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Accelerated microbial organic matter mineralization following salt-water intrusion into tidal freshwater marsh soils

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Abstract The impact of salt-water intrusion on microbial organic carbon (C) mineralization in tidal freshwater marsh (TFM) soils was investigated in a year-long laboratory experiment in which intact soils were exposed to a simulated tidal cycle of freshwater or dilute salt-water. Gas fluxes [carbon dioxide (CO_2) and methane (CH_4)], rates of microbial processes (sulfate reduction and methanogenesis), and porewater and solid phase biogeochemistry were measured throughout the experiment. Flux rates of CO_2 and, surprisingly, CH_4 increased significantly following salt-water intrusion, and remained elevated relative to freshwater cores for 6 and 5 months, respectively. Following salt-water intrusion, rates of sulfate reduction increased significantly and remained higher than rates in the freshwater controls throughout the experiment. Rates of acetoclastic methanogenesis were higher than rates of hydrogenotrophic methanogenesis, but the rates did not differ by salinity treatment. Soil organic C content decreased significantly in soils experiencing salt-water intrusion. Estimates of total organic C mineralized in freshwater and salt-water amended soils over the 1-year experiment using gas flux measurements (18.2 and 24.9 mol C m^{-2}, respectively) were similar to estimates obtained from microbial rates (37.8 and 56.2 mol C m^{-2}, respectively), and to losses in soil organic C content (0 and 44.1 mol C m^{-2}, respectively). These findings indicate that salt-water intrusion stimulates microbial decomposition, accelerates the loss of organic C from TFM soils, and may put TFMs at risk of permanent inundation.

Keywords Tidal freshwater marshes · Carbon · Organic matter mineralization · Sulfate reduction · Methanogenesis · Carbon dioxide · Methane · Delaware River

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Introduction

Tidal marshes have existed for at least the past 4000 years, when rates of sea level rise slowed enough to allow for their development (Redfield 1965). Sea level exerts an especially powerful influence on tidal marshes (Morris et al. 2002; Mudd et al. 2009). Tidal marshes are found at or near

current mean local sea level, and maintain their elevation relative to rising sea levels through net accretion and vertical growth. Accretion in tidal marshes is largely driven by deposition of watershed-derived sediments and autochthonous organic matter produced by marsh macrophytes and the subsequent storage of these materials in marsh soils (Reed 1995; Morris et al. 2002). The rate of sea level rise has increased in the past century due to anthropogenic climate change, and future acceleration of sea level rise is predicted (Nakada and Inoue 2005; Church and White 2006). Increased rates of sea level rise may exceed the ability of some marshes to accrete vertically, resulting in permanent inundation and loss of marsh area (Reed 1995; Morris et al. 2002).

Tidal marshes provide many critical ecosystem services, and the response of these ecosystems to climate change and sea level rise has received considerable attention from the scientific community (e.g., Morris et al. 2002; Mudd et al. 2009). Much of the attention has been on salt marshes, however, and relatively less is known about the impacts of climate change on tidal freshwater marshes (TFMs; see Neubauer and Craft 2009). TFMs are found in the tidally influenced freshwater portions of many estuaries, and approximately 20% of total tidal marsh area along the Atlantic and Gulf Coasts of the United States is TFM (Odum 1988; Mitsch and Gosselink 1993). Both TFMs and salt marshes are highly productive ecosystems (Odum 1988), serve as key habitats for many organisms (Mitsch and Gosselink 1993), and are efficient filters that can reduce the loading of nutrients from watersheds to coastal waters (Neubauer et al. 2005a; Gribsholt et al. 2005). Additionally, tidal marshes absorb storm surge and wave energy (Yang 1998), minimizing flooding and damage to adjacent upland areas during coastal storm events (Barbier et al. 2008). Although TFMs and salt marshes are functionally similar in many ways, differences in salinity and solute concentrations [especially sulfate (SO_4^{2-}) and hydrogen sulfide (H_2S)] lead to significant differences in microbial biogeochemical processes and dominant plant communities between these wetland types.

Climate change is predicted to alter future patterns and rates of precipitation, evaporation, and evapotranspiration (Smith et al. 2005; Milly et al. 2005). The combination of rising sea-levels and decreased river (freshwater) discharge will result in the upriver migration of the freshwater-saltwater mixing zone (i.e., salt-water intrusion) in some estuaries (Hamilton 1990; Knowles 2002), with potentially significant impacts on ecosystems in the tidal freshwater zone, including TFMs. Salinity-induced stress on freshwater plant communities is projected to decrease primary production and organic matter accumulation rates (Willis and Hester 2004; McKee and Mendelssohn 1989; Spalding and Hester 2007). In addition, rates and pathways of microbial organic matter mineralization can shift in response to changing salinities (Rysgaard et al. 1999; Canavan et al. 2006; Weston et al. 2006). Due to low SO_4^{2-} availability in freshwater (<0.1 mmol L^{-1}), methanogenesis (MG) is often a major pathway of anaerobic organic matter mineralization (Capone and Kiene 1988; Kelley et al. 1990), although microbial iron reduction and denitrification can also be important processes in freshwater wetlands (Roden and Wetzel 1996; Neubauer et al. 2005b; Gribsholt et al. 2005). Microbially-mediated SO_4^{2-} reduction (SR) replaces MG as a dominant anaerobic terminal C mineralization process in marine sediments and salt marsh soils (Jørgensen 1982; Capone and Kiene 1988) due to the greater availability of SO_4^{2-} in seawater ($\sim$28 mmol L^{-1}) and the higher energy yield of organic C degradation coupled to SR as compared to MG (Capone and Kiene 1988; Mishra et al. 2003). Therefore, salt-water intrusion into TFMs will likely alter pathways and rates of elemental cycling and drive shifts in overall ecosystem structure and function.

Previous studies have documented a positive relationship between salinity and decomposition in marsh soils (Craft 2007), and a shift from MG to SR following salt-water intrusion into tidal freshwater estuarine sediments (Weston et al. 2006). While these studies have suggested that salt-water intrusion may increase overall rates of organic matter decomposition, the impact of climate change on microbial C cycling in TFM soils remains unclear. Increased organic matter decomposition in response to salt-water intrusion has profound implications for the persistence of TFMs in coastal landscapes. In this study, we incubated TFM cores in the laboratory under freshwater and dilute salt-water conditions and measured emissions of carbon dioxide (CO_2) and methane (CH_4), rates of SR and MG, and soil biogeochemistry throughout the 1-year experiment. We specifically excluded plants from the experimental design to minimize confounding factors,

such as changes in C inputs as plants grow and senesce, and salinity-related deaths of freshwater plants, to focus on how salt-water intrusion impacts rates and pathways of microbial organic matter mineralization.

Methods

Study site

The Delaware River is tidal as far north as Trenton, New Jersey, although saline water seldom reaches north of the Delaware–Pennsylvania border. Extensive TFMs are found along the main channel and in tributaries to the Delaware River between approximately Wilmington, Delaware and Trenton, New Jersey (Patrick et al. 1973; Field and Philipp 2000). We collected soils from the Woodbury Creek TFM (39° 51′ 33.05″ N, 75° 10′ 23.33″ W), approximately 2 km from the confluence of this small tributary and the Delaware River. This site is towards the lower end of the freshwater tidal portion of the Delaware River; just upriver of the highest reach of saline water in recent years. Vegetation at this site includes freshwater *Peltandra virginica* (arrow arum), *Pontederia cordata* (pickerelweed) and *Nuphar lutea* (yellow pond lily).

Experimental design

We collected 40 intact soil cores from the marsh platform at the Woodbury Creek study site at low tide in early spring (17 April 2006), before plants emerged. Soils were collected in 10 cm (i.d.) polyvinylchloride tubes to a depth of approximately 25 cm, sealed at the bottom with gas- and water-tight end caps, and transported to the laboratory. Two cores were sectioned the following day for initial porewater biogeochemical measurements (see *Soil Biogeochemistry* below). Holes were drilled in the core barrel just above the soil surface. Subsequently, cores were randomly assigned to two separate tidal tanks which were housed in an environmental chamber at 20°C in the dark. The tidal tanks (100 L each) allowed the core surface to be exposed to air for a period of 6 h (low tide) followed by 6 h of inundation (high tide). Both tidal tanks were initially filled with artificial freshwater (AFW; Table 1), which was changed several times a week to maintain constant water chemistry. AFW chemistry was chosen to represent average ion and nutrient concentrations in the freshwater Delaware River.

