



Fate of Soil Organic Carbon During Wetland Loss

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Abstract We carried out a 1.5-year study of the fate of organic carbon during experimental wetland loss using an herbicide at freshwater, brackish and saltwater emergent wetlands. Total carbon stocks in the upper 50 cm of the soil horizon were 15.0 ± 0.5 kg C/m² (222.8 mt CO₂e/ac) at the freshwater site, 11.1 ± 0.9 kg C/m² (164.7 mt CO₂e/ac) at the brackish site, and 8.5 ± 1.4 kg C/m² (125.7 mt CO₂e/ac) at the saltwater site. There were no significant differences detected in decomposition between the treatment and reference plots, which had 39.4 to 41.5 % material remaining by the end of the study at the three sites. The plots treated with herbicide had decreased elevation of −4.24 cm, −1.56 cm and −1.48 cm at the freshwater, brackish and saltwater sites, respectively, which equate to a mass loss of soil organic carbon of 1273 g C/m², 389 g C/m² and 207 g C/m², respectively. Results indicate statistically greater greenhouse gasses were emitted at the brackish and saltwater plots treated with herbicide compared to the reference plots, with up to 4.2 mt CO₂e/ac emitted at the brackish site and 3.1 mt CO₂e/ac emitted at the saltwater site during the first 1.5 years of the study.

Keywords Carbon sequestration · Wetland loss · Greenhouse gas emissions · Blue carbon

Introduction

Although wetlands comprise only about 5–8 % of the terrestrial land surface (Mitsch and Gosselink 2015), wetland ecosystems are known to sequester 20–30 % of the Earth's soil pool of ~2500 Pg of carbon (Roulet 2000; Bridgman et al. 2006; Lal and Pimentel 2008). The high productivity of wetland systems coupled with slow organic matter (OM) decomposition rates due to anaerobic conditions in flooded soils make them an important sink for atmospheric CO₂ (Davidson and Janssens 2006; DeLaune and White 2011; Mcleod et al. 2011). Unfortunately, many wetlands globally are converting to open water due to anthropogenic and natural causes (Tessler et al. 2015), and the coastal wetlands of the Mississippi River delta are currently being lost at a rate of over 30 km²/yr (Barras et al. 2008). Overall, the amount of carbon sequestered is highly dependent on wetland health and productivity, as large amounts of previously stored carbon could potentially be released to the atmosphere when wetland vegetation dies (Davidson and Janssens 2006; DeLaune and White 2011; Mcleod et al. 2011; Pendleton et al. 2012).

Global warming has become a major worldwide concern that has facilitated significant growth in emissions trading programs collectively referred to as carbon markets. Projects that reduce greenhouse gas emissions generate 'carbon offsets'. A carbon offset (mt CO₂e), also referred to as a carbon credit, is a metric ton reduction in emissions of carbon dioxide or greenhouse gases made in order to compensate for, or to offset, an emission made elsewhere (Murray et al. 2011). For a variety of financial, environmental, and political reasons, substantial interest exists for carbon offsets derived from

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terrestrial landscapes including wetland ecosystems. The carbon sequestered in vegetated coastal ecosystems, specifically mangrove forests, seagrass beds, and salt marshes, has been termed ‘blue carbon’ (Sifleet et al. 2011; Mcleod et al. 2011). Allowing entities to privately invest in wetland restoration projects to offset greenhouse gas emissions elsewhere holds promise as a new carbon offset sector (Zhu et al. 2010; Mack et al. 2012; Blain et al. 2013; Silvestrum and Crooks 2014).

Carbon finance has potential to generate much needed revenue to support wetland restoration and conservation (Murray et al. 2011; Siikamäki et al. 2012). Generally, the amount of carbon sequestered that can be counted towards carbon offsets depends on the difference between the carbon sequestration rate under business-as-usual practices and the carbon sequestration rate that results from a respective restoration activity, referred to as additionality (IPCC 2006; Murray et al. 2007; Murray et al. 2011). Wetland restoration enhances carbon sequestration and prevents carbon release that would occur under business-as-usual as a wetland converts to open water. Insufficient information exists, however, on the fate and transport of carbon during wetland loss to carry out accurate carbon sequestration accounting that would provide offsets for actions that prevent wetland loss (Mittra et al. 2005; Sifleet et al. 2011; Siikamäki et al. 2012). Critical research is needed to determine the proportions of material that is decomposed in-situ and results in CO₂ and/or CH₄ emissions, versus the proportion that is transported out of the project boundary, or is buried (Mack et al. 2012). Scientifically addressing the fate and transport of carbon during wetland loss could optimize the amount of offsets achieved from a specific restoration project and increase the carbon finance available to support the restoration activity (Murray et al. 2011; Mack et al. 2015).

Deltaic wetlands are unique in that they provide a permanent geologic storage mechanism for carbon due to subsidence, caused by the compaction of deltaic sediments, and eustatic sea level rise, with a combined rate of over 1 cm/yr in the Mississippi River delta (Penland et al. 1990; Tornqvist et al. 2006). This subsidence is compensated for primarily by soil organic matter accumulation that results from largely in situ production by marsh plants, rather than transported into the marsh from other areas (Morris et al. 2016). Belowground carbon accumulation is a balance between belowground production and organic matter decomposition that are in turn dependent on a variety of factors such as nutrient availability, flooding status, elevation, and soil redox (Mitsch and Gosselink 2015).

Conversion of coastal marshes to inland open water is often associated with plant stresses such as saltwater intrusion and soil waterlogging, but the physical processes that initiate wetland deterioration are not clear (DeLaune et al. 1994). Hurricanes, such as Katrina and Rita, have triggered rapid large carbon losses of sequestered soil carbon through the destruction of large areas of marsh (Howes et al. 2010;

DeLaune and White 2011; Morton and Barras 2011). The top 50 cm of the wetland soil horizon generally includes the living root zone, which is most geomorphically unstable, and most susceptible to decomposition and erosion when the vegetation dies. Regardless of the mechanism, when coastal habitats are degraded, the detachment, transport and redistribution of organic matter leads to the breakdown of structural aggregates, and then exposure to anaerobic conditions accentuates methanogenesis and denitrification leading to efflux of CH₄ and N₂O to the atmosphere (Lal and Pimentel 2008; Pendleton et al. 2012; Krauss et al. 2016).

When vegetation death occurs, organic carbon undergoes complex cycling, with fate dependent on specific type and source of organic matter (Reddy and DeLaune 2008); part of the soil organic carbon can be chemically oxidized to either CO₂ or CH₄ during decomposition, and part can be buried, either in situ or exported and buried elsewhere, or lost as DOC and DIC in drainage water (Alewell et al. 2009; Crooks et al. 2011; Pendleton et al. 2012). Labile fractions of organic matter are rapidly decomposed into inorganic constituents, which are either lost to the air as CH₄ and CO₂ or exported to the water column, while recalcitrant fractions are buried in the soil column and decomposed more slowly or are permanently buried (Reddy and DeLaune 2008).

Decomposition of soil and plant detritus is carried out by heterotrophic bacteria that utilize organic matter as a source of energy and release CO₂ and CH₄ as byproducts. For example, Day et al. (1994, 2011) observed the presence of the chemoautotrophic bacterium (*Beggiatoa* sp.) in water draining from dying wetlands, indicating rapid decomposition of roots by anaerobic sulfate-reducing bacteria and flushing of that material (i.e., carbon) out of the area. In separate studies of the same marsh, DeLaune et al. (1994) and Nyman et al. (1995) described the physical collapse and anaerobic oxidation of the peat root structure as the cause of wetland loss rather than surface erosional forces. An additional possibility is that following death, root tissues are decomposed aerobically by filamentous fungi, such as *Clavospora bulbosa* or *Trichocladium achrasporum*, that rapidly decompose soil organic matter in salt marshes by transporting molecular oxygen downward through hyphae (Padgett et al. 1989; Padgett and Celio 1990).

