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# Sources and Chemical Stability of Soil Organic Carbon along a Salinity Gradient and a Chronosequence of Created Brackish Marshes in Coastal Louisiana

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SOURCES AND CHEMICAL STABILITY OF SOIL ORGANIC CARBON  
ALONG A SALINITY GRADIENT AND A CHRONOSEQUENCE OF  
CREATED BRACKISH MARSHES IN COASTAL LOUISIANA

A Thesis

Submitted to Graduate Faculty of the  
Louisiana State University and  
Agricultural and Mechanical College  
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requirements for the degree of  
Master of Science

in

The Department of Oceanography and Coastal Sciences

by  
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## ABSTRACT

Tidal marshes have been recognized for providing a number of important ecological services including soil carbon sequestration. However, the loss of tidal marsh habitat due to climate change and anthropogenic stressors exposes previously stored soil organic carbon (SOC) to oxidation. The vulnerability of SOC to oxidation depends on its chemical stability and environmental conditions limiting decomposition. Labile organic carbon (LC), decomposes quickly unless abiotic conditions limit decomposition. Recalcitrant organic carbon (RC) decomposes slower and is stored for longer time periods. Predicting long-term storage of SOC is complicated by the potential for multiple SOC sources, differences in chemical stability, and variation in environmental conditions that may preserve chemically LC. To increase understanding of soil carbon dynamics in tidal marshes, SOC sources and chemical stabilities were assessed along an estuarine salinity gradient, and along a time series of marsh creation. Additionally, relationships between labile and recalcitrant carbon, vegetation characteristics, and environmental factors (i.e. elevation, mineral sediment) were examined. Soil cores were collected in tidal freshwater (n = 4), brackish (n = 3), and salt (n = 4) marshes in Barataria Bay, Louisiana, and in six created dredge sediment marshes across a 32-year chronosequence, and a natural reference marsh (n = 6) in Sabine National Wildlife Refuge in southwestern Louisiana. To examine the source (algal, C<sub>4</sub>, or C<sub>3</sub> plant) of total organic and refractory SOC,  $\delta^{13}\text{C}$  analyses were used. Acid hydrolysis digestion was used to fractionate the SOC into 'labile' and 'refractory' components. Across all marshes an average of 73% of SOC was recalcitrant indicating it is chemically stable. Across the salinity gradient, LC, RC and SOC densities were highest in the freshwater

marshes. LC and RC stocks increased with marsh age in the created marshes, but the RC stock increased two times faster. LC and RC accumulation rates were not significantly different between created marshes of different ages and the natural reference marsh. Overall this study illustrates that a majority of SOC in coastal marshes of Louisiana is derived from local vegetation and is chemically stable therefore more likely to contribute to long term carbon storage.

## GENERAL INTRODUCTION

Tidal wetlands have been recognized for numerous ecosystem services including carbon sequestration (Chmura et al. 2003), however carbon stored in coastal soils is vulnerable to climate change and anthropogenic stressors (DeLaune and White 2012). Tidal marshes may become more saline with sea level rise or convert to open water, and it is not understood how these changes may affect carbon reservoirs. Additionally, in response to land loss marshes have been created by depositing dredge material into open water and vegetation allowed to colonize (Edwards and Proffitt 2003). Sources of carbon in tidal wetlands include biomass from local vegetation, algae, and allochthonous material that is deposited on the marsh surface and buried (Reddy and DeLaune 2008). The amount of each of these carbon sources will vary by spatially along the salinity gradient and temporally as created marshes age. Each carbon source has a different composition that determines the stability that may be important in determining the residence time and therefore long-term carbon storage. However, despite the importance of carbon stability for long term storage (Mudd et al. 2009), there are few studies examining carbon stability in coastal marshes. This study examines carbon source and stability spatially across a salinity gradient in Barataria Bay, Louisiana, and temporally across a chronosequence of created brackish marshes in southwest Louisiana to determine the capacity of long-term carbon storage in natural and created marshes.

# CHAPTER 1. SOURCES AND CHEMICAL STABILITY OF SOIL ORGANIC CARBON ACCUMULATION IN MARSHES ACROSS A SALINITY GRADIENT IN BARATARIA BAY, LOUISIANA

## 1.1. Introduction

Coastal wetlands have large stocks of soil organic carbon (SOC) due to high organic carbon accumulation rates (Chmura et al. 2003, Ouyang and Lee 2014) however, wetlands and their associated carbon stocks are vulnerable to climate change and anthropogenic impacts. Increased hurricane frequency may erode and increase exposure of coastal marsh sediment, while sea level rise may cause wetlands to become more saline or convert to open water (Boesch et al. 1994). Salinity increases in brackish and freshwater marshes will cause shifts in processes that affect SOC accumulation rates and stocks (Craft 2007, Herbert et al. 2015). Increases of salinity in freshwater and brackish marshes will stimulate mineralization rates (Weston et al. 2006, Chambers et al. 2011), and decrease plant productivity (Neubauer 2013). Coastal wetland loss increases the potential for microbial decomposition and release of previously sequestered SOC as CO<sub>2</sub> further contributing to increases in atmospheric CO<sub>2</sub> concentrations (DeLaune and White 2012). The coastal wetlands of Louisiana are particularly vulnerable to changes in salinity and loss due to high rates of relative sea level rise and anthropogenic alterations to the Mississippi River and installation of numerous channels and canals among other impacts (Boesch et al. 1994, Turner 1997), resulting in the loss of approximately 25% of the coastal plain from the 1932 to 2016 (Couvillion et al. 2017). It has been estimated that 1.86 metric tons x 10<sup>6</sup> of carbon is released on an annual basis from Louisiana marshes due to this land loss (DeLaune and White 2012), but it is still not well understood how carbon source, and chemical



stability, may influence carbon storage and accumulation as wetlands become more saline or convert to open water.

