soils of the grazed and ungrazed portion of a salt marsh on England’s Ribble Estuary. They recorded higher N2O flux from the grazed soil. Yang and Silver (2016) found positive and negative N2O fluxes in three zones of a salt marsh on California’s Tomales Bay watershed where cattle grazing was a source of NO3⁻–N. However, we have found no single field study that compares marshes on multiple eutrophic estuaries or eutrophic systems to a comparable system without excessive nutrient availability.

Our study compares salt marshes on multiple estuaries and includes a reference estuary. We questioned whether N2O fluxes from salt marshes located on watersheds with intensive agricultural land use and high N-loading on Prince Edward Island (PEI) in the Gulf of St. Lawrence are higher than those from our reference site—a salt marsh located in Kouchibouguac National Park, New Brunswick (table 1). Our study compared growing season N2O emissions from two grass-dominated vegetation zones characteristic of salt marshes along the northwestern Atlantic, the Spartina patens (commonly referred to as ‘high’ marsh) and Spartina alterniflora (commonly referred to as ‘low’ marsh) zones.

2. Methods

2.1. Study sites

The province of Prince Edward Island (PEI) has a cool maritime climate with a short growing season. The average, total rainfall for June to October (1981–2010) is 481 mm as measured at the Charlottetown A station (Environment Canada 2016). The average daily temperatures for June to October (1981–2010) range from 8.3°C in October to 18.7°C in July. Kouchibouguac National Park (Rexton station) shares a similar climate to PEI with an average, total rainfall for June to October (1981–2010) of 458 mm and an average daily temperature range (1981–2010) of 8.1°C in October to 19.4°C in July (Environment Canada 2016). Estuaries of PEI are subject to high levels of N-loading due to the intensive agriculture. Significantly higher NO3⁻–N concentrations were measured in a
PEI estuary with a watershed land use of 80% agriculture than one with 40% (Danielescu and MacQuarrie 2011); therefore, four salt marshes in watersheds with high agriculture land use (>50%) (PEI Department of Agriculture and Forestry 2003) and elevated predicted N-loads (Bugden et al 2014) were chosen as study sites in PEI (table 1). Our reference site, Kouchibouguac is located in the same region but has a predominantly forested watershed, low population density, and limited agriculture, therefore N-loading here is minimal (McIver et al 2015).

2.2. Experimental design and data analysis

We used an opaque, static chamber methodology with a vent tube on each chamber to convey changes in barometric pressure to headspace while minimizing gas leakage and contamination (Rochette 2011). An inner sampling tube perforated along its length and circulation of chamber air before sampling ensured collection of gas throughout the height of the chamber. Gas sample collection and analysis were carried out as described by Chmura et al (2011) using 17 L, 25 cm diameter chambers made of polycarbonate bottles covered with reflective material for insulation. Air was circulated by drawing it back and forth from the chamber using a large capacity syringe (60 ml) five times before collecting a sample. Changes in headspace concentration (gas flux) were calculated based on the Ideal Gas Law and the summation method (McVicar and Kellman 2014).

Gas samples were collected at Kouchibouguac in two previous studies by Chmura et al (2011) and Chmura et al (2016) which provided flux measurements from August 2006, July–November 2011 and April–June 2012.

Two weeks prior to gas sampling in PEI, we inserted PVC collars (25 cm-diameter, 10 cm high) into the soil. In the Tryon and DeSable salt marshes, 12 collars were installed in both the S. patens and S. alterniflora marsh zones, 3 m apart along a 36 m transect (for a total of 24 collars per marsh). In the Wilmot and Indian River salt marshes, four collars were installed in the S. patens marsh, 12 m apart along a 36 m transect. Relative elevation of collars was measured within each marsh using conventional surveying methods. In the Tryon marsh, the average elevation of collars in the S. alterniflora zone was 15 cm below those in the S. patens zone. In the DeSable marsh, the average elevation of collars in the S. alterniflora was an average of 5 cm higher than those in the S. patens zone, as the former were situated on a creek bank levee.