Another greenhouse gas of concern in coastal environments is nitrous oxide (N₂O), which is mainly formed during denitrification where nitrate (NO₃) is transformed anaerobically to dinitrogen gas (N₂), however, incomplete conversion results in release of nitric oxide (NO) and nitrous oxide (Reddy and DeLaune 2008). Nitrate can be formed during nitrification, where ammonia (NH₃) is transformed aerobically to NO₃, or typically introduced directly into a wetland from upland runoff or point source discharges. Nitrous oxide is particularly important in carbon accounting because N₂O has a global warming

potential (GWP) 298 times that of CO₂ (IPCC 2007). Though wetlands have a large effect on the Earth's radiative balance through high N₂O and CH₄ emissions, it is only to the extent that emissions change through time (i.e., during wetland deterioration) that they are a positive or negative radiative forcing and impact climate change (Bridgman et al. 2006).

The objective of this research was to investigate the fate of soil organic matter in coastal wetlands after vegetation death. Wetland loss was simulated in freshwater, brackish, and saltwater emergent wetlands using an herbicide. Greenhouse gas emissions, litterbag decomposition, soil organic carbon content, and mass loss of soil organic carbon were measured over a 1.5-year period. We hypothesized that there would be greater GHG emissions from the denuded plots than from nearby reference plots, and that mass loss of soil organic carbon would be higher at the denuded plots.

Methods

We carried out a 1.5-year study of the fate of soil organic matter during simulated wetland loss after applying herbicide to vegetation in wetland plots. Three areas were selected for study in freshwater, brackish, and saltwater emergent wetlands (Fig. 1). The freshwater site was located along Halpin canal in the Bayou Boeuf Basin (see Lane et al. 2015), and the brackish and saltwater sites were located off of Bayou Dularge in Terrebonne Basin. At each study site, four 2.5 × 2.5 m plots were delineated and boardwalks were constructed in order to minimize disturbance during monitoring efforts. Three of the four plots at each site were treated with RoundUp® in order to simulate wetland vegetation death. This technique has been used in past studies to simulate wetland death (DeLaune et al. 1994; Nyman et al. 1995; Williams 1999; Feagin et al. 2009). Plots were visited approximately every other month and measurements taken at control and treatment plots of greenhouse gas emissions (CO₂, CH₄, and N₂O), litterbag decomposition, porewater TOC and salinity. Cores for soil carbon content and data of soil elevation loss were taken at the end of the experiment.

CRMS Sites – Water Temperature, Salinity and Level

Data from the Coastwide Reference Monitoring System (CRMS) sites near to our study sites were used to characterize water temperature, salinity and water level (<http://www.lacoast.gov/crms>). Water level was calibrated to the marsh level at the study sites by regressing water level measured by the CRMS instrumentation with discrete water level measurements taken at the sites during the course of the study with a

staff gauge. The following are the CRMS sites and distances from the respective study sites: CRMS0206 located 4.5 km southeast from FT1; CRMS0396 located 3.6 km north of FT2; and CRMS0377 located 1.9 km west of FT3.

Porewater Dissolved Organic Carbon and Salinity

Wetland soil interstitial porewater was collected from each study plot and analyzed for dissolved organic carbon (DOC). Sample water was collected using a narrow diameter plastic tube connected to a 50 ml syringe (as in McKee et al. 1988). The rigid plastic tube (3 mm diameter) was perforated by several small holes at the end and was inserted into the soil to a depth of 15 cm. Sixty to 80 ml of water were collected, stored in acid-washed 125 ml glass bottles, and immediately stored on ice. Within 24 h, samples were filtered through pre-rinsed 25 mm 0.7 µm Whatman GF/F glass fiber filters into acid-washed bottles, salinity was measured using a salinity meter (American Marine, Inc., Pinpoint Salinity Monitor), and then frozen until DOC analysis using standard methods (APHA 1985).

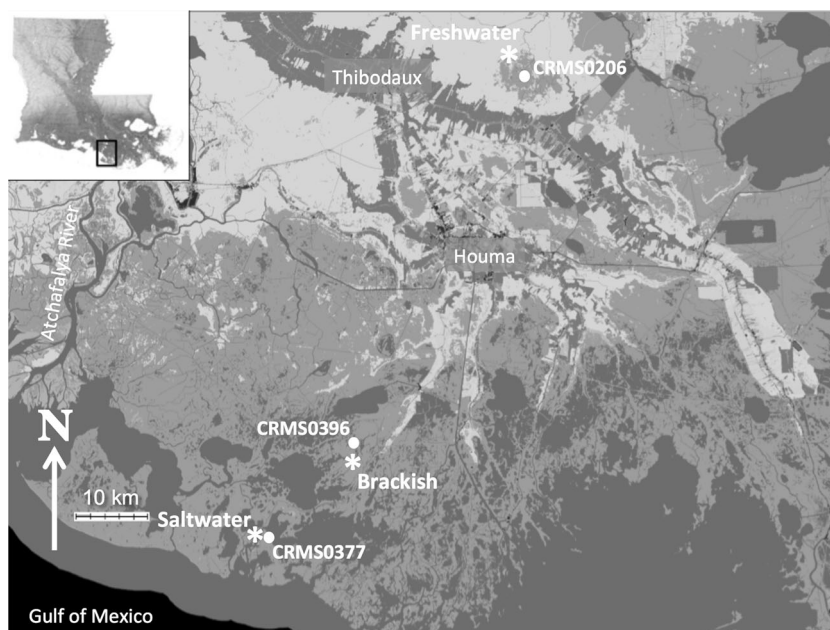
Decomposition

Decomposition rates were measured using the litterbag technique (Blum 1993; Fennessy et al. 2008). Litterbags were made of nylon screening (1 × 2 mm opening) sewn into 10 cm × 30 cm bags, filled with approximately 10 g of vegetation as well as an aluminum identification tag, and sewn closed. Aboveground plant material was collected from the respective study sites, oven dried until constant weight, and distributed into the mesh bags. At each site, half of the bags contained plant materials locally from the site, and the other half contained a mixture of plant materials from all study sites at roughly equal ratios. Bags were inserted into the soil so the plant material was 10–20 cm below the marsh surface. Bags containing local and mixed materials were collected during each site visit. After extraction from the sediment, litterbags were placed in plastic bags, sealed, put on ice, and returned to the laboratory where they were refrigerated until processing (within two weeks of collection). Bags were carefully washed to remove any mineral material. The material in the bags was dried at 60–80 °C to a constant weight (approximately 24 h) and weighed.

Greenhouse Gas Emissions

Carbon dioxide (CO₂), methane (CH₄), and nitrous oxide (N₂O) emissions were measured using the static chamber method (Smith et al. 1983; Klinger et al.

Fig. 1 Location of study sites (indicated by asterisks) and nearby CRMS sites in coastal Louisiana



1994; Livingston and Hutchinson 1995). Gas chambers consisted of an opaque surface chamber consisting of a 5-gal bucket placed on top of a lower sleeve-base positioned approximately 10 cm into the wetland soil if water levels permitted, or positioned on top of a floating Styrofoam ring if water levels were too high to use the bases. Bases were installed approximately one month before first measurements were taken. All samples were taken during daylight hours between 1000 and 1600. A rubber septum in the top of the chamber was used as a sampling port. Gas samples were taken at the beginning, middle and end of deployment, which lasted 1-h at the freshwater site, 1.5-h at the brackish site, and 2-h at the saltwater site. These time intervals were selected based on preliminary results to optimize detection of GHGs. Gas samples were injected into pre-vacuumed 10-cm³ vacutainers and brought back to the laboratory for CH₄, CO₂ and N₂O measurements using a gas chromatograph (e.g., Varian 3800) equipped with a dual Flame Ionization-Thermionic Specific (FID/TCD) system and an electron capture detector (ECD). The gas chromatograph was calibrated for all gases at the time of analysis and standards were used to determine the percent recovery of the sample. Concentrations were converted from volume/volume (given by instrument) to mass/volume concentrations using Eq. (1):

$$\text{Mass/volume concentration (mg/m}^3\text{)} = (C_v \times M \times P)/(R \times T) \quad (1)$$

where C_v is the volume/volume concentration of the gas (in ppm), M is molecular weight of the gas, P is barometric pressure (in atmospheres), T is air temperature (in degrees Kelvin) and R is the universal gas constant (0.0820575 L atm K mole;

Holland et al. 1999). For all samples, barometric pressure was assumed to be 1 atm. The slope of the flux series was used to calculate gas flux using Eq. (2):

$$\text{Gas flux (mg/m}^2\text{/h)} = V \times C_{\text{rate}}/A \quad (2)$$

where V is the internal volume of the air space inside the chamber (in m³), C_{rate} is the change in gas concentration over the enclosure period, or slope of the best-fit line as calculated above (in mg/m³/h), and A is the area of soil covered by the chamber (in m²) (Holland et al. 1999). Mean hourly rates were calculated as averages of the data collected. The total emissions were calculated by extrapolating mean hourly rates to the respective time periods between sampling.