Tidal marshes along estuary salinity gradients vary in their plant community composition (Odum 1988), microbial decomposition pathways (Weston et al. 2006, Neubauer 2013), and relative contributions of riverine and coastal sediment (Noe et al. 2016). In a meta-analysis of over 61 estuaries in the United States, average rates of SOC accumulation (CAR) were higher in brackish marshes ( $240 \pm 30 \text{ g m}^{-2} \text{ yr}^{-1}$ ) than freshwater marshes ( $140 \pm 20$ ) and salt marshes ( $190 \pm 40 \text{ g m}^{-2} \text{ yr}^{-1}$ ; Craft, 2007), potentially due to very high SOC concentrations, possibly associated with high phosphorus content, which can be a limiting nutrient. In riverine systems of coastal Georgia freshwater input promoted organic matter preservation and accumulation resulting in SOC accumulation rates three times greater in freshwater marshes ( $108 \pm 21 \text{ g m}^{-2} \text{ yr}^{-1}$ ) than brackish ( $32 \pm 6 \text{ g m}^{-2} \text{ yr}^{-1}$ ) and salt marshes ( $21 \pm 5 \text{ g m}^{-2} \text{ yr}^{-1}$ ; Craft, 2007). Relatively low SOC accumulation rates in salt marshes was linked to faster root decomposition rates in the salt marshes (41 - 49% lost after 1 year) compared to the brackish (29-30% lost), and freshwater marshes (30-36% lost; Craft, 2007). Along the Gulf Coast, where many marshes have been isolated from river sediment by levees, freshwater marshes tend to have low CAR (averaging  $150 \text{ g m}^{-2} \text{ yr}^{-1}$ ) compared to brackish marshes ( $\sim 300 \text{ g m}^{-2} \text{ yr}^{-1}$ ), and salt marshes ( $\sim 310 \text{ g m}^{-2} \text{ yr}^{-1}$ ; Craft 2007). Freshwater marshes in Barataria Basin tend to have less sediment and high SOC concentrations, while salt marshes closer to the Gulf of Mexico have greater exposure to coastal sediment and as a result have higher bulk densities and lower SOC concentrations (Reddy and DeLaune 2008, Dodla et al. 2012). What is not well

established is whether there are differences in SOC sources among fresh, brackish, and salt marshes, and whether the chemical stability of the SOC differs among these marsh types.

Freshwater marshes tend to be dominated by plants which use the C<sub>3</sub> photosynthetic pathway such as *Panicum hemitomon* and *Sagittaria latifolia* while brackish and salt marshes are dominated by C<sub>4</sub> grasses such as *Spartina patens* and *Spartina alterniflora* (reclassified as *Sporobolus alterniflorus*) (Chmura et al. 1987, DeLaune and Lindau 1987, Chmura and Aharon 1995). Allochthonous carbon may be variably sourced from terrestrial watersheds, eroded wetland soil, or phytoplankton (Ember et al. 1987, Leorri et al. 2018, Van de Broek et al. 2018) and the degree to which these allochthonous sources are deposited in marsh sediment along the salinity gradient may affect soil carbon dynamics. C<sub>3</sub> plants, which dominate terrestrial ecosystems and freshwater wetlands, have a greater photosynthetic discrimination against <sup>13</sup>CO<sub>2</sub> in favor of the <sup>12</sup>CO<sub>2</sub> resulting in δ<sup>13</sup>C values between -23 and -34‰ (Smith and Epstein 1971). The additional Hatch-Slack pathway of C<sub>4</sub> plants results in less discrimination of <sup>13</sup>CO<sub>2</sub> compared to the C<sub>3</sub> plants (Vogel 1980, O'Leary 1981). As a result, the δ<sup>13</sup>C range of most C<sub>4</sub> plants is between -6 and -19‰ (Smith and Epstein 1971). Algae are CAM plants and have intermediate δ<sup>13</sup>C values averaging -21‰ (Sullivan and Moncreiff 1990). Despite the potential for allochthonous carbon deposition, previous studies in Barataria Bay, Louisiana illustrated that the dominant SOC source in surface soils generally reflected the local vegetation in freshwater, intermediate, brackish and salt marshes (DeLaune 1986, Chmura et al. 1987). In the Scheldt Estuary, repeated sampling of soils in summer and winter months show an

algae  $\delta^{13}\text{C}$  signature in the surface sediments during the summer but not the winter. Indicating that algae are productive in the summer and contribute to SOC shortly after their deposition but decompose and do not contribute to long-term SOC storage since the  $\delta^{13}\text{C}$  of samples collected in winter did not reflect an algae signature (Van de Broek et al. 2018). In marshes of the Gulf Coast, algal production tends to be low compared to marshes of the Atlantic and California coasts (Sullivan and Currin 2002) and, in Louisiana, primary production of benthic algae in brackish and salt marshes was too low to influence soil  $\delta^{13}\text{C}$  values (Chmura et al. 1987).

Carbon sources may vary in their physical and chemical properties and, therefore, their susceptibility to microbial decomposition (Stagg et al. 2018). Algae, composed of labile compounds, decompose easily and are thought not to contribute to long term SOC storage (Van de Broek et al. 2018) unless environmental conditions such as anoxia limit labile organic carbon (LC) decomposition (Rovira and Vallejo 2003). LC is characterized by simple molecules such as carbohydrates and polysaccharides (Benner et al. 1987, Reddy and DeLaune 2008) that are thermodynamically favorable carbon sources for microbial respiration, and therefore, have residence times of a few years to several decades (Cheng et al. 2007). Carbon inputs from marsh grasses, such as *Spartina sp.*, tend to have a higher proportion of recalcitrant organic carbon (RC), composed of complex molecules including lignin, humic acids and phenolic compounds (Benner et al. 1987, Reddy and DeLaune 2008), which are more resistant to decomposition, and can have mean residence times of hundreds to thousands of years (Cheng et al. 2007). These recalcitrant carbon compounds are believed to be the dominant contributor to long-term SOC storage (Paul

et al. 1997, Silveira et al. 2008). Importantly, several mechanisms in coastal wetlands allow the preservation and accumulation of SOC at various stages of physical and chemical decay including labile compounds (Reddy and DeLaune 2008). Anaerobic conditions limit the organic carbon oxidation and associations with mineral substrates may also physio chemically protect labile compounds from decomposition (Benner et al. 1984, Rovira and Vallejo 2003, Unger et al. 2016).