Table 1 Composition of artificial freshwater (AFW) and artificial seawater (ASW) used in the salt-water intrusion experiment

Component	AFW	ASW
Cl^- (mmol L^{-1})	1.01	77.59
Mg^{2+} (mmol L^{-1})	0.10	3.58
Ca^{2+} (mmol L^{-1})	0.27	1.45
Na^+ (mmol L^{-1})	0.91	74.73
K^+ (mmol L^{-1})	0.18	1.52
SO_4^{2-} (mmol L^{-1})	0.04	3.98
HCO_3^- (μmol L^{-1})	668.8	668.8
NH_4^+ (μmol L^{-1})	28.1	28.1
PO_4^{3-} (μmol L^{-1})	11.0	11.0
NO_3^- (μmol L^{-1})	98.9	98.9
Salinity	0.06	4.95

After a 2 week pre-incubation period (days −14 to 0), the water in one tidal tank was replaced with dilute artificial seawater (ASW; Table 1). The ASW had a salinity of approximately 5 (about 14% of full strength seawater), which was attained through increasing major ion concentrations in proportion to seawater while maintaining nutrient and inorganic C concentrations as in the AFW (Table 1). Cores were exposed to simulated tidal flooding and drainage with AFW or ASW for 1-year (days 0–365). The water in both tanks was changed at least once weekly (more often during the first months of the experiments). We measured concentrations of dissolved inorganic C (DIC), chloride (Cl^-), SO_4^{2-}, ammonium (NH_4^+), nitrate + nitrite (NO_x), and phosphate (PO_4^{3-}) in the tidal tanks several times per week to ensure relatively constant chemistry (see *Soil Biogeochemistry* for analytical methods).

Gas flux rates

We measured rates of CO_2 and CH_4 gas emission from the soil cores 2 to 3 times per week during the initial 6 months of the experiment and once weekly in the last 6 months. Gas fluxes were measured during the low-tide portion of the tidal cycle when the

soil surface was exposed. Cores were fitted with a gas-tight cap, providing approximately 1.2 L of headspace that was circulated with a small fan. An infra-red gas analyzer (PP Systems EGM-4) was connected to the cap in a flow-through configuration, and CO_2 concentration was measured in the headspace every 1 min for 10 min. When CO_2 measurements were complete, an initial headspace sample (3 mL) for CH_4 was obtained with a gas-tight syringe. Final CH_4 samples were obtained after approximately 1 h. CH_4 samples were analyzed immediately by flame ionization detection gas chromatography (Agilent 6890 N with Porapak Q column). Changes in CO_2 and CH_4 gas concentrations over time in the headspace were used to determine gas flux rates.

CO_2 gas flux rates were measured on all cores during each of 85 sampling dates for a total of 1453 CO_2 flux measurements. Due to logistical constraints, CH_4 flux was measured on a subset of 4 AFW and 4 ASW cores during each sampling (72 dates for a total of 618 CH_4 flux measurements). Equipment failure resulted in no CH_4 measurements from days 200 to 270.

To assess whether CO_2 and CH_4 flux rates differed between periods of core inundation and core exposure, we compared gaseous flux rates as described above with aqueous flux measurements. Aqueous flux rates were measured on duplicate cores from each treatment on 6 different dates (day 0, 5, 12, 27, 47 and 82; on day 0 only duplicate freshwater cores were incubated). Cores were capped without a gas headspace and incubated for approximately 8 h with continuous mixing of the overlying water. Water samples were obtained about every 2 h. For DIC measurements, 8 mL of headspace water was removed and placed into a glass vial, 50 μL of $HgCl_2$ was added to halt microbial activity, and the vial was capped without headspace. DIC concentrations were determined on a Shimadzu TOC-V_{CSH} instrument. For dissolved CH_4, 5 mL of sample was injected into a 12 mL headspace vial and preserved with 2 mL of 1 N HCl. Following equilibration, the concentration of CH_4 in the gas headspace of these vials was determined by gas chromatography. DIC and CH_4 flux rates under inundated conditions were then calculated from the changes in DIC and CH_4 concentrations in the flooded core headspace over time.

Rates of microbial sulfate reduction and methanogenesis

We sectioned soil cores periodically throughout the experiment to determine depth-specific rates of both microbial SR and MG and porewater and solid-phase biogeochemistry (see *Soil Biogeochemistry* below). Duplicate cores were sectioned after field collection (on day −14) and just prior to salt-water amendment (day 0). Duplicate cores were removed from the ASW tank and sectioned on days 5, 12, 27, 47, 82, 160 and 364, with sampling from the AFW tank occurring the following day. Due to the destructive nature of the sampling, the number of cores in each tidal tank decreased by two following each sampling timepoint.

Soil cores were sectioned in 2 cm depth increments to a depth of 20 cm in an O_2-free (N_2) atmosphere. Depth-specific rates of microbial SR, hydrogenotrophic MG (HMG) and acetoclastic MG (AMG) were determined on duplicate 2 cm^3 sub-samples from the 0–2, 2–4, 4–6, 8–10, 12–14 and 18–20 cm depths. Six intact sub-samples from each section were taken using 5 mL cut-off syringes that were immediately capped with silicon stoppers. Approximately 0.2 μCi of $^{35}SO_4^{2-}$, 1 μCi of $H^{14}CO_3^-$, and 0.2 μCi of $^{14}CH_3COOH$ were injected into separate sub-cores (2 each) and the samples were incubated at 20°C for 12-16 h. Sub-samples containing ^{35}S were then fixed in 10 mL of 20% zinc acetate and immediately frozen. Sub-samples containing ^{14}C were injected into a 12 mL headspace vial and immediately fixed with 2 mL of 6 N HCl to stop metabolic activity and convert DIC into CO_2. Activity of the total reduced sulfur (TRS) pool was quantified by liquid scintillation counting following cold distillation (Kallmeyer et al. 2004), and rates of SR were calculated as

$$SR = TR^{35}S \times \left(^{35}SO_4^{2-}\right)^{-1} \times \left[SO_4^{2-}\right] \times \varphi \times \alpha SR \times t^{-1} \quad (1)$$

where $^{35}SO_4^{2-}$ is the activity of the initial SO_4^{2-} added, $[SO_4^{2-}]$ is the concentration of SO_4^{2-} in the soil porewater, φ is the porosity of the soil (cm^3 water cm^{-3} soil), αSR is the isotope fractionation factor of SR (1.06; Jørgensen 1978) and t is incubation time.

The ^{14}C activities of CH_4 and CO_2 in MG samples were determined by gas chromatography. The gas headspace from acidified soil slurries was purged for 10 min with helium and trapped onto a 5 cm length

of Porapak Q column under liquid nitrogen. The trapped gases were then injected into a gas chromatograph (Agilent 6890 N) with a 1 m Porapak Q column for separation and quantification of CH_4 (by flame ionization detection) and CO_2 (by thermal conductivity detection), and quantification of $^{14}CH_4$ and $^{14}CO_2$ activities by gas counting (Raytest Raga Star). Purging and trapping efficiency was >99% for CH_4 and >95% for CO_2. The activity of samples was determined relative to the activity of $^{14}CO_2$ standards, after determining that the counting efficiency of $^{14}CH_4$ and $^{14}CO_2$ was equivalent. Rates of hydrogenotrophic HMG and AMG were quantified in a similar manner to SR rates (Eq. 1):

$$\mathrm{HMG} = {}^{14}\mathrm{CH_4} \times \left(\mathrm{DI}^{14}\mathrm{C}\right)^{-1} \times [\mathrm{DIC}] \times \varphi \times \alpha\mathrm{HM} \times \mathrm{t}^{-1} \quad (2)$$

$$\mathrm{AMG} = {}^{14}\mathrm{CH_4} \times \left({}^{14}\mathrm{CH_3COOH}\right)^{-1} \times [\mathrm{CH_3COOH}] \times \varphi \times \alpha\mathrm{AM} \times \mathrm{t}^{-1} \quad (3)$$

where $^{14}CH_4$ is the activity of the measured CH_4, ($DI^{14}C$) and ($^{14}CH_3COOH$) are the activities of the DIC and acetate additions, respectively, [DIC] and [CH_3COOH] are the porewater concentrations of DIC and acetate, respectively, and αHM and αAM are the isotope fractionation factors for HMG and AMG, respectively (both 1.06, Orcutt et al. 2005).