Greenhouse gas emission calculations were made using CO₂ equivalents (CO₂e) based on the 100-year Global Warming Potential (GWP) factors listed in the IPCC Fourth Assessment Report (IPCC 2007). Factors used were 25 for CH₄ and 298 for N₂O. These scaling factors represent the global warming potential for CH₄ and N₂O over a 100-year time horizon. Carbon dioxide was omitted from the calculations from the reference plots since live vegetation was present and thus respiration was occurring, but included in the treatment units where no vegetation was present and thus all emissions were additive.

Soil Bulk Density, Percent Carbon, and Carbon Density

The carbon content of wetland soil profiles were measured at the end of the study using a 9.8 cm diameter thin walled aluminum corer. A core was taken from each of the treatment plots as well as from nearby unimpacted wetlands. Compaction was measured and accounted for in final

calculations. The cores were sectioned in the field into 2.5-cm increments and brought to the laboratory, dried at 55 °C to a constant weight, weighed for bulk density (Brady and Weil 2001), and then analyzed for percent carbon using an elemental analyzer (Chatterjee et al. 2009). Carbon density was calculated by multiplying bulk density by percent carbon.

Elevation Surveys

Elevation surveys were carried out at the beginning and end of the study using standard surveying equipment to measure relative elevation differences of the treatment and control plots (accuracy ± 1.6 mm @ 33 m). Measurements were made at eight points in each plot treated with herbicide as well as at eight points nearby (<2 m) that did not receive herbicide.

Statistical Analysis

All statistical analyses were performed using JMP IN Version 12 produced by SAS Institute, Inc. (Sall et al. 2012). Carbon dioxide, CH₄ and N₂O fluxes were calculated by applying a linear least squares regression to the chamber headspace concentration of each gas plotted against time. Analysis of variance (ANOVA) was carried out to detect differences between means, and *z*-tests were used to compare samples means to hypothesized values (in this case, values from the reference plots). Comparisons of means with significant ANOVA tests (alpha < 0.05) were made using the Tukey–Kramer Honestly Significant Difference (HSD) test (Sall et al. 2012). All analyses were carried out using a *p*-value of 0.05 to determine significance.

Results

CRMS Sites – Water Temperature, Salinity and Level

Mean daily water temperature during 2013 and 2014 ranged from 2.4 °C to 32.9 °C, with lowest temperatures during January and February, and the highest during August and September (Fig. 2). Salinity at the freshwater site never rose above 0.2 PSU, while salinity at the brackish site ranged from 0.9–15.4 PSU with a mean of 5.2 ± 0.12 PSU, and the saltwater site ranged from 4.8–23.3 PSU with a mean of 13.9 ± 0.14 PSU (Fig. 2). Water level was generally highest at the freshwater site (mean: 5.6 ± 0.43 cm), followed by the saltwater site (mean: 1.25 ± 0.27 cm), with the brackish site having the lowest water levels (mean: -16.9 ± 0.73 cm). Percent time inundated followed the same pattern as mean water level, with the freshwater site flooded the most at 67.1 % of the time, followed by the saltwater site at 48.8 %, and the brackish site at 23.8 % (Fig. 2).

Porewater

Mean porewater salinity at the freshwater site was 0.1 PSU at both the treatment and reference plots (Fig. 3). At the brackish site, porewater salinity was significantly lower at the treatment plots (10.6 ± 0.82 PSU) compared to the reference plot (15.4 ± 0.74 PSU; *p* = 0.038), presumably due to a lack of evapotranspiration at the treatment plots. There was no significant difference between the treatment plots (19.3 ± 1.32 PSU) and the reference plot (21.2 ± 2.53 PSU) at the saltwater site.

Porewater DOC concentrations had no discernible trends over time or between treatment and control plots (Fig. 4). However, the pooled mean DOC concentrations were 31.8 ± 2.63 mg/L at the freshwater site, 25.4 ± 1.17 mg/L at brackish site, and 22.1 ± 2.36 mg/L at the saltwater site, with the freshwater and saltwater sites being significantly different from each other (*p* = 0.083), but not from the brackish site.

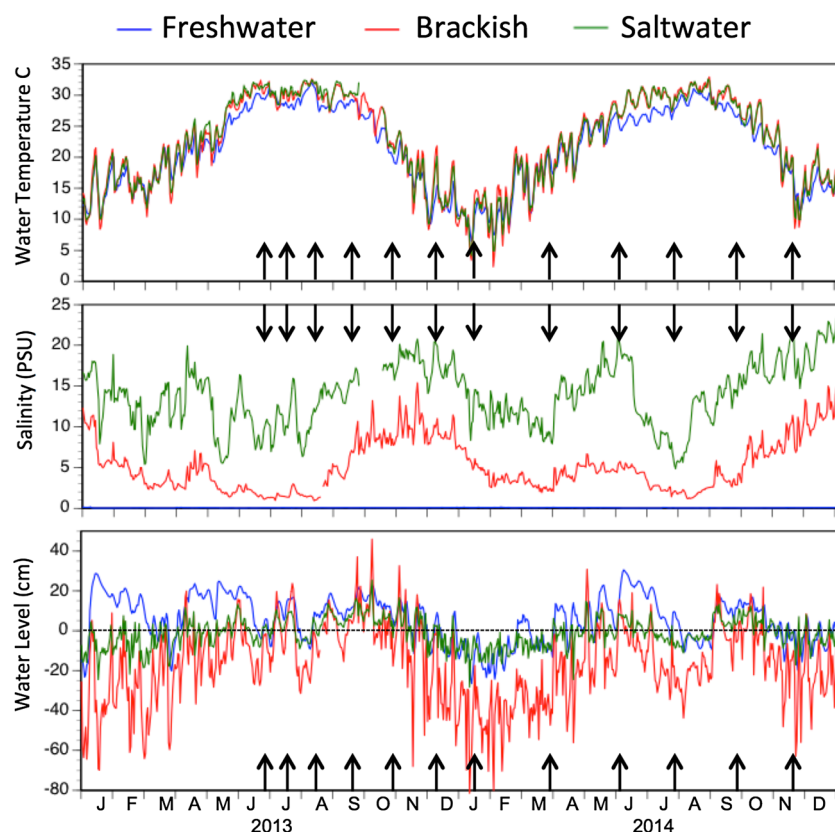
Decomposition

There were no significant differences detected in the rate of decomposition between the treatment and reference plots or between sites (Fig. 5). There was, however, significantly more decomposition of the mixed vegetation (31.7 % remaining) compared to the native vegetation (47.8 % remaining) at the freshwater site (*p* = 0.0022), suggesting freshwater vegetation is more recalcitrant than the brackish and saltwater vegetation mix. The amount of material left at the end of 436 days was remarkably similar between sites, with the means of the three sites ranging from 39.4 to 41.5 % remaining, with an overall pooled mean of 40.3 ± 1.42 % remaining.