The relative lability or recalcitrance of SOC is operationally defined by methodologies that attempt to isolate easily oxidized compounds from more refractory compounds. Methods used to separate LC and RC include cold DI water, hot water (70°C), or salt extraction with K<sub>2</sub>SO<sub>4</sub> (Dodla et al. 2012). Other methods used include measuring differences in SOC in microbial biomass before and after fumigation (McLauchlan and Hobbie 2004), or measuring the amount of carbon dioxide, or methane respired during an incubation (McLauchlan and Hobbie 2004, Dodla et al. 2012). Methods such as acid hydrolysis dissolves the LC component and the remaining recalcitrant carbon is measured to calculate the percent of LC removed. A comparison of various methodologies show positive but varying slopes in the relationship between LC and SOC, indicating that the proportion of LC removed varied by method (McLauchlan and Hobbie 2004, Dodla et al. 2012), therefore caution should be used when comparing studies of differing methods, but when comparing within the same method, the patterns are reliable. The assumption for these methods is that the property that makes the SOC degradable, makes it susceptible to chemical, thermal, or microbial extraction (McLauchlan and Hobbie 2004, Dodla et al. 2012). Solid-state <sup>13</sup>C Nuclear Magnetic Resonance (NMR) spectroscopy has illustrated that labile compounds such as

carbohydrates and amino acids are removed during acid hydrolysis, and the remaining residue is composed of chemically resistant compounds (Silveira et al. 2008).

Additionally,  $^{14}\text{C}$  dating indicated that the non-hydrolyzable RC had an average age 1200 - 1400 years older than the SOC in surface soils providing support for the refractory nature of carbon remaining post-hydrolysis (Paul et al. 2006).

Freshwater, brackish, and salt marshes may differ in their process rates, such as primary productivity, microbial decomposition, and sedimentation, which affect SOC accumulation rates and the relative quantity of labile and recalcitrant SOC (DeBusk and Reddy 1998, Jobbágy and Jackson 2000, Lützow et al. 2006, Kirwan and Blum 2011). Aboveground productivity tends to be higher in brackish and salt marshes than freshwater marshes along the Gulf Coast (Figure 1.1; White and Simmons 1988, Pezeshki and DeLaune 1991, Stagg et al 2017b). Fewer studies have measured belowground productivity, however, across the salinity gradient in Louisiana, belowground productivity was found to be similar among wetland types

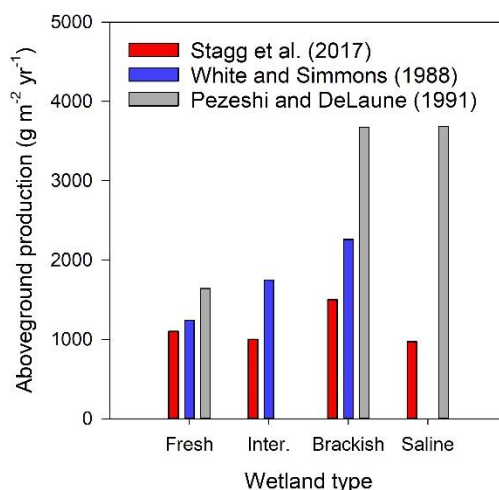


Figure 1.1. Published values of aboveground productivity in freshwater, brackish, and salt marshes in Barataria Bay, Louisiana.

(Stagg et al. 2017b). Higher aboveground productivity rates in brackish and saline marshes have been attributed to the dominance of the C<sub>4</sub> plants that are efficient in water use and photosynthesis, compared to C<sub>3</sub> plants that dominate in freshwater marshes (DeLaune 1986, Baldocchi 1994, Chmura and Aharon 1995, Stagg et al. 2017b). Decomposition rates of local plant litter tend to be lower in salt marshes as compared to freshwater marshes (McKee and Seneca 1982, Foote and Reynolds 1997, Laursen 2004, Stagg et al. 2018), which has been linked to higher lignin: nitrogen ratios in salt marsh plants compared to freshwater plants (Stagg et al. 2018). Lignin content tends to be higher in polyhaline species including *Spartina alterniflora*, *Spartina patens*, and *Juncus roemerianus*, and higher elevation freshwater species including *Typha latifolia*, and *Panicum hemitomon* than lower elevation freshwater species such as *Sagittaria lancifolia*, and *Schoenoplectus americanus* (Odum 1988, Stagg et al. 2018). Lignin content of soils across a salinity gradient was lower in the brackish marsh, dominated by *J. roemerianus* and *S. patens* than the salt marsh, dominated by *S. alterniflora*, and the freshwater marsh which composed of *S. lancifolia*, *Zizaniopsis miliacea*, and *Alternanthera philoxeroides* (Chambers et al. 2013). SOC accumulation rates are therefore expected to be highest in salt marshes and lowest in freshwater marshes with higher rates of aboveground productivity, lower rates decomposition, and higher rates of sediment deposition (Nyman et al. 1990, Stagg et al. 2017b, Stagg et al. 2018). The relative fraction of LC and RC, however, may be influenced by both litter quality and local environmental conditions that can protect and preserve LC.