Total organic C (CH_2O) mineralized through each anaerobic microbial pathway was estimated assuming the following stoichiometries:

$$\mathrm{SR}: 2\mathrm{CH_2O} + \mathrm{SO_4^{2-}} + 2\mathrm{H^+} \rightarrow 2\mathrm{CO_2} + \mathrm{H_2S} + 2\mathrm{H_2O} \quad (4)$$

$$\mathrm{AMG}: 2\mathrm{CH_2O} \rightarrow \mathrm{CH_4} + \mathrm{CO_2} \quad (5)$$

$$\mathrm{HMG}: \mathrm{CO_2} + 4\mathrm{H_2} \rightarrow \mathrm{CH_4} + 2\mathrm{H_2O} \quad (6)$$

The amount of SO_4^{2-} reduced (for SR; Eq. 1) or CH_4 produced (for MG; Eqs. 2 and 3) via each process was used to determine the total amount of organic C mineralized to CO_2 and CH_4. For SR and AMG, 2 mol C are mineralized per SO_4^{2-} reduced (Eq. 4) or CH_4 produced (Eq. 5), while there is no net C mineralization for HMG (Eq. 6). The rates of CO_2 and CH_4 production and total organic C (TC) mineralization are then:

$$\mathrm{CO_2} = 2 \cdot \mathrm{SR} + \mathrm{AMG} \quad (7)$$

$$\mathrm{CH_4} = \mathrm{AMG} + \mathrm{HMG} \quad (8)$$

$$\mathrm{TC} = 2 \cdot \mathrm{SR} + 2 \cdot \mathrm{AMG} \quad (9)$$

Soil biogeochemistry

Porewater and solid-phase biogeochemistry was determined on the same soil cores used for microbial SR and MG rates on each 2 cm soil section between the surface and 20 cm depth. Two cm^3 of soil was placed into an aluminum weigh dish for determination of bulk density, porosity, and elemental analysis after drying at 90°C. C and N content was determined on dried, ground soil using a Leco TruSpec CN analyzer. Carbonates did not contribute to the C content of these soils [unacidified = 0.997 (acidified) + 0.18; $R^2 = 0.88$; $n = 87$ samples from throughout the experiment and from both treatments] and the CN content reported here is for unacidified samples. For determination of porewater CH_4 concentrations, 2 cm^3 of soil was placed into duplicate 12 mL headspace vials which were immediately sealed. Four mL of 1 N HCl was injected into the vial, and the contents shaken vigorously to stop microbial activity and equilibrate the porewater gases with the vial headspace. CH_4 concentration was determined on the headspace of these vials by gas chromatography.

We placed 50 cm^3 of soil into centrifuge tubes under an N_2 atmosphere, centrifuged the soil at 4000 rpm for 15 min, and split aliquots of porewater into several vials for various analyses. One mL of unfiltered porewater was preserved with 50 µL of a saturated $HgCl_2$ solution for DIC analysis on a Shimadzu TOC-V_{CSH}. One mL of unfiltered porewater was pipetted into a 20% zinc acetate solution for later determination of reduced sulfide concentrations (Cline 1969). Four mL of 0.7 µm nominal filtered (GF/F) porewater was preserved with 50 µL of 6 N HNO_3, 2 mL of filtered sample was immediately frozen, and the remaining sample (1–5 mL) was filtered and refrigerated.

Porewater Cl^- and SO_4^{2-} (Dionex DX 500 ion chromatograph) and PO_4^{3-} (phosphomolybdate method; Murphy and Riley 1962) concentrations were determined on nitric acid acidified samples. Dissolved organic carbon (DOC) concentrations were determined by high-temperature combustion following sparging of acidified samples on a Shimadzu TOC-V_{CSH}. NH_4^+ (phenolhypochlorite method; Solorzano

1969) and NO_x (flow injection autoanalyzer following cadmium reduction) concentrations were measured on un-acidified, refrigerated samples. Acetate was determined on frozen samples by high-pressure liquid chromatography (Agilent 1200 series) following sample derivitization (Albert and Martens 1997).

Data analysis

Porewater and solid phase biogeochemical variables and microbial rates were integrated over a 20 cm depth, with linear interpolations between data points when data were missing (e.g., rates were measured on only 6 of 10 depths). Porewater and solid phase measurements were converted to volumetric units (i.e., mmol cm^{-3}) using measured soil porosity and bulk density, respectively. Statistical analyses of the data were conducted using linear regressions and univariate analysis of variance (ANOVA) with least squares difference corrections of confidence intervals for main effects using SPSS (v16.0). Additional pairwise comparisons of means were made using T tests for independent samples.

Results

Gas flux

Gaseous CO_2 flux rates were significantly higher for cores undergoing salt-water intrusion (Fig. 1, $p < 0.001$, $F_{1,1452} = 95.38$). The CO_2 flux from the salt-water amended marsh soils increased above flux rates from freshwater controls rapidly (<1 week) following salt-water intrusion and remained

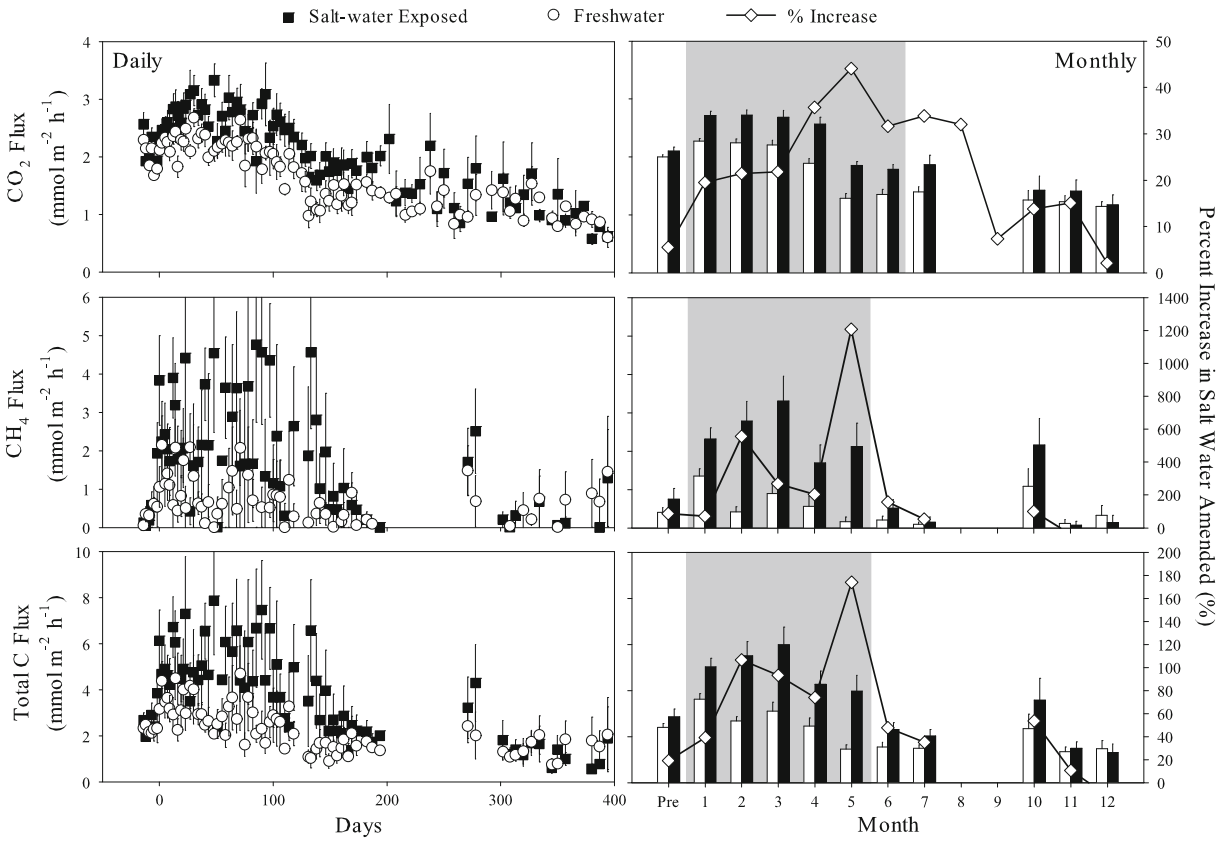

Fig. 1 Daily and monthly carbon dioxide (CO_2; *top*), methane (CH_4; *middle*) and total C (*bottom*) gas fluxes (mmol m^{-2} h^{-1}; mean ± SE) from freshwater soil cores and soil cores exposed to dilute salt-water. The percent increase in flux from salt-water amended soils versus freshwater controls for monthly averages are shown, and shading indicates months for which differences between treatments are significant ($p < 0.05$; months 1–6 for CO_2, 1–5 for CH_4 and 1–5 for total C). No CH_4 (and therefore total C) data are available from day 200 to day 270 due to equipment failure

significantly higher for the first 6 months of the experiment (Fig. 1, $p < 0.05$). Maximum flux rates in both treatments were measured during months 1 through 3 averaging $\sim$2.7 mmol m^{-2} h^{-1} in the salt-water amended cores and $\sim$2.2 mmol m^{-2} h^{-1} in the freshwater cores (Fig. 1). The relative difference in CO_2 flux from salt-water amended soils increased to 20% in the first several months following salt-water intrusion, with a peak of 45% in the 5th month (Fig. 1). There was a significant decline in CO_2 gas flux over time in both the freshwater (CO_2 flux $= -0.0037 \times$ day $+ 2.19, t = -18.02, p < 0.001, R^2 =$ 0.32, $F_{1,696} = 324.61$) and salt-water amended soils (CO_2 flux $= -0.0040 \times$ day $+ 2.61$, $t = -12.61$, $p < 0.001, R^2 = 0.17, F_{1,756} = 159.08$).