Greenhouse Gas Emissions

Carbon dioxide emissions had a seasonal signal of decreased emissions during cooler months (Fig. 6). Mean carbon dioxide emissions at the freshwater treatment and reference sites were 84.3 ± 11.2 mg/m²/h and 103.6 ± 16.0 mg/m²/h, respectively (Table 1). Total CO₂ flux during the course of the experiment was 1003.4 ± 163.8 g/m² at the treatment site and 1291.6 g/m² at the reference site (Table 1). At the brackish site, mean CO₂ emissions were significantly different (*p* = 0.0181) with 116.3 ± 19.1 mg/m²/h and 191.0 ± 38.1 mg/m²/h measured at the treatment and reference sites, respectively. Total CO₂ emissions at the brackish treatment site were 901.0 ± 171.0 g/m² and were significantly lower (*p* = 0.0059) than emissions measured at the reference site of 2457.7 g/m² (Table 1). The saltwater site had mean CO₂ emissions of 75.1 ± 8.3 mg/m²/h and 131.4 ± 63.2 mg/m²/h at the treatment and reference sites, respectively. Total CO₂ emissions at the

Fig. 2 Mean daily water temperature, salinity, and water depth above the wetland surface, indicated as 0, at the three study locations. Arrows indicate sampling times



treatment site were $813.8 \pm 59.2 \text{ g/m}^2$, which was significantly lower ($p = 0.0056$) than the 1366.5 g/m^2 measured at the reference site (Table 1).

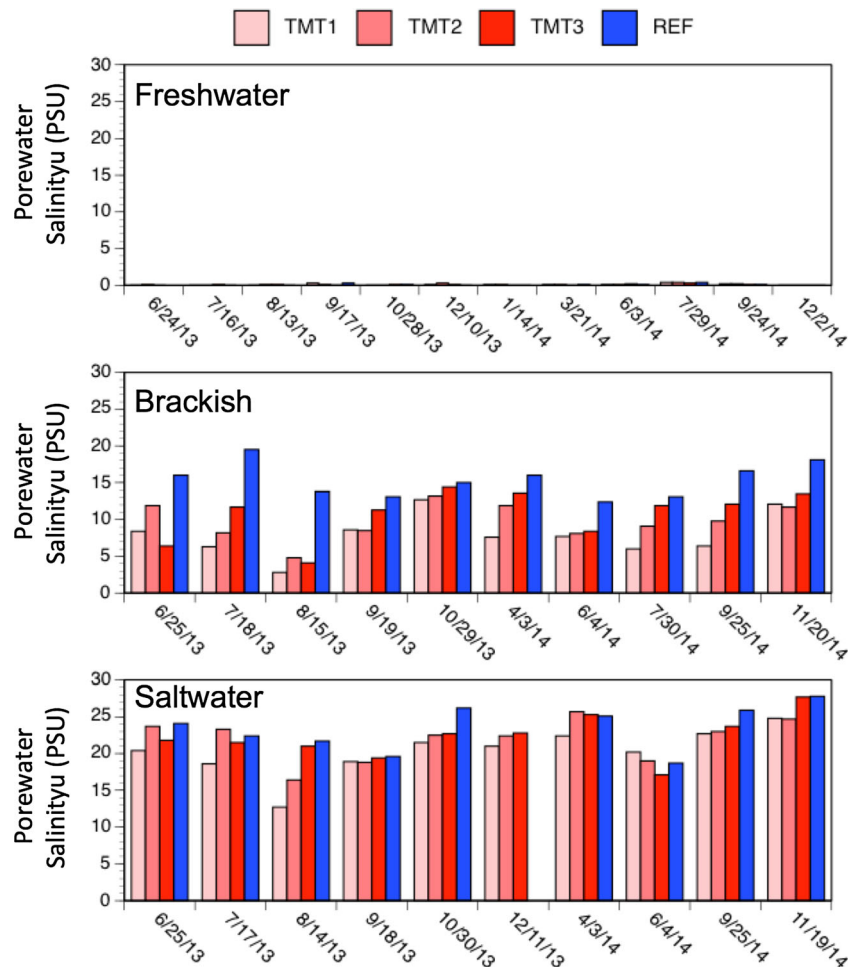
Methane emissions were over an order of magnitude higher at the freshwater site compared to the brackish and saltwater sites (Fig. 6). The freshwater site had mean CH_4 fluxes of $24.1 \pm 3.9 \text{ mg/m}^2/\text{h}$ and $35.9 \pm 10.4 \text{ mg/m}^2/\text{h}$ at the treatment and reference sites, respectively (Table 1). Total CH_4 flux during the course of the experiment was $311.4 \pm 1.9 \text{ g/m}^2$ at the treatment site, which was significantly lower ($p = 0.0003$) than the 421.3 g/m^2 measured at the reference site. Methane emissions at the brackish site averaged $2.3 \pm 0.7 \text{ mg/m}^2/\text{h}$ and $1.3 \pm 0.4 \text{ mg/m}^2/\text{h}$ at the treatment and reference sites, respectively. Total CH_4 emissions were $19.0 \pm 2.5 \text{ g/m}^2$ and 14.0 g/m^2 at the brackish treatment and reference sites, respectively (Table 1). At the saltwater site, mean CH_4 emissions were $0.5 \pm 0.2 \text{ mg/m}^2/\text{h}$ and $0.9 \pm 0.4 \text{ mg/m}^2/\text{h}$ at the treatment and reference sites, respectively. Total CH_4 emissions were $4.1 \pm 1.0 \text{ g/m}^2$ and 7.6 g/m^2 at the treatment and reference sites, respectively (Table 1).

The freshwater site had mean N_2O emissions of $0.0084 \pm 0.0044 \text{ mg/m}^2/\text{h}$ and $0.0067 \pm 0.0039 \text{ mg/m}^2/\text{h}$ at the treatment and reference sites, respectively (Table 1). Total N_2O flux during the course of the experiment was

$0.1244 \pm 0.0385 \text{ g/m}^2$ and 0.1085 g/m^2 at the treatment and reference sites, respectively. At the brackish site, N_2O emissions at the treatment and reference sites had means of $0.0016 \pm 0.0009 \text{ mg/m}^2/\text{h}$ and $0.0013 \pm 0.0009 \text{ mg/m}^2/\text{h}$, respectively. Total N_2O emissions at the brackish treatment and reference sites were $0.0242 \pm 0.0193 \text{ g/m}^2$ and 0.0254 g/m^2 , respectively (Table 1). Nitrous oxide emissions at the treatment and reference sites were $0.0109 \pm 0.0078 \text{ mg/m}^2/\text{h}$ and $0.0025 \pm 0.0021 \text{ mg/m}^2/\text{h}$, respectively. Total N_2O emissions were $0.1679 \pm 0.0932 \text{ g/m}^2$ and 0.0347 g/m^2 at the treatment and reference sites, respectively (Table 1).

Driven by increased methane emissions, which have 25 times the greenhouse warming potential of CO_2 , greenhouse gas emissions were over a magnitude higher and much more highly variable at the freshwater site compared to the brackish and saltwater sites, however, there was not a significant difference between the treatment and reference plots at the freshwater site (Fig. 7). There was a significant difference at both the brackish and saltwater sites, however, where the treatment plots emitted significantly more greenhouse gasses compared to the reference plots, with a net difference of 1026.4 and $766.1 \text{ g CO}_2\text{e/m}^2$, respectively, for the brackish and saltwater sites over the 1.5 years of the study. This is equivalent to $4.2 \text{ mt CO}_2\text{e/acre}$ and $3.1 \text{ mt CO}_2\text{e/acre}$, respectively, for the brackish and saltwater sites (Fig. 7).

Fig. 3 Porewater salinity at the three study locations



Soil Bulk Density, Percent Carbon, and Carbon Density

Bulk density increased with depth at the freshwater site from approximately 0.1 g/cm^3 at the surface to $>0.5 \text{ g/cm}^3$ at $>30 \text{ cm}$ depth, averaging $0.32 \pm 0.03 \text{ g/cm}^3$ (Fig. 8). The brackish site had an average bulk density of $0.26 \pm 0.01 \text{ g/cm}^3$ and relatively uniform bulk density ranging $0.2\text{--}0.3 \text{ g/cm}^3$, with exception of several sections at approximately 20 cm depth that had bulk density of $>0.40 \text{ g/cm}^3$ and were visually distinguished as containing gray clay. Bulk density at the saltwater site was much more variable than the brackish or freshwater sites, ranging from $0.3\text{--}1.0 \text{ g/cm}^3$, with a mean of $0.64 \pm 0.03 \text{ g/cm}^3$. Soil bulk density was not statistically significant different between the treatment and reference plots at any of the three sites (Fig. 8).