Numerical models have shown that recalcitrant SOC is vital to long term carbon storage in coastal marshes (Mudd et al. 2009), however, only a few studies have

included this stable fraction in their assessment of soil carbon (see Cheng et al. 2008, Dodla et al. 2012, Gao et al. 2016, Unger et al. 2016, Leorri et al. 2018, Van de Broek et al. 2018). In the Mississippi River Delta, chemical fractionation of the SOC illustrated a tight relationship between SOC and LC ( $R^2 = 0.76$ ), with LC accounting for 27.5% of SOC in the top 1.5 meters (Dodla et al. 2012). In four marshes of the North Atlantic, marshes within larger estuaries with steeper watershed gradients had a greater fraction of SOC as older RC (Leorri et al. 2018). Estuaries separated from their respective river had the smallest RC fraction (the percent of SOC that was RC) and youngest  $^{14}\text{C}$  ages, since more SOC by concentration was sequestered in-situ (Leorri et al. 2018). In one of the only studies to measure LC and RC accumulation rates (salt marshes of the mid-Atlantic), RC accumulation rate was relatively uniform (63 to 95  $\text{g m}^{-2}\text{yr}^{-1}$ ) across these marshes with a large range of SOC accumulation rates. LC accumulation rate, however, ranged between 37 to 238  $\text{g m}^{-2}\text{yr}^{-1}$  and accounted for 69% of the variability in SOC accumulation rates and was positively related to mineral sediment accumulation (Unger et al. 2016). The positive relationship between LC accumulation rate and mineral sediment was attributed to high LC input coincident with mineral sediment leading to rapid burial of LC to anaerobic depths, LC adsorbing to mineral surfaces (Lehmann et al. 2007), and/or protection by association within microaggregates (Rovira and Vallejo 2003).

The purpose of this study was to examine organic carbon sources and chemical stabilities in marshes across an estuarine salinity gradient. I hypothesized that the dominant SOC source, indicated by  $\delta^{13}\text{C}$ , will differ among fresh, brackish, and salt marshes, and changes in  $\delta^{13}\text{C}$  following hydrolysis (i.e., chemical removal of LC) will

reflect the relative labile and recalcitrant carbon content of the soil. Freshwater marshes were predicted to have a higher fraction of LC than brackish and salt marshes due to higher quality carbon sources, although these higher quality carbon sources may be decomposed at a faster rate than brackish or salt marshes (Figure 1.2). Salt marshes, which have plants with high rates of productivity and lower litter quality, lower decomposition rates, and greater mineral sediment supply are predicted to have greater RC accumulation as compared to brackish and freshwater marshes. LC is predicted to be positively related to live belowground biomass and mineral sediment stock, while RC is predicted to be positively related to dead biomass.

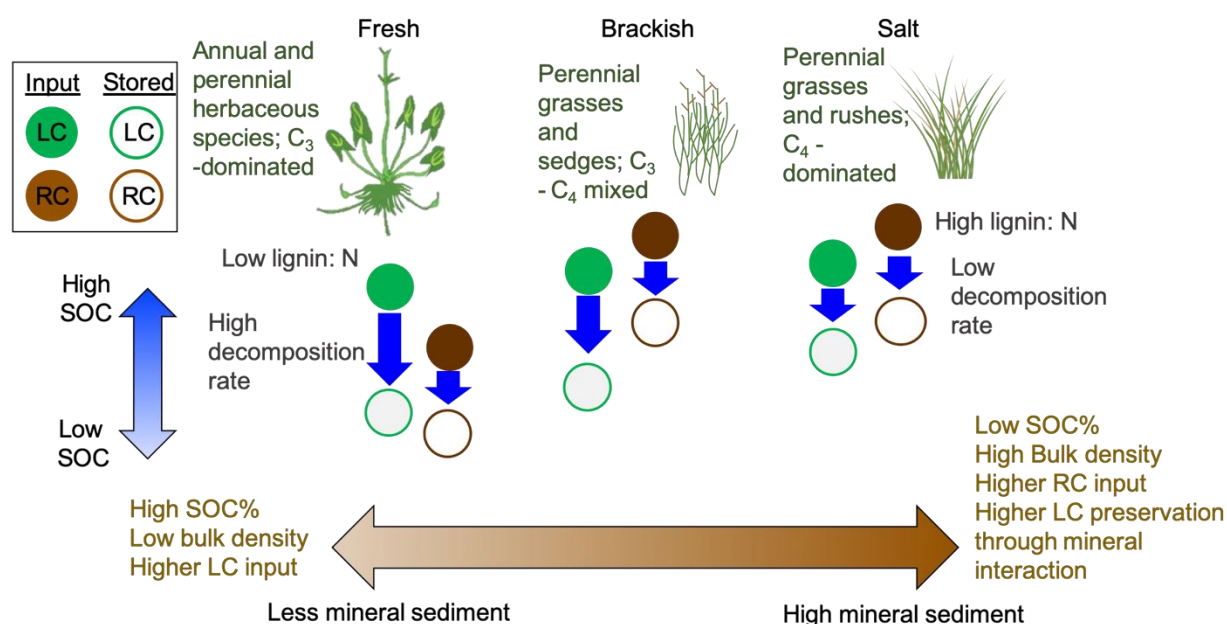


Figure 1.2. Conceptual diagram of labile and recalcitrant organic carbon dynamics in freshwater, brackish, and salt marshes. The height of the solid circles represents relative predicted SOC derived from plant productivity and the open circles indicate amount stored after decomposition, the length of the blue arrow reflects the relative amount of decomposition.