CH_4 fluxes (Fig. 1) were significantly higher for cores undergoing salt-water intrusion ($p < 0.001$, $F_{1,617} = 44.04$) and this difference persisted for 5 months ($p < 0.05$, Fig. 1). CH_4 flux from salt-water amended cores peaked in month 3 with an average rate of about 3.3 mmol m^{-2} h^{-1} (Fig. 1). The flux of CH_4 from salt-water amended soils was 70% (in month 1) to 1200% (month 5) higher than flux rates from freshwater soils (Fig. 1). Note that CH_4 flux rates were not significantly different for 2 months (months 6 and 7) prior to data loss during months 8 and 9 (Fig. 1). As was observed for CO_2 flux, the flux of CH_4 declined significantly over time from the freshwater (CH_4 flux $= -0.0021 \times$ day $+ 0.84, t = -3.26, p =$ 0.001, $R^2 = 0.03$, $F_{1,313} = 10.65$) and salt-water amended soils (CH_4 flux $= -0.0050 \times$ day $+ 2.15$, $t = -3.75, p < 0.001, R^2 = 0.04, F_{1,305} = 14.05$).

Overall, total gaseous C fluxes ($CO_2 + CH_4$) were significantly higher from salt-water amended marsh soils for 5 months following salt-water intrusion ($p < 0.001$, $F_1 = 52.46$), and C emissions ranged from 40% (in month 1) to 175% (in month 5) higher from the cores undergoing salt-water exposure than from freshwater cores (Fig. 1). Total C flux from salt-water impacted marsh soils peaked in month 3 at a rate of about 6 mmol C m^{-2} h^{-1} (Fig. 1).

During the first 90 days of the experiment (when inundated flux measurements were conducted) the average DIC flux rates when the soils were flooded were not significantly different than CO_2 gas fluxes when soils were exposed (Table 2; $p = 0.84$, $F_{1,865} = 0.66$). In contrast, there was a significant difference between exposed and inundated CH_4 fluxes (Table 2; $p = 0.02$; $F_{1,379} = 5.79$). The ratio of CH_4 emissions in inundated versus exposed cores ($R_{(Ind/Exp)}$) was 0.46 and 0.22 in freshwater and salt-water amended cores, respectively (Table 2).

Total CO_2 and CH_4 emissions over the 1-year experiment were calculated. As there was no significant difference between inundated and exposed CO_2/DIC flux (Table 2), the measured CO_2 gas fluxes (Fig. 1) were integrated over 1-year for 24 h per day. 14.2 mol CO_2 m^{-2} was emitted from freshwater soils compared with 17.3 mol CO_2 m^{-2} from soils exposed to salt-water. Because of the lower CH_4 emissions when soils were flooded (Table 2), the CH_4 gas flux measurements (Fig. 1) were assumed to represent CH_4 emissions for 12 h per day when soils were exposed. To determine CH_4 emissions for the remaining 12 h per day when soils were flooded, the CH_4 gas flux measurements (Fig. 1) were multiplied by the appropriate $R_{(Ind/Exp)}$ (Table 2). The total CH_4 flux over the 1-year experiment was calculated

Table 2 Average (±standard deviation; SD) carbon dioxide (CO_2) and methane (CH_4) flux rates (mmol m^{-2} h^{-1}) from freshwater and salt-water amended soil cores under exposed and inundated conditions, and the ratio of inundated to exposed flux ($R_{(Ind/Exp)}$) rates during the initial 90 days of the experiment

	CO_2						CH_4					
	Freshwater			Salt-amended			Freshwater			Salt-amended		
	Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*
Exposed	2.17	0.54	396	2.54	0.91	449	0.86	1.37	182	2.12	2.68	177
Inundated	2.36	2.00	14	2.28	1.72	10	0.40	0.36	14	0.46	0.48	10
$R_{(Ind/Exp)}$	1.09			0.90			0.46			0.22		

The number of measurements (*n*) is shown. Note that the difference between exposed and inundated measurements is significantly different for CH_4 ($p = 0.02$; $F_{1,379} = 5.79$) but not for CO_2 ($p = 0.84$, $F_{1,865} = 0.66$)

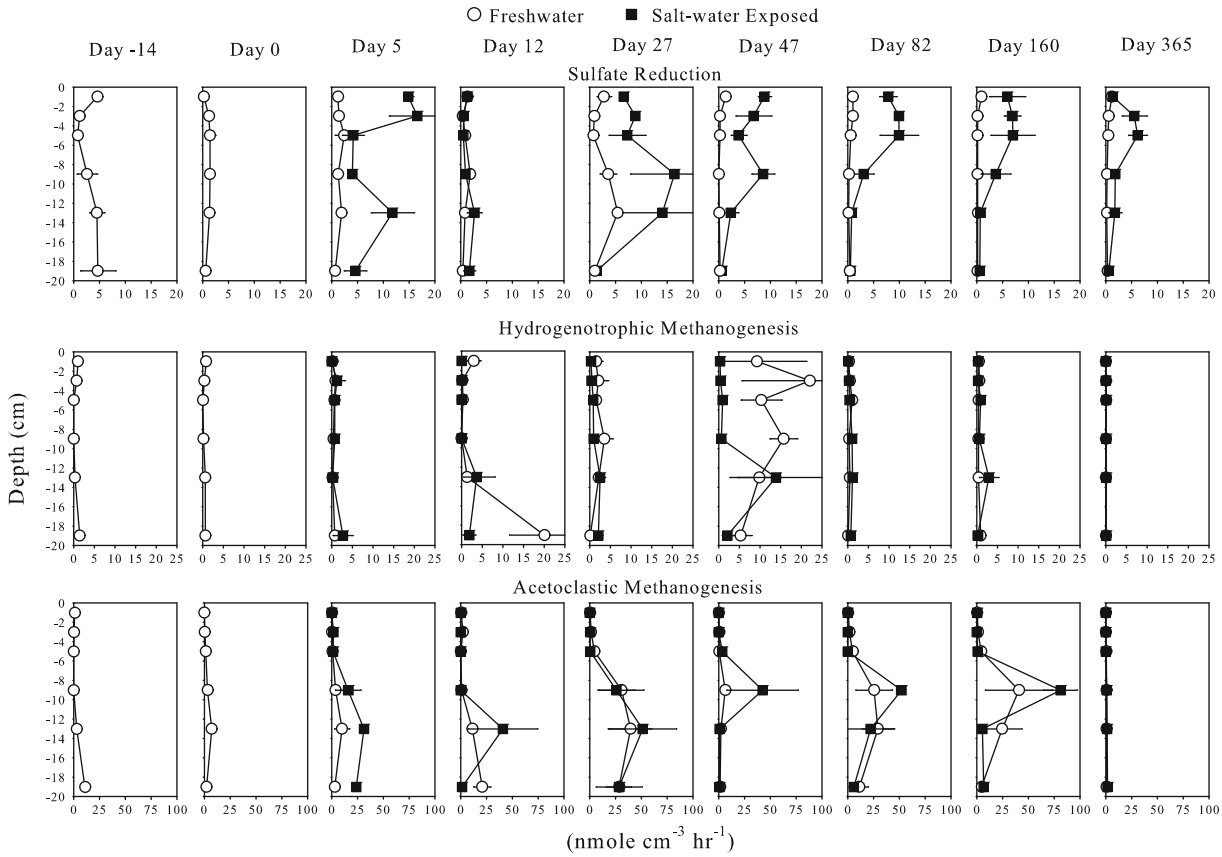

Fig. 2 Depth-specific rates of sulfate reduction (*top*; nmol SO_4^{2-} cm^{-3} h^{-1}), hydrogenotrophic methanogenesis (*middle*; nmol CH_4 cm^{-3} h^{-1}) and acetoclastic methanogenesis (*bottom*; nmol CH_4 cm^{-3} h^{-1}) in soils undergoing salt-water intrusion and in freshwater controls over time (average ± SE)

to be 3.9 and 7.5 mol m^{-2} from freshwater and salt-water amended cores, respectively. The total C gas flux over the 1-year experiment from freshwater cores was 18.2 mol m^{-2}, compared with 24.9 mol m^{-2} from salt-water amended soils.