Percent carbon decreased with depth at the freshwater site, ranging from 32 % at the surface to 4 % at $>40 \text{ cm}$, with a mean of $16.7 \pm 0.98 \%$ (Fig. 9). The brackish site ranged from 6 to 13 % with a mean of $9.7 \pm 0.42 \%$. Percent carbon at the saltwater site ranged from $>4 \%$ near the surface to $<2 \%$ below 20 cm depth, with a mean of $2.8 \pm 0.21 \%$. There was not a statistically significant difference in percent organic

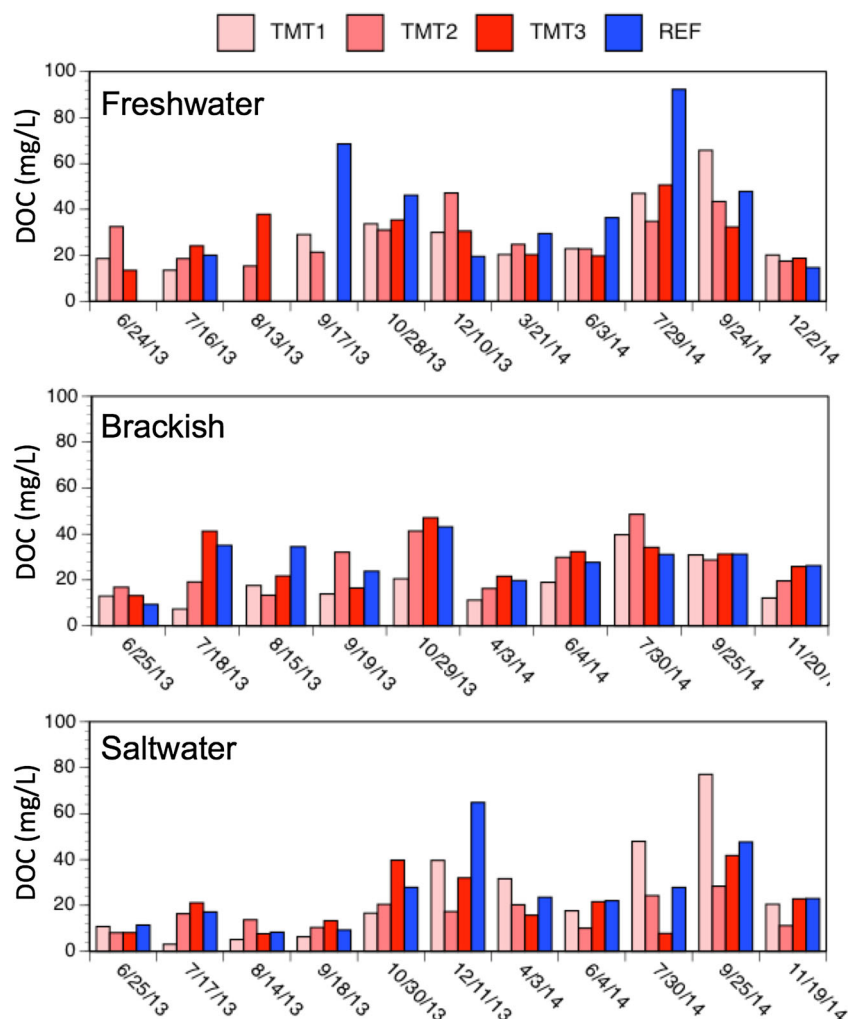
carbon between the treatment and reference plots at any of the three sites (Fig. 9).

Carbon density did not differ statistically between the treatment and reference plots at any of the sites, except for the topmost slices at the saltwater site, which were highly variable but statistically greater at the treatment site ($p = 0.0425$; Fig. 10). The freshwater site had a carbon density that ranged from $0.02\text{--}0.05 \text{ g C/cm}^3$, with a mean of $0.033 \pm 0.001 \text{ g C/cm}^3$, and a total carbon content of $1.50 \pm 0.05 \text{ g C/m}^2$ ($15.0 \pm 0.5 \text{ kg C/m}^2$). The brackish site had a total carbon content of $1.11 \pm 0.09 \text{ g C/cm}^2$ ($11.1 \pm 0.9 \text{ kg C/m}^2$), ranging from $0.01\text{--}0.04 \text{ g C/cm}^3$, with a mean of $0.024 \pm 0.001 \text{ g C/cm}^3$. The total carbon content at the saltwater site was $0.85 \pm 0.14 \text{ g C/cm}^2$ ($8.5 \pm 1.4 \text{ kg C/m}^2$), ranging $0.005\text{--}0.045 \text{ g C/cm}^3$, with a mean of $0.019 \pm 0.002 \text{ g C/cm}^3$.

Elevation Surveys

The elevation surveys found decreases in elevation at all the treatment plots compared to the surrounding wetland elevation (Table 2). The freshwater site had an average of $-4.24 \pm 0.57 \text{ cm}$ decrease, and was significantly greater than

Fig. 4 Porewater DOC concentrations at the three study locations



the brackish and saltwater sites ($p = 0.0062$), which had elevations of -1.56 ± 0.35 cm and -1.48 ± 0.34 cm, respectively. The average carbon density of the surface core slices was 0.030 ± 0.0024 g C/cm³ at the freshwater site, 0.025 ± 0.0034 g C/cm³ at the brackish site, and 0.014 ± 0.0040 g C/cm³ at the saltwater site. These carbon density values multiplied by the decrease in elevation reported in Table 2 provide an estimate of the carbon missing from each plot of 1273.3 g C/m² at the freshwater site, 388.9 g C/m² at the brackish site, and 206.9 g C/m² at the saltwater site.

Discussion

The decomposition data showed no difference in the rate of decomposition between the treatment and reference plots at any salinity level. Therefore, the increased emissions from the treatment plots at the brackish and saltwater sites were most likely due to the decomposition of the newly labile material derived from the recently killed vegetation. DeLaune et al. (1990) estimated that at least 600–700 g C/m²/yr must be

sequestered to balance soil oxidation-decomposition losses and maintain accretion of wetlands in the Barataria Basin. Organic matter may be particularly important in the structure of many Gulf coast and Atlantic coast marsh soils because most soil volume is pore space supported by organic matter (Nyman et al. 2006; DeLaune and White 2011; DeLaune et al. 2013). Organic matter contributes to the soil matrix and increases structural strength by forming an interlocking network of roots, and the loss of root turgor following plant death likely leads to peat collapse (DeLaune et al. 1994). Thus, when vegetation dies, the highly organic marsh soils compact, often initiating new ponds in coastal marshes, after which marsh loss proceeds via edge erosion as reported elsewhere (Nyman et al. 1994; Schwimmer 2001).

DeLaune and Lindau (1987) found that a majority of the organic matter in coastal bay sediment was from phytoplankton as opposed to salt marsh plants, suggesting that much of the C lost from marshes was oxidized instead of exported and buried. Oxidation first removes easily decomposable material causing oxidative losses to decline over time. This study found that approximately 60 % of vegetation in litter bags