## 1.2. Methods

### 1.2.1. Study site

Barataria Basin is located south west of the Mississippi River and covers 6,300 km<sup>2</sup>, including 615 km<sup>2</sup> of swampland, 700 km<sup>2</sup> of freshwater marsh, 240 km<sup>2</sup> of intermediate marsh, 415 km<sup>2</sup> of brackish marsh, and 540 km<sup>2</sup> of salt marsh with the remaining area open water, unclassified, or inhabited (CWPPRA 2018). Barataria Basin is an intertributary basin between the former St. Bernard delta (4.0-2 ka), Lafourche delta (2.5-.5 ka), and the modern Plaquemine-Balize delta (1.3 ka – present; Blum and Roberts 2009). Abandoned delta lobes experience high rates of land loss due to subsidence and sediment compaction that can exceed sediment deposition in areas without river sediment input (Roberts 1997). Levees along the Mississippi River have eliminated the hydrologic connection between the river and adjacent wetlands, leading to increased subsidence (Twilley et al. 2016) and a high rate of relative sea-level rise (Penland and Ramsey 1990). The large wetland area, rapid sea-level rise and marsh deterioration make Barataria Basin an important region to examine marsh soil carbon properties.

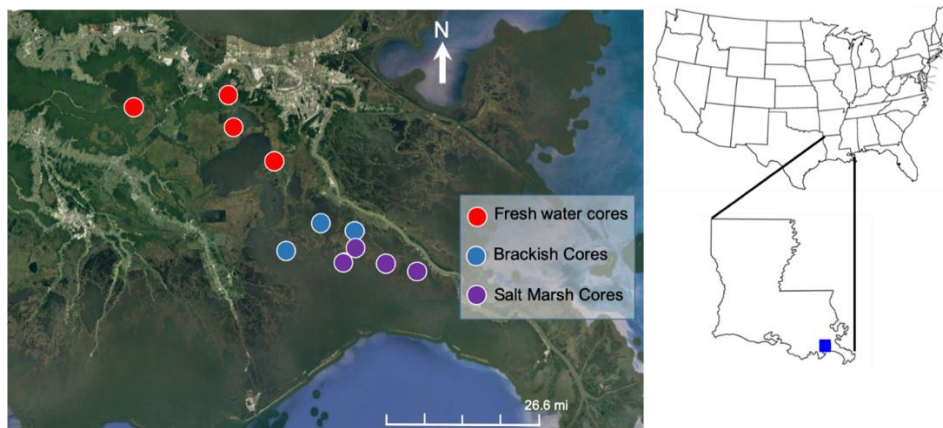


Figure 1.3. Map of core locations in Barataria Bay, LA. Red, blue and purple represent core locations in freshwater (n = 4), brackish (n = 3), and salt marsh (n=4), respectively.

### 1.2.2. Field sampling

Data for this study were obtained from eleven soil cores selected from Shrull (2018), that were 35 cm deep and 10.16 cm diameter, collected in the spring and fall of 2016 at Coast-wide Reference Monitoring Station (CRMS) sites across Barataria Basin, Louisiana, USA (Table A.1). Only 4 of the original 25 cores had sufficient depths for  $^{210}\text{Pb}$  accretion calculations, therefore cores were selected based on having a clear  $^{137}\text{Cs}$  peak in freshwater ( $n = 4$ ), brackish ( $n = 3$ ), and salt ( $n = 4$ ) marshes in the 2016 growing season (Figure 1.3). One freshwater core (FW3) was collected near the Davis Pond Diversion and has higher bulk densities and lower percent SOC concentrations than the other freshwater marsh cores. Cores were extracted using an aluminum push core device that was 60.8 cm long. Following collection, cores were frozen to ensure no internal mixing or mold growth, and that the stratigraphy remained intact. A band saw was used to cut cores into slices at 2 cm intervals.

### 1.2.3. Soil analysis

Bulk density was measured for each 2 cm section by drying samples to constant weight at 60°C and dividing by the volume (*in* Shrull 2018). Loss on Ignition (LOI) was used to determine the percent organic matter (OM) content and mineral mass weight percentages by incinerating samples at 550°C, and the difference in mass before and after burning is the organic matter fraction, and the remaining mass is the fraction of mineral sediment. Bulk density was lower in the freshwater marsh ( $0.14 \pm 0.10 \text{ g cm}^{-3}$ ) than the brackish ( $0.25 \pm 0.12 \text{ g cm}^{-3}$ ) and salt ( $0.30 \pm 0.09 \text{ g cm}^{-3}$ ) marshes (Shrull 2018). OM percent was about two times higher in the freshwater marsh ( $60 \pm 20\%$ )

compared to the brackish ( $33 \pm 8\%$ ), and salt ( $23 \pm 7\%$ ) marshes (Shrull 2018). Bulk density was inversely related to percent OM (LOI;  $R^2 = 0.83$ ; Shrull 2018).

OM volume and mineral sediment volume were calculated by the following equations:

$$\text{OM volume} = \left( \frac{(\text{dry weight (g)} * \text{OM\%/100})}{1.14 \text{ g/cm}^3} \right) * 100 \quad (1)$$

Where dry weight is the dry weight of the sample slice, OM % determined by LOI, and 1.14 is the average particle density of OM in marshes (DeLaune et al. 1983).

$$\text{Mineral Volume} = \left( \frac{(\text{dry weight (g)} * \text{Mineral \%}/100)}{2.61 \left( \frac{\text{g}}{\text{cm}^3} \right)} \right) * 100 \quad (2)$$

Where dry weight is the dry weight of the sample slice, mineral % of determined by LOI, and 2.61 is the average particle density of mineral sediment in marshes (DeLaune et al. 1983). Profiles of OM, and mineral sediment, show variation within and among marshes (Figure A.1).