At PEI salt marshes, gas samples were collected monthly from June–October 2013. In the Wilmot and Indian River salt marshes, gas samples were collected from each collar plot over a one hour sampling interval at low tide. At the Tryon and DeSable salt marshes, gas samples were collected in both grass species from four unique collar plots during three separate sampling intervals—immediately after high tide receded, between high and low tide, and at low tide.

In PEI, soil porewater was collected from 15 cm depth within 1 m of each collar using a porewater sampler (Chmura et al 2011) during each sampling event outlined for gas sample collection. Porewater was transferred to acid-washed Nalgene bottles, stored on ice, and passed through a 0.45 µm filter. Porewater concentrations of NO$_3^-$ -N were analyzed colorimetrically by flow injection analysis on a Lachat QuickChem 8500 using Lachat Method 30-107-04-1-C.

The annual C storage, or blue carbon, potential of PEI salt marshes reported in this study (7.9 mol C m$^{-2}$ yr$^{-1}$) is the average of three measurements collected from Malpeque Bay, Brackley Bay, and Rustico Bay: 5.9, 7.4, and 10.4 mol C m$^{-2}$ yr$^{-1}$, respectively (Chmura and Hung 2004). Average N$_2$O and CH$_4$ fluxes were extrapolated to the eight month period over which marsh soils are unlikely to be frozen (April–November). Because no measurements were taken during the months of April, May and November in PEI, we used the average flux for October at each site to represent the missing data. For SGWP and SGWP calculations, we then calculated the average yearly flux by multiplying the average hourly flux over the eight months by 24 hours and then 244 days (April to November). For the reference site, we used the average April to November fluxes reported by Chmura et al (2016) and incorporated August flux data from Chmura et al (2011) by averaging the August fluxes from the two years.

Predictive Analytics Software statistics 18 and SPSS statistics 24 were used for statistical analysis. For each site, hourly gas fluxes were averaged to produce an average monthly flux. Because the data did not meet the requirements of normality of the residuals, the non-parametric Kruskal-Wallis test was performed. Pairwise comparisons for the Kruskal-Wallis test were corrected for overall error rate (5%) via the Bonferroni method. The difference between fluxes from S. patens and S. alterniflora was tested using the non-parametric Mann-Whitney U test. Fluxes from the Tryon and DeSable marshes were measured at different tidal stages and the Kruskal-Wallis test indicated no significant differences between the average high, mid- and low tide fluxes in the S. patens marsh. Thus, the average daily fluxes (using data from all three tidal stages) were compared to the Wilmot, Indian River, and Kouchibouguac fluxes, which only had measurements from one tidal stage. A constant was added to N$_2$O flux (19.35) and NO$_3^-$ -N values (0.001) so that all data (including negative or 0 values) could be log-transformed. To determine the relationship of fluxes and NO$_3^-$ -N porewater concentrations, we used the averages from four different plots during each month and each sampling event (low, mid or high tide) for the Tryon and DeSable salt marshes and same month for the Wilmot and Indian River salt marshes as
described above. Unless noted otherwise, all errors are reported as standard errors.

3. Results and discussion

In the four PEI N-loaded salt marshes, N₂O emissions from the S. patens marsh were higher than fluxes measured by Chmura et al. (2016) at the reference site, however only the average flux from DeSable was significantly higher (Kruskal-Wallis $p=0.003$). Lower letters denote significant differences between S. patens marsh zones. In both Tryon and DeSable, the average N₂O flux of the S. alterniflora zone was significantly higher than that in the corresponding S. patens zone (Mann-Whitney $p<0.01$). Error bars = ±1 SE.