Rates of microbial sulfate reduction and methanogenesis

Rates of SR ranged from 0 to approximately 16 nmol SO_4^{2-} cm^{-3} h^{-1}. SR was lower in the freshwater soils than in salt-water amended soils throughout the experiment. In the salt-water amended cores, SR rates increased at all depths on day 5, were not significantly different than freshwater rates on day 12, and were higher in the upper 10 cm of the soil column for the duration of the experiment (Fig. 2). SR became more confined to surface (0–5 cm) soils from 3 months until the termination of the experiment.

Depth-integrated rates of SR remained under 0.7 mmol SO_4^{2-} m^{-2} h^{-1} in freshwater soils and reached a maximum of 2.0 mmol SO_4^{2-} m^{-2} h^{-1} in salt-water impacted soils on day 27 after salt-water intrusion (Fig. 3). Salt-water amendment had a significant effect on SR rates ($p < 0.001$, $F_{1,29} = 25.40$). SR rates were significantly higher in salt-water impacted soils on all dates ($p < 0.05$, $t = 3.01$, $df = 2$) except for day 47 (Fig. 3). Total SR integrated over the 1-year experiment was 0.9 mol SO_4^{2-} m^{-2} in freshwater soils and 6.8 mol SO_4^{2-} m^{-2} in the salt-water amended soils.

Rates of HMG ranged from 0 to 22 nmol CH_4 cm^{-3} h^{-1}. HMG rates were variable and there was no clear pattern with depth (Fig. 2). Depth-integrated rates of HMG peaked in both freshwater and salt-water impacted soils on day 47, with the highest rates

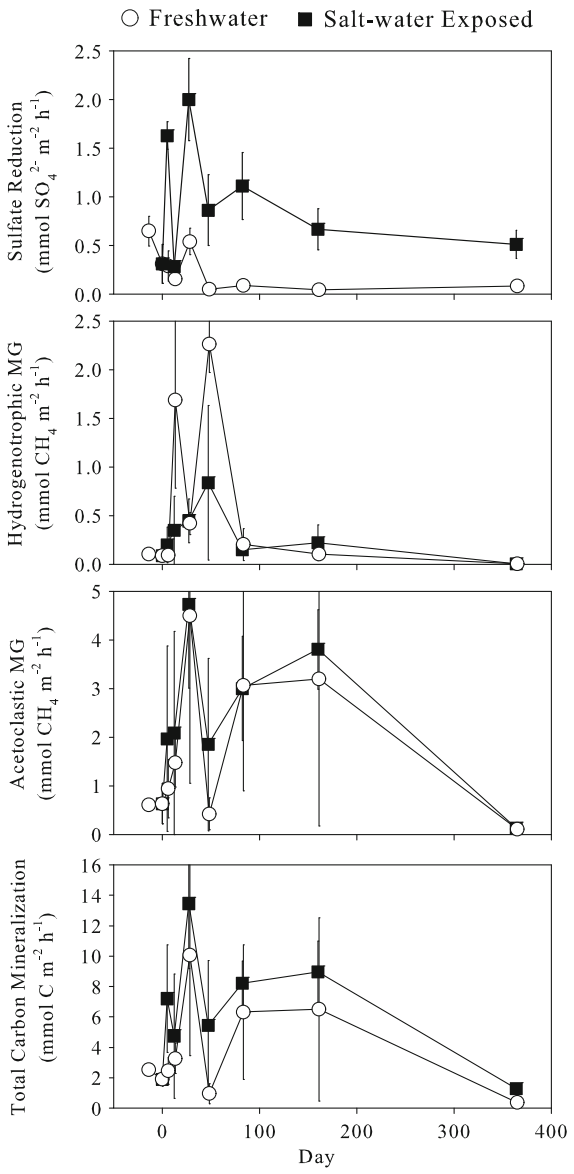

Fig. 3 Depth-integrated rates (mmol m^{-2} h^{-1}; average ± SE) of sulfate reduction, hydrogenotrophic and acetoclastic methanogenesis and total carbon mineralization (see text) over time in soil cores undergoing salinity intrusion and in freshwater controls

measured in freshwater soils (2.3 mmol CH_4 m^{-2} h^{-1}; Fig. 3). There were no significant differences in HMG rates between treatments ($p = 0.43$, $F_{1,29} = 0.63$), although note the high rates in freshwater soils on days 12 and 47 (Fig. 3). Rates of HMG integrated over 1-year were 2.8 mol CH_4 m^{-2} in the freshwater soils and 1.8 mol CH_4 m^{-2} in soils exposed to salt-water.

AMG rates of up to 80 nmol CH_4 cm^{-3} h^{-1} were measured. AMG was generally low in surface soils, and maximum rates were usually observed at deeper depths (>8 cm; Fig. 2). Depth integrated rates of AMG of over 4.0 mmol CH_4 m^{-2} h^{-1} were measured in both freshwater and salt-water impacted soils on day 27 (Fig. 3). There was no significant effect of salt-water amendment on AMG rates ($p = 0.25$, $F_{1,29} = 1.37$). Integrated over the 1-year experiment, rates of AMG were 18.1 mol CH_4 m^{-2} in freshwater and 21.4 mol CH_4 m^{-2} in salt-water amended soils.

Estimates of total C mineralized via anaerobic microbial pathways, calculated from measurements of SR and MG together with reaction stoichiometries in Eqs. 7–9, ranged from 0.4 mmol C m^{-2} h^{-1} (in freshwater soils at day 364) to 13.5 mmol C m^{-2} h^{-1} (in salt-water impacted soils on day 27; Fig. 3). Salt-water amendment significantly affected overall rates of TC ($p = 0.048$, $F_{1,29} = 4.26$), although differences were not significant between specific sampling dates ($p > 0.05$) except on day 364 ($p < 0.05$, $t = 3.27$, $df = 2$; Fig. 3). Rates of total C mineralization integrated over the 1-year experiment were 37.8 mol C m^{-2} (5% SR and 95% AMG) in freshwater soils and 56.2 C mol m^{-2} (24.0% SR and 76% AMG) in soils exposed to salt-water.

Soil biogeochemistry

Soil porosity (0.694 ml cm^{-3} ± 0.003, mean ± SE) and dry bulk density (0.500 g cm^{-3} ± 0.004) varied little with depth, time, or between salt-water and freshwater treatments (data not shown). Porewater Cl^- concentrations remained low in the freshwater soils throughout the experiment (Fig. 4). In the soils undergoing experimental salt-water intrusion, Cl^- concentrations in surface soils increased rapidly to reflect Cl^- concentrations in the ASW (Table 1), while concentrations at depth remained lower throughout most of the experiment. Total inventories of Cl^- in salt-water amended cores increased throughout the experiment (Fig. 4), reflecting the relatively slow diffusion-driven increase of Cl^- at depth. It took almost 3 months before Cl^- in the salt-water amended soils at depth (>16 cm) became significantly higher than Cl^- concentrations in the freshwater control soils, and a full year before inventories of Cl^- in amended cores were fully equilibrated with Cl^- concentrations in the overlying water (Fig. 4). There was an overall