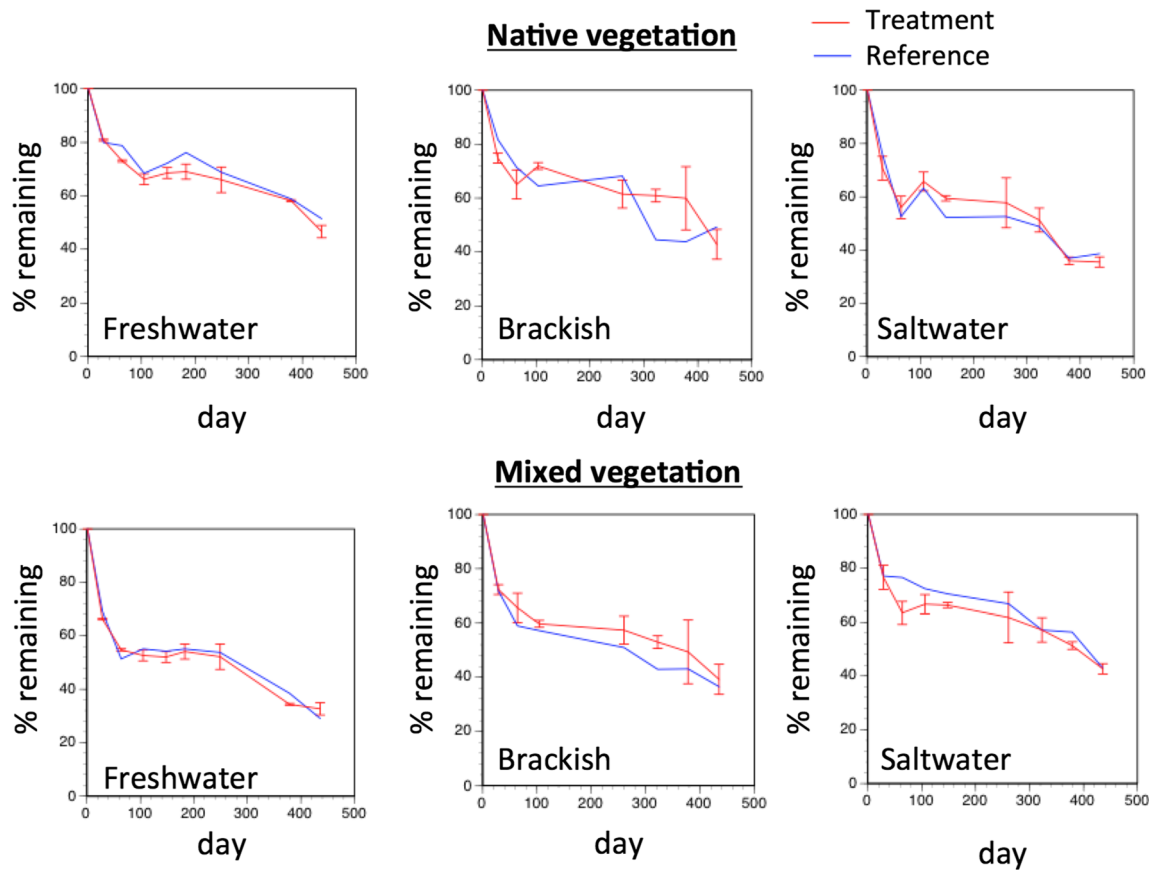


Fig. 5 Decomposition at the Treatment and Reference plots at the three study locations

was lost due to decomposition regardless of wetland type, with a majority of the loss occurring during the first half of the study, similar to other decomposition studies (i.e., Benner

et al. 1991; Rybczyk et al. 2002; Shaffer et al. 2015). Labile fractions of organic matter are rapidly decomposed into inorganic constituents, which are lost to the air (as CH_4 or CO_2) or

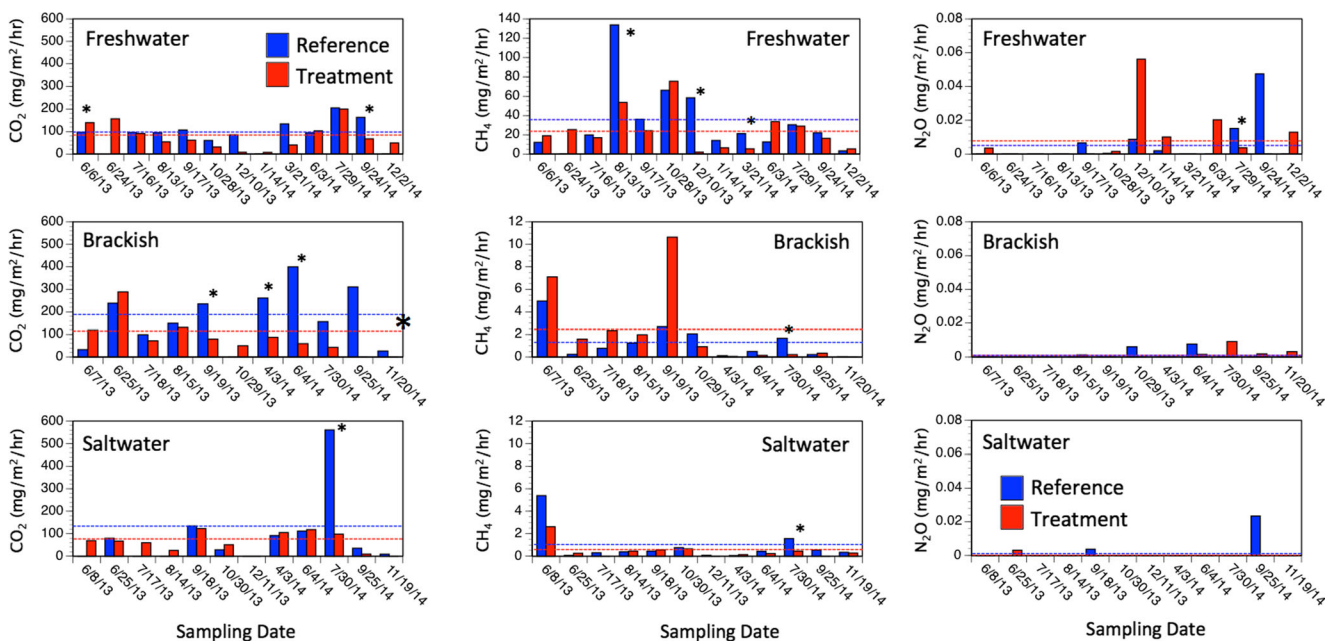


Fig. 6 Emissions of CO_2 , CH_4 and N_2O at the study plots. Asterisks indicate statistically significant difference between the treatment and reference plots. Dashed lines indicate overall means

Table 1 Mean rate and total emissions of CO₂, CH₄ and N₂O at the study sites

	CO ₂ (mg/m ² /h)	CO ₂ (g/m ²)	CH ₄ (mg/m ² /h)	CH ₄ (g/m ²)	N ₂ O (mg/m ² /h)	N ₂ O (g/m ²)
Freshwater - Tmt	84.3 ± 11.2	1003.4 ± 163.8	24.1 ± 3.9	311.4 ± 1.9	0.0084 ± 0.0044	0.1244 ± 0.0385
Freshwater - Ref	103.6 ± 16.0	1291.6	35.9 ± 10.4	421.3 *	0.0067 ± 0.0039	0.1085
Brackish - Tmt	116.3 ± 19.1	901.0 ± 171.0	2.3 ± 0.7	19 ± 2.5	0.0016 ± 0.0009	0.0242 ± 0.0193
Brackish - Ref	191.0 ± 38.1 *	2457.7 *	1.3 ± 0.4	14.0	0.0013 ± 0.0009	0.0254
Saltwater - Tmt	75.1 ± 8.3	813.8 ± 59.2	0.5 ± 0.2	4.1 ± 1.0	0.0109 ± 0.0078	0.1679 ± 0.0932
Saltwater - Ref	131.4 ± 63.2	1366.5 *	0.9 ± 0.4	7.6	0.0025 ± 0.0021	0.0347

Asterisks indicate statistically significant difference between the Treatment and Reference plots

exported in the water column as DOC, while recalcitrant fractions are mostly permanently buried in the soil column (Reddy and DeLaune 2008). Bouillon et al. (2008) found that CO₂ efflux from sediments and creek waters and tidal export of dissolved inorganic carbon (DIC) resulting from mineralization via pore water drainage to be major pathways for carbon in mangrove systems, and DIC was on average about eight times higher than for DOC (Bouillon and Connolly 2009).