Despite its lower areal extent, the total N₂O flux from the S. alterniflora marsh can be disproportionately large. For example, a comparison of the hourly emissions weighted by area reveals the S. alterniflora marsh emission of N₂O is approximately three times the S. patens marsh emission at our DeSable site. The average N-loaded N₂O emission we measured in the S. alterniflora marsh, 156.9 ± 88.8 $\mu$g m$^{-2}$ hr$^{-1}$, is substantially higher than fluxes reported in Massachusetts sites dominated by S. alterniflora (Emery and Fulweiler 2014). The difference in flux between these two vegetation zones merits further study to isolate the roles played by marsh elevation and species where N-loading is high.

One of the main drivers of N₂O flux in coastal systems is the availability of dissolved inorganic N (DIN) especially NO₃$^-$-N, which is the dominant form of N entering estuaries from rivers and groundwater (Megonigal and Neubauer 2009). In N-limited ecosystems, such as salt marshes, available NO₃$^-$-N is often used over N₂O as a terminal electron acceptor by denitrifying microorganisms and complete denitrification to N₂ is less likely (Megonigal and Neubauer 2009). Our mean porewater NO₃$^-$-N concentrations were positively correlated with mean N₂O fluxes (figure 2), explaining 53% of variation in N₂O flux across both the S. patens and S. alterniflora marsh ($p<0.001$).

We also measured CH₄ flux which did not significantly vary among marshes. We extrapolated both N₂O and CH₄ fluxes over the period when we assume soil is not frozen, then converted these values to sustained flux global warming potentials (SFGWP) and sustained flux global cooling potentials (SFGCP) (Neubauer and Megonigal 2015) to determine to what extent these two greenhouse gases would reduce the climate mitigation value (i.e. the annual C storage) of these salt marshes (table 2). We based our calculations on a 20 year time horizon as we cannot be certain of the period over which the high N-loading has occurred in the PEI estuaries. At our reference site, the CO₂ equivalence of the negative N₂O flux (i.e. uptake) was less than that of the CH₄ emissions so that, overall, the site sequestered 371.87 g CO₂-equivalents m$^{-2}$ yr$^{-1}$. The CO₂-equivalents of gas emissions from the N-loaded S. patens marshes range from 0.5 to 8.5 g CO₂-equivalents m$^{-2}$ yr$^{-1}$, reducing the value of the average CO₂ stored as soil organic matter (348.33 g m$^{-2}$ yr$^{-1}$) by up to 2.4%. More importantly, a comparison of the greater emissions from the S. alterniflora marsh at Tryon reduces the value of the CO₂ stored as organic matter by 16% while emissions from the S. alterniflora marsh in DeSable result in a SFGWP that almost completely offsets the SFGCP of the soil organic matter (327.6 g m$^{-2}$ yr$^{-1}$ or 94% of average C stored in PEI soil)—a difference that would be greater with the use of a 100 year time horizon. Thus, cultural eutrophication may transform a salt marsh from an ecosystem that mitigates global warming to one that contributes to global warming. Such marshes may