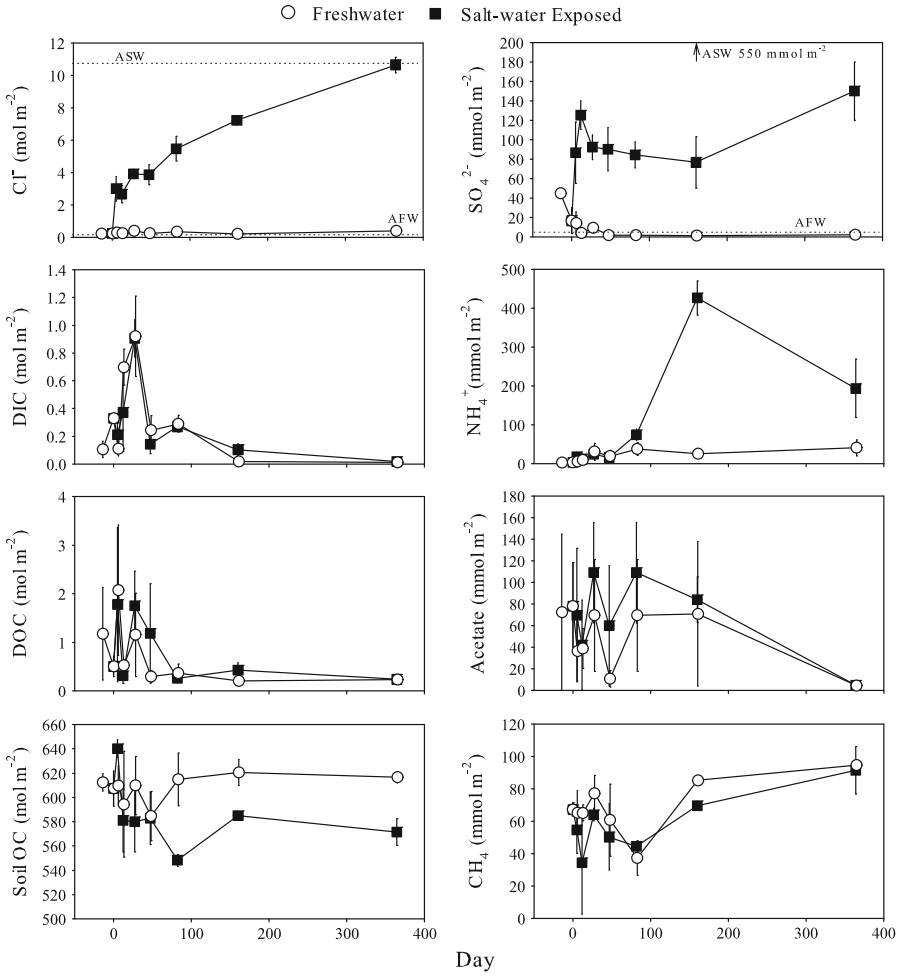

Fig. 4 Depth-integrated inventories (integrated to 20 cm; mean ± SE) of porewater chloride (Cl^-), sulfate (SO_4^{2-}), dissolved inorganic carbon (DIC), ammonium (NH_4^+), dissolved organic carbon (DOC), acetate and methane (CH_4), and inventories of soil organic C in freshwater and salt-water amended cores over time. Horizontal lines on select graphs denote theoretical inventories of cores fully equilibrated with overlying artificial freshwater (AFW) and/or seawater (ASW) used in the experiment (Table 1)

significant difference between Cl^- inventories in salt-water amended and freshwater soils ($p < 0.001$, $F_{1,29} = 88.70$), and Cl^- inventories were significantly greater in salt-water amended cores on all sampling dates post-amendment ($p < 0.05$, $t > 3.51$, $df = 2$).

Initial porewater SO_4^{2-} concentrations in cores collected from the TFM (day 14) indicated a sub-surface SO_4^{2-} maximum of about 700 µmol L^{-1} at a depth of 7 cm (data not shown). This mid-depth peak in SO_4^{2-} concentrations in the freshwater cores decreased during the first several weeks of the experiment, such that by day 12 porewater SO_4^{2-} in freshwater cores did not exceed 100 µmol L^{-1} and this decrease is reflected in the SO_4^{2-} inventories (Fig. 4). SO_4^{2-} concentrations in salt-water amended cores increased rapidly on days 5 and 12, and then declined slightly through day 160 before increasing again at the end of the experiment (Fig. 4). SO_4^{2-} inventories remained far below equilibration with the overlying ASW (550 mmol m^{-2}) throughout the experiment (Fig. 4). SO_4^{2-} was limited to surface soils, and concentrations at depths below 10 cm remained low relative to overlying water concentrations (<500 µmol L^{-1}; see Table 1). There was a significant treatment effect on SO_4^{2-} inventories ($p < 0.001$, $F_{1,29} = 93.39$), and inventories of SO_4^{2-} were significantly greater in salt-water amended cores on days 12, 27, 82 and 364 ($p < 0.05$, $t > 3.99$, $df = 2$).

Porewater NH_4^+ concentrations were low initially (<150 µmol L^{-1}) and remained below 500 µmol L^{-1} in freshwater cores throughout the experiment (Fig. 4). Salt-water amendment impacted NH_4^+ inventories significantly ($p = 0.02$, $F_{1,29} = 6.01$), although NH_4^+ concentrations were not significantly different between freshwater and salt-water amended soils

except on day 160 when NH_4^+ inventories in salt-water amended soils peaked at over 400 mmol m^{-2} (Fig. 4, $p < 0.05$, $t = 9.06$, $df = 2$). DIC concentrations were variable between replicate cores, and there were no significant differences between treatments for whole core inventories ($p = 0.90$, $F_{1,29} = 0.02$, Fig. 4). There was a consistent pattern over time for both treatments, in which inventories increased in both freshwater and salt-water amended soils until day 27 and then decreased. Porewater DIC inventories were quite low by the termination of the experiment (Fig. 4).

Porewater acetate concentrations were consistently higher at depth than in surface soils. Acetate concentrations in the top 4 cm remained below 400 μmol L^{-1}, while maximum concentrations exceeded 1 mmol L^{-1} at depths below 8 cm. Acetate concentrations were variable and there were no significant differences between treatments for whole core inventories ($p = 0.27$, $F_{1,29} = 1.27$), although acetate inventories were consistently larger in salt-water amended soils (Fig. 4). Inventories of porewater DOC were highly variable and there were no significant differences between salt-water amended and freshwater control soils (Fig. 4, $p = 0.57$, $F_{1,29} = 0.34$). DOC concentrations were consistently low by day 82, however, and remained low for the duration of the experiment (Fig. 4).

Whole core CH_4 inventories were not significantly different between freshwater and salt-water amended soils (Fig. 4, $p = 0.10$, $F_{1,29} = 2.96$), although inventories in both treatments increased over time ($p = 0.002$, $F_{1,29} = 12.18$). Soil inventories of PO_4^{3-} and NO_x were consistently low (<10 mmol m^{-2}), were not significantly different between treatments ($p > 0.05$) and did not exhibit patterns over time (data not shown). Porewater sulfide concentrations were below detection (~ 1 μmol L^{-1}) in all cores at all depths (data not shown).

Soil solid phase organic C ranged between 5.0 and 9.5% by weight, and total N ranged from 0.3 and 0.8% by weight. There was no significant change in N over time or between treatments ($p > 0.05$, data not shown). Inventories of organic C were significantly different between salt-water amended and freshwater soils ($p = 0.043$, $F_{1,29} = 4.50$), and organic C was significantly lower in salt-water amended soils on days 82, 160 and 364 compared to freshwater soils and to initial organic C values (Fig. 4, $p < 0.05$, $t < -2.98$, $df = 2$). The average soil inventory of organic C on these 3 sampling dates ($n = 6$ cores per treatment) was 568.2 (±7.7 SE) mol m^{-2} for salt-water amended cores versus 617.3 (± 7.5 SE) mol m^{-2} for freshwater cores and 612.2 (±8.3 SE) mol m^{-2} for the initial organic C inventory on day 14 (Fig. 4).

Discussion

Our research has documented that salt-water intrusion into TFM soils can significantly increase rates of microbial C mineralization (Fig. 5). We used three independent approaches to assess C mineralization, including (1) changes in soil organic C, (2) microbial sulfate reduction and methanogenesis rate measurements, and (3) C gas fluxes from soils following simulated salt-water intrusion and in freshwater controls. The lack of measurable decrease in the organic C in freshwater controls (Figs. 4, 5), coupled with the differences between rates of CO_2 and CH_4 production by microbial mineralization and the flux of these gases from the soils (Fig. 5), suggests these three measures of soil C dynamics were prone to some uncertainties. That these three independent approaches agree on the relative impact of salt-water intrusion on microbial C cycling in TFM soils, however, clearly indicates that the mineralization of organic C accelerates following salt-water intrusion into tidal freshwater marshes.