Freshwater wetlands tend to be major sources of CH₄, a greenhouse gas 25 times more potent than CO₂, whereas the presence of sulfate in brackish and saltwater soils inhibits the production of CH₄ (Reddy and Delaune 2008). Similarly, in this study, CH₄ emissions from the reference plot at the freshwater site (280.9 g CH₄/m²/yr) was more

than an order of magnitude larger than that of the brackish (9.3 g CH₄/m²/yr) or saltwater (5.1 g CH₄/m²/yr) reference sites. This is in agreement with other studies that found CH₄ emissions to range from 0.8–365 g CH₄/m²/yr at freshwater wetlands (Alford et al. 1997; Delaune and Pezeshki 2003; Li and Mitsch 2016), 0.1–27 g CH₄/m²/yr at brackish wetlands (Poffenbarger et al. 2011; Li and Mitsch 2016), and 0–5.7 g CH₄/m²/yr at saltwater wetlands (Delaune et al. 1983; Poffenbarger et al. 2011). Holm et al. (2016) suggest using the relationship between salinity and CH₄ emissions provided by Poffenbarger et al. (2011) to determine baseline emissions of CH₄. This would correspond to approximately 27, 13, and 4 g CH₄/m²/yr, respectively, for the freshwater, brackish and saltwater sites of this study, suggesting that this method adequately estimates brackish and saltwater wetland emissions, but underestimates CH₄ emissions for freshwater wetlands. The main factors besides salinity controlling methane emissions from wetlands are soil temperature, water depth, and the amount and quality of decomposable substrate (Reddy and Delaune 2008). In addition, the species of vegetation has been shown to influence the relative production of CH₄ versus CO₂ (Roden and Wetzel 1996). Since the saline environment of brackish and saltwater marshes inhibits the production of methane, brackish and saltwater marshes have a much greater capacity for net carbon storage than freshwater wetlands (Bridgman et al. 2006).

Total carbon stocks in the upper 50 cm of the soil horizon in this study were 15.0 ± 0.5 kg C/m² at the freshwater site, 11.1 ± 0.9 kg C/m² at the brackish site, and 8.5 ± 1.4 kg C/m² at the saltwater site. These values are comparable to ranges of C stocks reported by other studies, such as 3.4–4.7 kg C/m² for wetlands in the northern Gulf of Mexico coastal region (Hansen and Nestlerode 2014); 10–25 kg C/m² at a coastal wetland in north Florida (Choi and Wang 2004); and 1.2 to 22.2 kg C/m² for mangroves surrounding Términos Lagoon in Carmen Island, Campeche-México (Cerón-Bretón et al. 2010). When the carbon stocks reported in this study are converted to CO₂ equivalents (CO₂e) on a per acre basis, as is used in carbon accounting, they equate to 222.8 mt

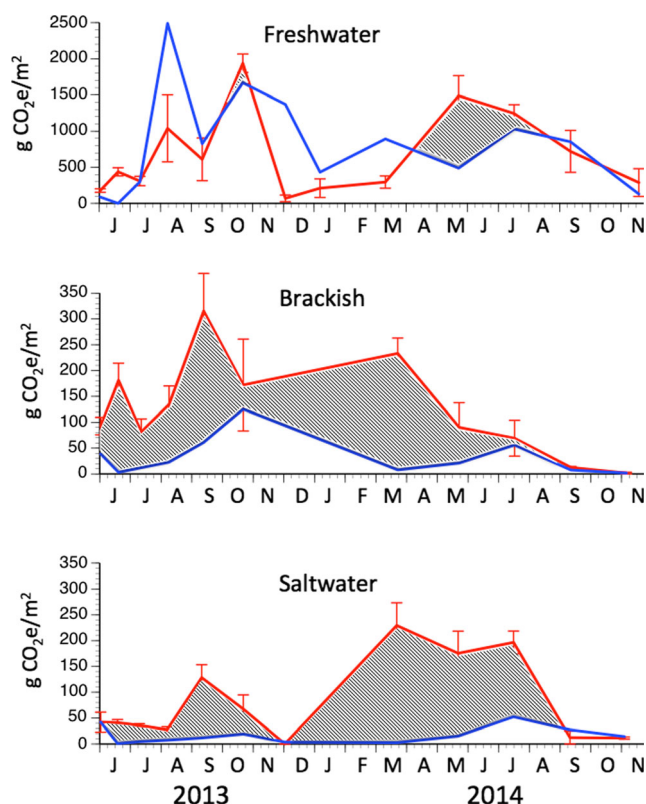
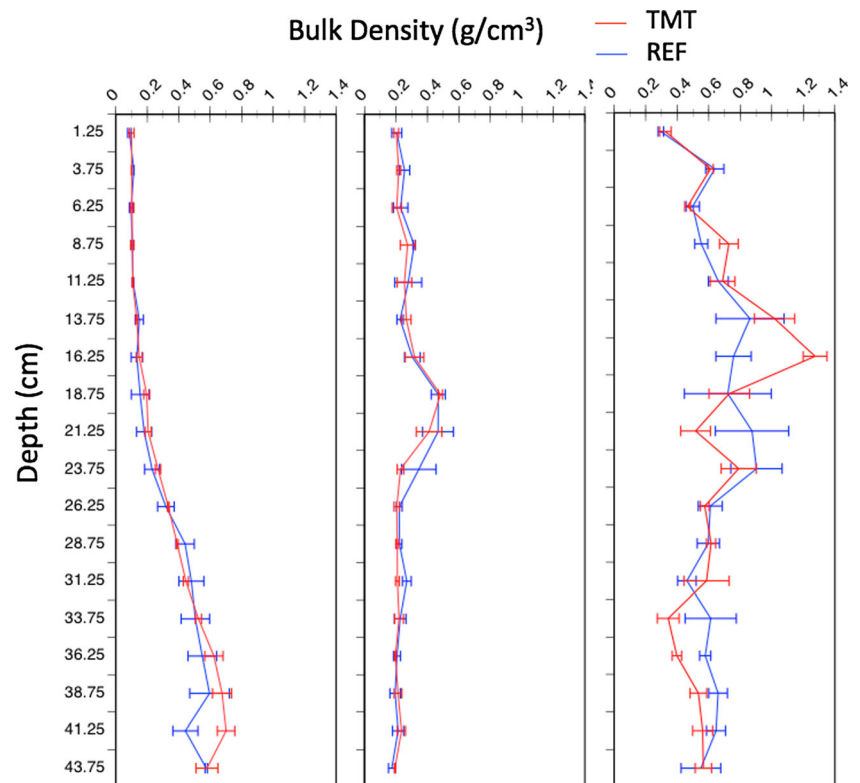


Fig. 7 Greenhouse gas emissions as CO₂e for the treatment (red) and reference (blue) plots at the three study locations

Fig. 8 Bulk density with depth at the three study locations



$\text{CO}_2\text{e/ac}$ for freshwater, 164.7 mt $\text{CO}_2\text{e/ac}$ for brackish, and 125.7 mt $\text{CO}_2\text{e/ac}$ for saltwater wetlands.

There was a decrease in elevation at the treatment sites compared to the surrounding wetlands. The 1273.3 g C/m^2