![Figure 1. Spartina patens marsh N₂O fluxes from the 4 PEI sites were higher than fluxes measured by Chmura et al. (2016) at the reference site (Kouchibougauac); however, only the average flux from DeSable was significantly higher (Kruskal-Wallis $p=0.003$, figure 1). The average S. patens marsh N₂O flux measured from these four marshes was 5.2 ± 1.6 $\mu$g m$^{-2}$ hr$^{-1}$. Although we had no reference site for the S. alterniflora marsh (its area was too small at the Kouchibougauac site), our results show notably higher fluxes in the S. alterniflora-dominated zone. At the two sites (Tryon and DeSable) where fluxes from both zones were measured, N₂O fluxes were significantly greater in the S. alterniflora marsh (figure 1, Mann-Whitney $p<0.01$). No differences were detected between tidal stages in the Tryon marsh S. alterniflora zone, but differences were detected in the DeSable marsh S. alterniflora zone (Kruskal-Wallis $p=0.007$), with the average high tide flux (630.5 ± 159.8 $\mu$g m$^{-2}$ hr$^{-1}$) significantly higher ($p=0.005$) than the low tide flux (52.3 ± 13.5 $\mu$g m$^{-2}$ hr$^{-1}$). The results of this study suggest that only measuring N₂O flux during one tidal stage may greatly over- or underestimate the average daily flux; therefore, future research should investigate how N₂O flux varies throughout a complete tidal cycle (through both flooding and ebbing tides). The lack of correspondence of N₂O fluxes in the PEI marshes to N-loading estimates indicates that it cannot be used as a direct predictor of N₂O. It is clear that other factors that drive NO₃$^-$ concentrations in soil porewater need to be explored.}
Figure 2. Relationship between N$_2$O flux and soil porewater NO$_3^-$-N concentration by month and vegetation zone (a) and site and vegetation zone (b). N$_2$O flux was positively correlated with NO$_3^-$-N concentration from four marshes and two vegetation zones in PEI ($r^2 = 0.53$, $p < 0.001$, $n = 61$). N$_2$O = 0.41 (NO$_3^-$-N) + 2.26. Analyses by marsh zone generate an $r^2 = 0.52$ ($n = 26$, $p < 0.001$) for S. alterniflora and an $r^2 = 0.12$ ($n = 35$, $p = 0.024$) for S. patens. Each sample point represents the mean N$_2$O and NO$_3^-$-N measurements of four plots taken during the same tidal sampling event (low, mid or high) and month. Error bars = ±1 SE.

Table 2. CO$_2$ equivalents (g CO$_2$ m$^{-2}$ yr$^{-1}$) of soil organic matter stored in PEI and reference marshes (Chmura et al. 2003) compared to CO$_2$ equivalents derived from SGCP and SGWP (Neubauer and Megonigal 2015) applied to gas fluxes over a 20 yr time horizon. Negative values indicate global cooling potential (uptake) and positive values indicate global warming.

| Salt marsh                | Soil storage | N$_2$O | S. patens marsh | CH$_4$ | sum   | N$_2$O | CH$_4$ | sum   |
|---------------------------|--------------|--------|-----------------|--------|-------|--------|--------|-------|
| DeSable                   | -348.33      | 8.29   | 0.19            | 8.48   | 300.46| 27.13  | 327.59 |
| Indian River              | -348.33      | 6.99   | 0.28            | 7.27   |       |        |        |       |
| Tryon                     | -348.33      | 3.39   | 0.20            | 3.60   | 15.35 | 41.17  | 56.52  |
| Wilmot                    | -348.33      | 0.24   | 0.24            | 0.48   |       |        |        |       |
| Kouchibouguac (reference) | -373.33      | -1.26  | 3.16            | 1.90   |       |        |        |       |

a SGCP multiplier = 264, SGWP multiplier = 250.
b SGWP multiplier = 96.

not provide CO$_2$ offsets and would have no value on carbon markets.

Our results, however, are in contrast to those of Pastore et al. (2017) who calculated the sustained global warming potential of fertilized and unfertilized plots and concluded that N$_2$O emissions from fertilized S. patens marsh were not high enough to counteract the effect of increased belowground C sequestration in fertilized plots. Differences in our finding could be due to the limited fertilization period (9 years) in the Maryland site or the method of calculating C sequestration rates and the use of a 100 year time horizon for the SFGWP and SFCCP. Pastore et al. (2017) based C sequestration rates on aboveground biomass and belowground gain as the product of soil elevation change over 9 years and the C density of the surface 10 cm of soil, while our study utilized C sequestration rates from representative marshes. These differences highlight the need for more studies that include analyses of both gas flux and C storage at marshes.

Our observation of elevated N$_2$O emissions as a consequence of cultural eutrophication suggest that the value of blue carbon in salt marshes will be reduced within N-loaded watersheds, and that N$_2$O flux should be quantified to properly assess the value of the annual C storage in such areas. Coastal eutrophication may
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