The total amount of CO_2 and CH_4 released from salt-water amended cores (24.9 mol m^{-2}) was 36.9% greater than the total inorganic C flux from freshwater cores (18.2 mol m^{-2}) over the 1-year experiment. Similarly, the amount of organic matter mineralized via SR and MG within soils experiencing salt-water intrusion (56.2 mol m^{-2}) was greater than mineralization in freshwater soils (37.8 mol m^{-2}) by 49%. Finally, the higher rates of organic matter decomposition in salt-water amended soils were reflected in a loss of soil organic C (44.1 mol m^{-2}) from these soils (Fig. 5). These results reinforce earlier work about the effects of salinity on C turnover in a short-term (30 d) experiment with freshwater riverine sediments (Weston et al. 2006) and in a year-long root decomposition study along an estuarine salinity gradient (Craft 2007). However, unlike these earlier studies, our research has shown that salt-water intrusion can accelerate rates of CH_4 emission to

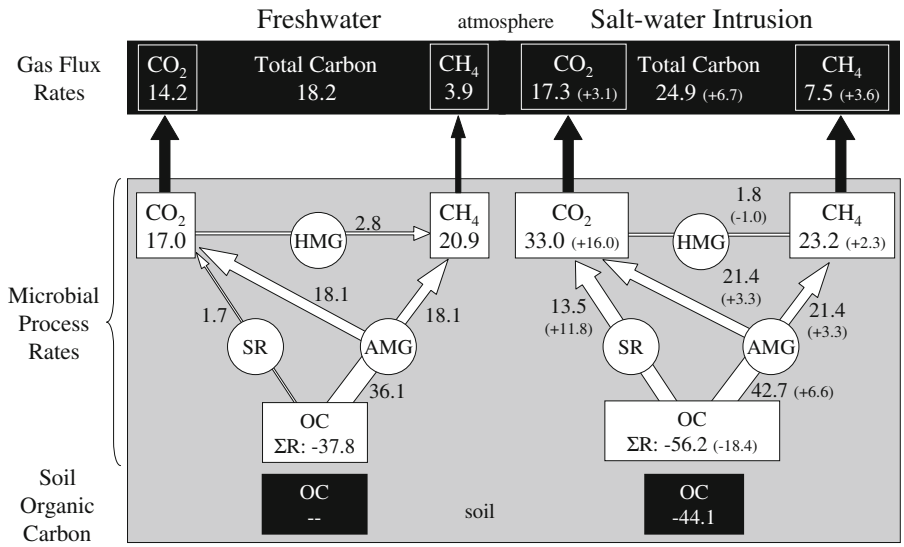

Fig. 5 Schematic of C cycling in freshwater tidal marsh soils (*left*) and soils undergoing salt-water intrusion (*right*) as calculated from measured gas fluxes (*top*, in black), microbial process rates (*middle*, in white) and soil organic C measurements (*bottom*, in black) from cores incubated for 1-year. All values are in units of mol C m^{-2}. Values for gas flux rates (Fig. 1) and microbial process rates (Fig. 3) are integrated over 1-year; organic C content (Fig. 4) is the difference between the initial organic C inventory (612.2 mol m^{-2}) and average soil organic C inventories from days 82, 160 and 364 in salt-water amended (568.2 mol m^{-2}) and freshwater soils (617.3 mol m^{-2}). The net production of carbon dioxide (CO_2) and methane (CH_4), the net consumption of organic carbon (OC) calculated as the sum of microbial process rates or the loss of soil organic carbon, and the rates of sulfate reduction (SR), acetoclastic methanogenesis (AMG) and hydrogenotrophic methanogenesis (HMG) are shown. Increases (+) or decreases (−) in salt-water amended soils relative to freshwater controls are shown in parentheses

the atmosphere, a finding that has implications not only for local rates of C preservation and marsh accretion, but also for regional-scale greenhouse gas budgets.

Experimental design considerations

The overall responses of TFMs to rising sea levels and salt-water intrusion will be determined by changes in microbial dynamics as well as plant processes. Our experimental design, in which soil cores were collected prior to spring plant emergence and incubated in the dark, intentionally precluded new C inputs to the soils via primary production so that we could focus our attention on understanding the effects of salt-water intrusion on microbially-mediated soil C mineralization. We acknowledge that wetland plants can influence C cycling by increasing soil C concentrations (Hines et al. 1989), accelerating rates and modifying pathways of anaerobic metabolism (Neubauer et al. 2005b), and "priming" the microbial utilization of recalcitrant soil C (Wolf et al. 2007). Further, rates of plant production and community composition can themselves be affected by salt-water intrusion (Spalding and Hester 2007). We suggest that the overall effects of excluding plants in our experimental design were to (1) lower total rates of organic C remineralization relative to a vegetated marsh and (2) cause organic matter limitation, leading to a progressive decline in CO_2 and CH_4 production and emission rates over the course of the experiment (Figs. 1, 3). Shifts in hydrology and drainage due to long-term incubation of soils in the laboratory, and the step-increase in salinity when simulating salt-water intrusion rather than pulses of saline water as would accompany salt-water intrusion in the field, also likely alter the overall rates of microbial processes in these soils. Regardless, we do not expect that the chosen experimental design would cause any difference in the relative patterns of soil C mineralization that were observed between freshwater and salt-exposed cores.

Salt-water effects on anaerobic C mineralization rates

Salt-water inundation of TFM soils resulted in shifts in microbial pathways and increases in the overall rate of organic matter decomposition. Higher concentrations of SO_4^{2-} following salt-water intrusion fueled increased rates of SR (Figs. 2, 3) and likely contributed to higher CO_2 emissions from the cores (Fig. 1). Surprisingly, rates of MG did not decrease with salt-water intrusion (Figs. 2, 3) and CH_4 emissions from the salt-exposed cores increased by 70–1200% for 5 months relative to freshwater cores (Fig. 1). The overall gaseous C loss (Fig. 1) was significantly greater in TFM soils following salt-water intrusion relative to freshwater controls. Similarly, the inventory of organic C was significantly lower in salt-water amended soils 3, 6 and 12 months after exposure than in freshwater soils (Fig. 4), reflecting the increased mineralization of organic matter in these soils under higher salinity regimes. Weston et al. (2006) found a similar increase in organic matter decomposition in freshwater sediments following salt-water intrusion in a short-term experiment. In addition, Craft (2007) documented a negative relationship between both soil organic content and accumulation and the salinity of the overlying water in a survey of tidal freshwater and salt marshes, which he attributed to the availability of SO_4^{2-} and thus higher rates of SR in the more saline sites. Our results support these findings, and suggest that salt-water intrusion will stimulate decomposition in TFM soils.

SR and MG are terminal steps in the breakdown of organic matter, and are limited to relatively small organic compounds such as acetate (Weiss et al. 1991). These terminal metabolic processes therefore depend on the generation of low molecular weight DOC substrates by other processes. A microbial consortium converts particulate organic matter into low molecular weight DOC through hydrolysis and fermentation reactions (Arnosti et al. 1994; Fenchel and Findlay 1995). Greater inorganic C fluxes from TFM soils amended with salt-water (Fig. 1), which can be attributed to increased rates of SR and MG (Fig. 3), require either; (1) the utilization of a previously unused pool of low molecular weight DOC in the soils or (2) an increased supply of low molecular weight DOC via hydrolysis and fermentation.

Low molecular weight dissolved organic matter can adsorb onto mineral particles, and ion exchange plays an important role in the sorption of some compounds such as amino acids (Wang and Lee 1993; Liu and Lee 2007). Therefore, the intrusion of saline water with greater concentrations of dissolved ions into previously freshwater soils may desorb organic compounds from exchange sites making them available for terminal metabolism (e.g., Liu and Lee 2007) and may alter the availability of larger dissolved and particulate organic C, perhaps promoting hydrolytic and fermentative production of labile, low molecular weight, dissolved organic compounds. Similarly, NH_4^+ is a surface reactive ion that can also be desorbed upon addition of other cations (Rosenfeld 1979); evidence of NH_4^+ desorption in our study is reflected by the increase in porewater inventories of NH_4^+ in the salt-water amended soils from about 3 months until the termination of the experiment (Fig. 4). The increasing ionic strength of the porewater in the salt-water amended soils therefore clearly altered the soil sorption dynamics. There was no evidence of DOC or acetate desorption, however (Fig. 4), and other potential substrates for these terminal metabolic processes were not measured. Further investigation of the mechanisms influencing organic matter availability upon salt-water intrusion is required.