Accretion rates were determined using  $^{137}\text{Cs}$  chronology (*in* Shrull 2018). Briefly, samples were dried at  $55^\circ\text{C}$  to a constant weight, pulverized to homogenize the samples, and packed in plastic petri dishes, weighed, sealed with electrical tape, and allowed to reach secular equilibrium (3 weeks). Low Energy Germanium (LEGe) and Broad Energy Germanium (BEGe) detectors were used to analyze the samples. Both the LEGe and BEGe detectors from Canberra styles accurately assess the Ke V values that are necessary for  $^{137}\text{Cs}$ . Within 20 - 24 hours, samples were processed in the detectors. Radioactivity units of measurement used were disintegrations per minute (dpm) or  $1/60^{\text{th}}$  of a Becquerel (Bq) (Shrull 2018).

Vertical accretion rates were calculated using the  $^{137}\text{Cs}$  activity peak occurring in ca. 1963 (Richie et al. 1975, DeLaune et al. 1978, Chmura & Kosters 1994, Milan et al. 1995, Walling & He 1997). To calculate the vertical accretion rate, the midpoint depth of the interval ( $D_m$ ) of the  $^{137}\text{Cs}$  peak is divided by the time between collection and deposition. Samples in this study were collected in 2016 and therefore,  $^{137}\text{Cs}$  peak depth was divided by 53 years (Shrull 2018). Accretion rates from the original study, using 25 cores including one adjacent to the Davis Pond, did not vary significantly by wetland type, with  $0.65 \pm 0.12 \text{ cm yr}^{-1}$  in the freshwater,  $0.62 \pm 0.12 \text{ cm yr}^{-1}$  in the brackish, and  $0.64 \pm 0.10 \text{ cm yr}^{-1}$  in the salt marshes (Shrull 2018).

Labile and refractory C fractions were measured on every other depth interval, down to 35 cm, the depth of the shallowest core. RC was isolated using acid hydrolysis methods described by Unger et al. (2016). Briefly, 1.0 g of dry soil was placed into a glass vial, and 10 ml of deionized (DI) was added, followed by 20 ml of 6 M HCL. Samples were then placed on a digestion block at  $150^\circ\text{C}$  for 18 hours. After hydrolysis, samples were then transferred to a centrifuge tube and centrifuged at 2500 revolutions per minute for 10 minutes and the supernatant was immediately decanted. The sample was then dried for at  $60^\circ\text{C}$  until a constant weight was reached, and the dry weight was measured. Both the bulk sample (pre-hydrolysis) and residual fraction (post-hydrolysis) were analyzed for elemental carbon and  $\delta^{13}\text{C}$  using an elemental analyzer interfaced to a continuous flow isotope ratio mass spectrometer (IRMS) by University of California, Davis Laboratory. Samples were prepared by weighing 10  $\mu\text{g}$  of dry sample into silver capsules, inorganic carbon was removed by fuming with 12 M HCL for 24 hours, and then allowing samples to dry by blowing air over them under a heat lamp for 4-6 hours,

similar to (Hedges and Stern 1984). The percent of SOC that was refractory was calculated as:

$$\text{RC\% of SOC} = \frac{\text{Cpost hydrolosis (g)}}{\text{SOC original (g)}} = \frac{\text{Mass post hydrolosis (g)} * (\text{post hydrolosis C\%}) / 100}{\text{Mass pre hydrolosis (g)} * (\frac{\text{TOC (\%)}}{100})} \quad (3)$$

Where the mass post hydrolysis is the mass measured after hydrolysis, and post hydrolysis C% is the elemental carbon concentration measured on the recalcitrant sample after hydrolysis. The mass pre hydrolysis was the exact amount of 1 g of sample measured, and TOC% is the elemental C percent measured on bulk sample before hydrolysis.

The percent of organic carbon that was labile was calculated as:

$$\text{LC\% of SOC} = 100 - \text{RC\% of SOC} \quad (4)$$

The percent of the total soil mass that was refractory carbon (RC %) was calculated as:

$$\text{RC\%} = \text{SOC\%} * \frac{\text{RC\% of SOC}}{100} \quad (5)$$

Where SOC% is the elemental carbon percent of the bulk sample before hydrolysis.

The percent of the total soil mass that was labile carbon (LC %) was calculated as:

$$\text{LC\%} = \text{SOC(\%)} * \frac{\text{LC\% of SOC}}{100} \quad (6)$$

LC, RC, and SOC accumulation rates were calculated by multiplying carbon concentration by the bulk density to yield a carbon density. The carbon density for each depth was then multiplied by 2 cm (the depth of slice) and summed to the depth of the

$^{137}\text{Cs}$  peak to calculate a carbon stock ( $\text{g cm}^2$ ), multiplied by 10,000 to put on a square meter basis and divided by 53 to determine the accumulation rate ( $\text{g m}^{-2} \text{yr}^{-1}$ ).

#### 1.2.4. Vegetation analysis

Aboveground plant biomass, stem density and height were measured between August and October 2016 in three plots per core location (33 plots total) that were  $0.25 \text{ m}^2$  in size (Snedden pers. comm. CPRA). Aboveground biomass was sorted into live and dead and then dried and weighed. Stem density and average height were determined for each species. The average height was measured on 5 stems as a representative for the plot. Belowground biomass was collected in fall 2016 at the end of the growing season using three replicate cores of 10.16 cm diameter X 32 cm depth at each core location. Each core was divided into four sections of 8 cm intervals down to 32 cm (Snedden 2018). No biomass was collected at site FW4. Eight of the 33 belowground biomass cores did not extend the full 32 cm and the last section was excluded. Belowground biomass was sieved through a 2 mm sieve and material retained on the sieve was sorted into live and dead biomass. Macro-organic matter passed through a 2 mm sieve but was retained on a 0.5 mm sieve and was generally comprised of dead organic material. Relationships between SOC and belowground biomass was examined by using either the average SOC concentration (%) or SOC stock ( $\text{g C m}^{-2}$ ) and the belowground biomass stock ( $\text{g m}^{-2}$ ) at each respective depth interval.