Fig. 9 Percent carbon with depth at the three study locations

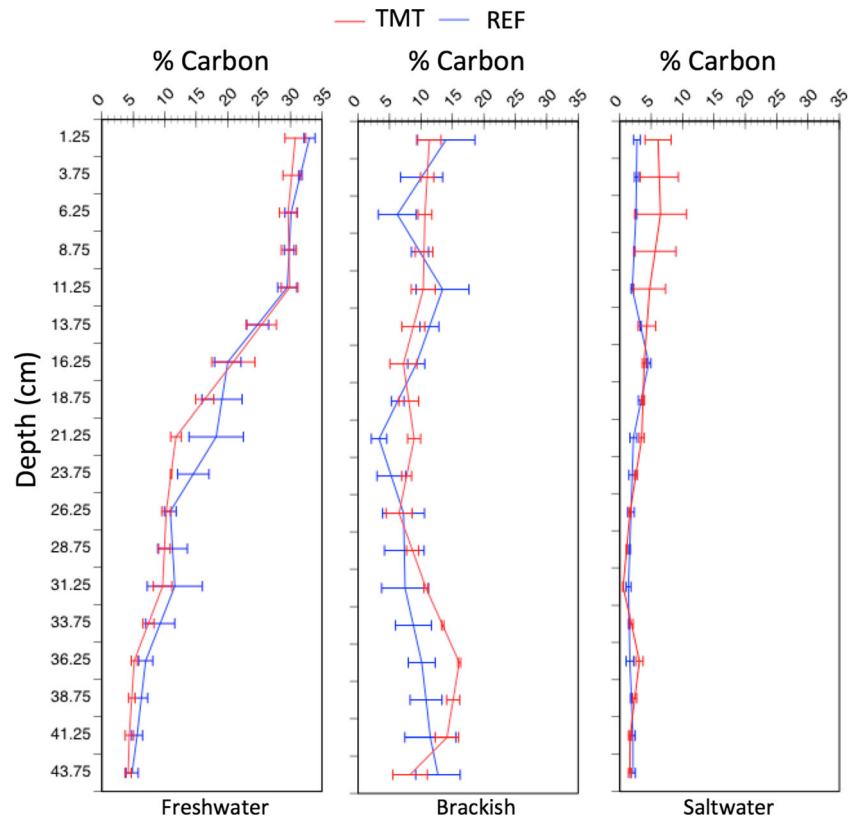
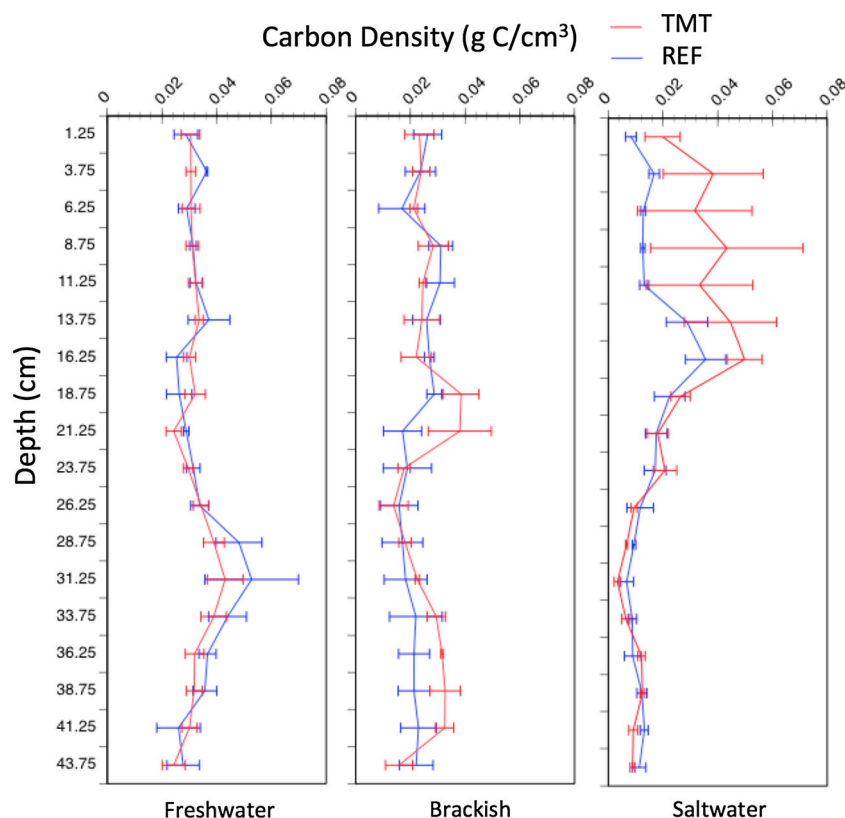


Fig. 10 Carbon density with depth at the three study locations



missing at the freshwater site is very close to the amount of carbon released as CH_4 and CO_2 , which was 1314.7 g C/m^2 , respectively. Thus, approximately 8.5–8.8 % of the total carbon content in the surface 50 cm of the soil profile at the freshwater site ($15,010 \text{ g C/m}^2$) was lost during the experiment. The brackish and the saltwater sites did not agree as well, with an observed surface carbon loss of 388.9 g C/m^2 and 206.9 g C/m^2 , respectively, but measured emissions of 920.0 g C/m^2 and 817.9 g C/m^2 , respectively. The ranges of these losses compared to the carbon content of the soil profiles at the brackish ($11,100 \text{ g C/m}^2$) and saltwater (8470 g C/m^2) sites were 3.5–8.3 % and 2.4–9.6 %, respectively (Table 3). This suggests that some other process was occurring at the brackish and saltwater sites other than the decomposition of organic matter.

Although the results at the freshwater site were inconclusive, the treatment plots at both the brackish and

saltwater sites emitted significantly more greenhouse gasses compared to the reference plots. There was a net difference of 1026.4 and $766.1 \text{ g CO}_2\text{e/m}^2$, respectively, for the brackish and saltwater sites over the 1.5 years of the study. This is equivalent to 4.2 and $3.1 \text{ mt CO}_2\text{e/acre}$, respectively, for the brackish and saltwater sites over the 1.5-year study.

When this experiment was first conceived, we hypothesized that over 50 % of the available carbon would be mineralized within one to two years. This percentage of mineralization was supported by the literature (Mittra et al. 2005), which was more speculative rather than proven. For example, Pendleton et al. (2012) carried out an entire analysis of emissions from converted and degraded coastal ecosystems using 25–100 % mineralization, and Siikamäki et al. (2012) used a 75 % conversion rate for their analysis of emissions after destruction of mangroves. DeLaune and White (2011) estimated that the majority of the organic matter lost through marsh deterioration would be exported to estuaries or offshore areas, with a significant portion of the organic matter oxidized in the process. However, this study found less than 10 % of available carbon was actually mineralized. Given this diminished response, additional reference plots would have been useful to aid in determining significant differences in this study. Since oxidation first removes the most easily decomposable material, oxidative losses decline with time, thus the experiment

Table 2 Elevation (cm) change of treatment plots at the end of the 1.5 year study

Plot	Freshwater	Brackish	Saltwater
1	-4.01	-1.60	-1.23
2	-3.40	-2.14	-2.15
3	-5.32	-0.93	-1.06
mean $\pm$ s.e.	-4.24 ± 0.57	-1.56 ± 0.35	-1.48 ± 0.34

Table 3 Total carbon stock in the top 50 cm of the soil horizon followed by the amount of carbon lost from the decrease in elevation and measured as greenhouse gasses, and the relative the percentage of the total soil carbon stock lost through each pathway

Wetland Site	C stock <50 cm (g C/m ²)	Elevation C lost (g C/m ²)	% C stock <50 cm	GHG C released (g C/m ²)	% C stock <50 cm
Fresh	15,010	1273.3	8.5 %	1314.7	8.8 %
Brackish	11,100	388.9	3.5 %	920.0	8.3 %
Saltwater	8470	206.9	2.4 %	817.9	9.6 %

presented here most likely captured the majority of the emission response from the treatment plots and further significant emissions were unlikely.

Overall, the amount of carbon sequestered is highly dependent on the health and productivity of the wetland, as large amounts of previously stored carbon could potentially be released to the atmosphere when wetland vegetation dies (Davidson and Janssens 2006; DeLaune and White 2011; Mcleod et al. 2011; Pendleton et al. 2012). Successful wetland restoration creates conditions for healthy, thriving wetland systems that are optimal for the sequestration and burial of carbon and prevent the release of carbon to the atmosphere. Thus, the most effective method to maintain wetland carbon pools and prevent emissions to the atmosphere is to avoid conversion and drainage through protection and sustainable management. Louisiana's Comprehensive Master Plan for a Sustainable Coast provides a long-term, state-wide, action-specific plan to achieve the state's coastal protection and restoration. The plan details that restoration activities can prevent up to 2850 km² from converting to open water over the next 50 years (CPRA 2012). Unfortunately, Louisiana could be more than \$70 billion short of the funding needed to implement the state's 50-year coastal protection and restoration plan (Davis et al. 2015).

Providing wetland offset credits for prevented wetland loss in the Mississippi River Delta may be essential to creating a strong business case for carbon investment into wetland restoration projects. The costs of most restoration techniques exceed carbon revenue streams when accounting for wetland sequestration alone. The large offset potential contained in the top 50 cm of the soil profile demonstrates the importance of quantifying the carbon benefit of preventing wetland loss. Quantifying wetland sequestration and prevented loss will optimize the amount of offsets that can be achieved from a specific restoration project. This research supports claiming carbon offsets for restoration efforts that are successful in preventing the loss of the wetland soil horizon. Further research is needed to more fully quantify the proportions of material in the top 50 cm of the soil profile that is decomposed in-situ and results in CO₂ and/or CH₄ emissions, versus the proportion that is transported out of the project boundary and

results in emissions, or is buried (Mack et al. 2012). Including the prevention of wetland loss in carbon monitoring and accounting may provide stronger financial incentives to develop wetland offset projects to support private investment into wetland restoration and conservation.

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