The amount of both CO_2 and CH_4 produced by the measured microbial processes in marsh soils in both freshwater and salt-water amended soils exceeded the flux of these gases from the soils (Fig. 5). While processes other than SR and MG, such as iron reduction and denitrification (Roden and Wetzel 1996; Neubauer et al. 2005b; Gribsholt et al. 2005), may have contributed to the mineralization of organic matter, the measured rates of SR and MG were more than enough to support the measured inorganic C fluxes from these soils.) There was an increase in CH_4 production in soils following salt-water intrusion (Fig. 5; 2.3 mol CH_4 m^{-2}), which was lower than the increase in measured CH_4 flux (Fig. 5; 3.6 mol CH_4 m^{-2}). The difference between CH_4 production and flux ($\Delta = 17.0$ and 15.7 mol CH_4 m^{-2} for freshwater and salt-water amended soils, respectively; Fig. 5) suggests a difference between production and flux from the soils and/or errors in the rate measurements. Porewater acetate concentrations measured in both freshwater and salt-water amended soils were

relatively high (exceeding 1 mmol L^{-1} in some cases; data not shown), which may reflect increased acetate following centrifugation of soils (Shaw and McIntosh 1990; Hines et al. 1994). Artificially increased concentrations of porewater acetate would result in higher rates of AMG (Eq. 3), and lead to elevated estimates of CH_4 and CO_2 production (Fig. 5). CH_4 oxidation may also have played an important role in mitigating CH_4 emission from these TFM soils (Megonigal and Schlesinger 2002). The oxidation of CH_4 produces CO_2, but as with CH_4, the measured CO_2 gas fluxes could not account for the CO_2 produced via sulfate reduction and methanogenesis ($\Delta = 2.7$ and 15.7 mol CO_2 m^{-2} for freshwater and salt-water amended soils, respectively; Fig. 5). A total of 19.7 and 31.4 mol C m^{-2} was therefore apparently mineralized but not accounted for in gas fluxes from freshwater and salt-water soils, respectively (Fig. 5). The fate of this 'missing' carbon is unclear, though we suspect that estimates of CH_4 and CO_2 production were elevated due to artificially high porewater acetate concentrations. Some amount of the organic carbon substrate used during SR and MG would be assimilated by the microbes mediating these reactions, though growth yields do not typically exceed 10% and are often much lower (Widdel and Bak 1992; Maillacheruvu and Parkin 1996; Reeve et al. 1997; Habicht et al. 2005). Chemoautotrophic fixation and assimilation of CO_2 and CH_4 via methanotrophy, nitrification, reduced sulfur oxidation and other reactions may also reduce fluxes of these gasses from soils (e.g., Howarth 1984; Hadas et al. 2001). Fixation of carbon and an increase in microbial biomass and/or subsequent release of fixed C as DOC (DOC fluxes were not measured in this study, though soil inventories of DOC were substantial: Fig. 4) may account for some of this missing C. Alternatively, ebullition, which can be patchy both in space and time, could be responsible for some loss of CO_2 and CH_4 from soil that was not captured by the exposed or inundated core incubations. For example, ebullition accounted for $\sim$50% of the total CH_4 flux (diffusion + ebullition) from subtidal freshwater river sediments (Chanton et al. 1989).

Pathways of anaerobic C mineralization

The energy yield of SR is greater than that of MG, and when SO_4^{2-} is available, sulfate reducers are expected to outcompete methanogens for organic matter substrates (Capone and Kiene 1988; Mishra et al. 2003). Increased rates of SR upon salt-water intrusion were therefore expected, and these findings support previous studies. For instance, Weston et al. (2006) found that the sulfate reducing microbial community in freshwater sediments of the Altamaha River, GA was able to adjust rapidly (<2 weeks) to higher SO_4^{2-} availability since sulfate reducers can multiply quickly upon the onset of positive growth conditions (e.g., Raskin et al. 1996). Although rates of SR increased in the current experiment, as hypothesized, the apparent stimulation (or at a minimum the lack of suppression) of methanogens was unexpected.

Depth-integrated rates of HMG in general were less than rates of AMG (Fig. 3). Although the differences were not significant, rates of HMG tended to be greater in freshwater soils than in salt-exposed soils, while rates of AMG tended to be higher in salt-water-impacted soils (Fig. 3). The two pathways of MG measured here are usually the major pathways of CH_4 production, but the utilization of other low molecular weight organic substrates, such as methanol and methyl amines, were not directly measured and could therefore account for a portion of the CH_4 generation (Oremland and Polcin 1982). Regardless of the specific substrate, however, results indicate that the increased CH_4 flux from TFM soils experiencing salt-water intrusion was due to the response of the methanogens utilizing organic matter substrates rather than hydrogen as the reductant.

MG was largely limited to deeper soils (>8 cm) while rates of SR were generally greater in surface soils (Fig. 2). SO_4^{2-} concentrations below about 10 cm remained relatively low in the salt-water amended cores, due to consumption of SO_4^{2-} via SR in surface soils and the slow diffusion of SO_4^{2-} at depth (see also Cl^- profiles; Fig. 4). The diffusion of Cl^- deeper into the soils relative to SO_4^{2-} may have desorbed organic matter and stimulated AMG at depth below the zone of active SR. However, rates of AMG were highest in the mid-depth soils, and there was substantial overlap in the zones of active SR and MG (Fig. 2). In the salt-water-impacted soils, there was actually a very weak but statistically significant positive relationship between AMG and SR [$AM = 0.91 \times SR$, $R^2 = 0.05$, $p = 0.04$]. Salt-water intrusion therefore stimulated both SR and MG

(Fig. 5), and the apparent mechanism is enhanced availability of SO_4^{2-} (for SR) and organic matter (for both processes). While SR and MG often compete for substrates, contemporaneous SR and MG can occur when noncompetitive substrates are available (such as methanol and methylamines, which are not available to sulfate reducers; Oremland and Polcin 1982), when organic substrates are in abundance (e.g., Yoda et al. 1987) or due to fine scale heterogeneity in the distribution of electron acceptors and electron donors (Højberg et al. 1994). The increased CH_4 flux from soils experiencing salt-water intrusion was unexpected and conflicts with measurements along estuarine salinity gradients (e.g., Bartlett et al. 1987), and with prior experimental results using tidal freshwater river sediments (Weston et al. 2006). Further work is needed to determine the mechanism leading to enhanced CH_4 emissions following salt-water intrusion.

Implications for TFMs

Marsh accretion, which is necessary if marshes are to keep pace with rising sea levels, occurs through the accumulation of both organic matter and mineral sediments (Reed 1995; Morris et al. 2002). Across a diversity of TFMs, the accumulation of organic matter from both autochthonous and allochthonous sources contributes an average of 62% to vertical marsh growth (Neubauer 2008). Based on the loss of 44.1 mol soil C m^{-2} over 1-year due to salt-water intrusion (Fig. 5), we estimate that the increased rate of decomposition will lead to the loss of 5.8 mm of marsh elevation (assuming the % organic matter is twice the % organic C and a volumetric leverage of 5.5 cm^3 g^{-1} for organic matter in TFM soils; Neubauer 2008). For Delaware River TFMs, which have vertical accretion rates averaging 10 mm yr^{-1} (based on ^{137}Cs, ^{210}Pb, and pollen horizons; Orson et al. 1992; Church et al. 2006) and are exposed to a relative sea level rise rate of about 4 mm yr^{-1}, the loss of 5.8 mm of soil elevation is the difference between a site that is accreting considerably faster than sea level is rising and one that is growing at roughly the rate of today's sea level rise. While it is likely that the response of soil C mineralization to salt water intrusion will moderate after long-term exposure (e.g., Fig. 1), decreases in plant production also are likely and may hinder the vertical growth response of TFMs.

The tidal marsh plant community plays a key role in marsh accretion by supplying organic matter and by trapping allochthonous sediments and associated organic matter from tidal waters as water velocity slows due to friction within the plant canopy (Reed 1995; Pasternack and Brush 2001). In TFMs, salt-water intrusion associated with sea-level rise will adversely affect plant productivity (Willis and Hester 2004; Spalding and Hester 2007), and declines in plant production will limit the accretion potential of these marshes. Shifts in the dominant marsh macrophyte (from freshwater to salt-tolerant species) may play an important role in determining the fate of TFMs experiencing salt-water intrusion, and the rate of both sea-level rise and salinity increases relative to plant community shifts will likely determine the resilience of these ecosystems to climate change. Declines in plant productivity, coupled with increased organic matter decomposition rates as described here, create a scenario in which organic matter sequestration is severely limited in TFMs following salt-water intrusion. Future work involving experimental mesocosms, field transplants, or in situ manipulations that expose both TFM soils and plants to elevated salinities will be necessary since the overall response of TFMs to climate change and salt-water intrusion will be a complex interaction of the processes that drive plant production, microbial decomposition, sediment deposition and, ultimately, marsh accretion. Our work highlights that salt-water intrusion will increase microbial decomposition rates in TFM soils, can change the importance of metabolic pathways in unexpected ways (e.g., increases in CH_4 emissions), and may put TFMs at risk of permanent inundation as rates of sea level rise continue to accelerate.

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