Vegetation percent cover for sites in 2016 were obtained from CPRA (2019) online database. This data was used to calculate the estimated value of soil  $\delta^{13}\text{C}$  by using the following isotope mixing model below (equation 7), modified from Chmura et

al. (1987) which used percent biomass instead of percent cover. Since vegetation species were recorded at time of soil collection, the percent cover value provides a more complete representation of species present than the aboveground biomass species.

$$\delta^{13}\text{C soil} = \frac{\sum_{n=1}^i (\% \text{ cover } i)(\delta^{13}\text{C})}{\sum_{n=1}^i (\% \text{ cover})} \quad (7)$$

Where  $i$  = each species found in the wetland site

#### 1.2.5. Statistical analysis

Data were transformed either by log, square root, or logit functions with the latter used for percentage data, to improve data normality and homogeneity of variance when necessary. One-way analysis of variance (ANOVA) was used to test for differences in LC, RC, and SOC concentration, density and accumulation rates between wetland types (freshwater, brackish marsh, and salt marsh). ANOVA was also used to test for differences between marsh types in other soil parameters including soil bulk density, OM content, mineral sediment content, OM volume percent, and mineral sediment volume percent. Differences among marshes in biomass categories including aboveground (live, dead, total, stem density), and belowground (live, dead, macro-organic matter, and total) were tested using ANOVA. Regression and correlation analyses were conducted to examine relationships among soil and vegetation parameters and LC, RC, and SOC concentration, density and accumulation rates (i.e. Figure 1.4). All statistical analyses were conducted using JMP ® version 14.2, SAS institute Inc. Figures were created using Sigma Plot © 13.0.

## 1.3. Results

### 1.3.1. Carbon concentrations, densities and accumulation rates

SOC concentration averaged two times higher in freshwater marshes compared to brackish and salt marshes ( $F_{2,193} = 18.63$ ;  $p < 0.01$ ; Table 1.1). There was a strong linear relationship between RC and SOC concentration ( $R^2 = 0.98$ ) and LC and SOC concentration ( $R^2 = 0.93$ ; Figure 1.4), and therefore, both RC and LC concentration were about two times higher in freshwater marshes than in brackish and salt marshes. The LC: RC concentration ratio was higher in freshwater marshes ( $0.39 \pm 0.01$ ) compared to brackish ( $0.34 \pm 0.01$ ) and salt marshes ( $0.38 \pm 0.02$ ;  $F_{2, 193} = 3.52$ ;  $p = 0.03$ ), indicating that LC comprised a slightly larger fraction of SOC in the freshwater marshes. RC is always the dominant form of SOC across all marshes and depth profiles illustrate that RC concentrations generally mirror the SOC concentrations (Figure 1.5). LC makes up a smaller fraction of SOC and depth profiles show less variability overall being relatively uniform with depth. Two freshwater marsh sites had relatively high SOC concentrations; FW1 averaged  $47.0 \pm 0.3\%$ , and did not vary with depth, and FW4 averaged  $29.6 \pm 1.5\%$  and was higher at the surface (34%) and bottom of the core (47.7%; Figure 1.5). The other two freshwater marsh sites had lower SOC concentrations both averaging  $11 \pm 1\%$  and were generally higher at the surface. In brackish marshes, SOC concentration profiles were highly variable with no consistent patterns with depth. In salt marshes, SOC generally increased with depth at all sites with one exception, SM11, where SOC concentration was higher at the surface (25%) than subsurface depths (1.8 – 10%).



Table 1.1. Soil properties averaged to 35 cm depth, and total soil organic carbon (SOC), recalcitrant carbon (RC) and labile carbon (LC) accumulation rates based on  $^{137}\text{Cs}$ -dating in freshwater (n = 4), brackish (n = 3) and salt (n = 4) marshes in Barataria Bay, LA. Values are means  $\pm$  standard error. Significant differences were calculated by one-way ANOVA for soil parameters, accretion rates and carbon concentration, density, and accumulation rates. Values represented by different letters are significantly different from Tukey's honestly significant post-hoc test.

Parameter	Fresh	Brackish	Salt	F value	p value
Bulk density (g cm <sup>-3</sup> )	0.20 $\pm$ 0.02 <sup>B</sup>	0.27 $\pm$ 0.03 <sup>A</sup>	0.25 $\pm$ 0.02 <sup>A</sup>	6.97	<0.01
OM content (%)	52.70 $\pm$ 0.04 <sup>A</sup>	33.30 $\pm$ 0.02 <sup>B</sup>	26.50 $\pm$ 0.01 <sup>B</sup>	17.98	<0.01
Mineral content (%)	47.30 $\pm$ 0.04 <sup>B</sup>	66.70 $\pm$ 0.02 <sup>A</sup>	73.50 $\pm$ 0.01 <sup>A</sup>	20.66	<0.01
OM volume (%)	5.85 $\pm$ 0.21 <sup>A</sup>	5.37 $\pm$ 0.09 <sup>AB</sup>	4.87 $\pm$ 0.16 <sup>B</sup>	5.95	<0.01
Mineral volume (%)	5.34 $\pm$ 0.70 <sup>B</sup>	7.90 $\pm$ 1.20 <sup>A</sup>	7.39 $\pm$ 0.58 <sup>A</sup>	10.67	<0.01
<b>Carbon concentration (%)</b>					
SOC	25.72 $\pm$ 1.94 <sup>A</sup>	14.46 $\pm$ 1.00 <sup>B</sup>	11.93 $\pm$ 0.65 <sup>B</sup>	18.63	<0.01
RC	18.04 $\pm$ 1.32 <sup>A</sup>	10.68 $\pm$ 0.71 <sup>B</sup>	8.53 $\pm$ 0.46 <sup>B</sup>	18.46	<0.01
LC	7.65 $\pm$ 0.62 <sup>A</sup>	3.79 $\pm$ 0.31 <sup>B</sup>	3.28 $\pm$ 0.23 <sup>B</sup>	17.59	<0.01
<b>Carbon density (mg cm<sup>3</sup>)</b>					
SOC	29.87 $\pm$ 1.15 <sup>A</sup>	26.04 $\pm$ 0.73 <sup>B</sup>	24.63 $\pm$ 0.97 <sup>B</sup>	8.24	<0.01
RC	21.47 $\pm$ 0.82 <sup>A</sup>	19.51 $\pm$ 0.52 <sup>AB</sup>	17.78 $\pm$ 0.74 <sup>B</sup>	6.74	<0.01
LC	8.39 $\pm$ 0.39 <sup>A</sup>	6.53 $\pm$ 0.33 <sup>B</sup>	6.60 $\pm$ 0.36 <sup>B</sup>	9.08	<0.01
<b>Accretion rate (cm yr<sup>-1</sup>)</b>	0.51 $\pm$ 0.10	0.49 $\pm$ 0.10	0.58 $\pm$ 0.05	0.3	0.75
<b>Carbon accumulation rate (g m<sup>-2</sup> yr<sup>-1</sup>)</b>					
SOC	123.6 $\pm$ 19.2	126.4 $\pm$ 27.9	132.1 $\pm$ 23.0	0.04	0.96
RC	87.7 $\pm$ 13.1	93.4 $\pm$ 18.8	98.1 $\pm$ 15.6	0.12	0.89
LC	35.8 $\pm$ 6.4	33.1 $\pm$ 9.3	34.9 $\pm$ 6.8	0.03	0.97

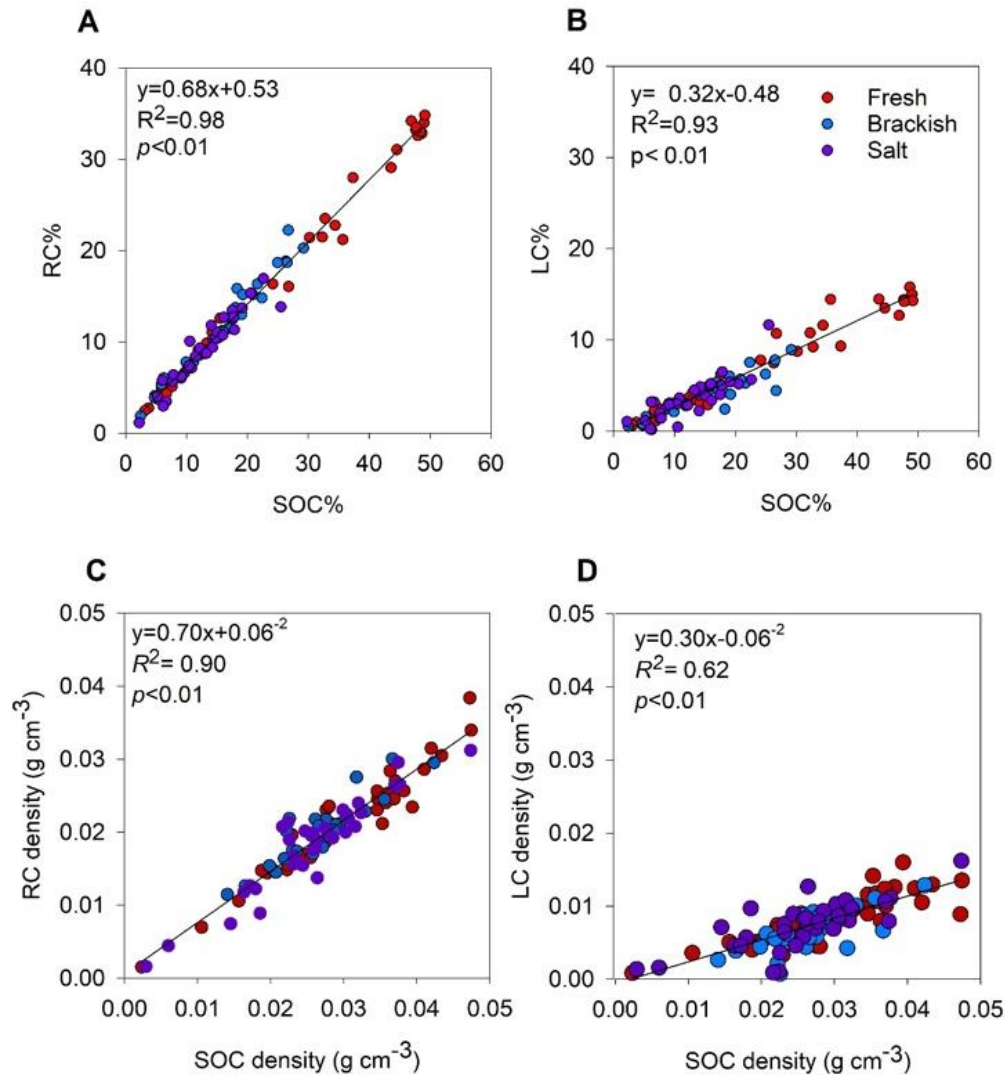


Figure 1.4. Relationships between the percentage (A and B) and density (C and D) of total soil organic carbon (SOC) and labile (LC) and recalcitrant (RC) carbon in freshwater, brackish, and marshes of Barataria Bay, Louisiana.

SOC, RC, and LC density were also highest in the freshwater marsh compared to the brackish and salt marshes ( $p < 0.01$ ; Table 1.1). SOC and RC density were about 15% lower in both the brackish and salt marsh compared to the freshwater marsh, while LC density was 33% lower in the brackish and saltmarsh compared to the freshwater marsh. All sites, except BM7, had low SOC density at the surface (0 - 2 cm), which then peaked between 10 and 20 cm depths (Figure 1.6). BM7 varied with depth without a

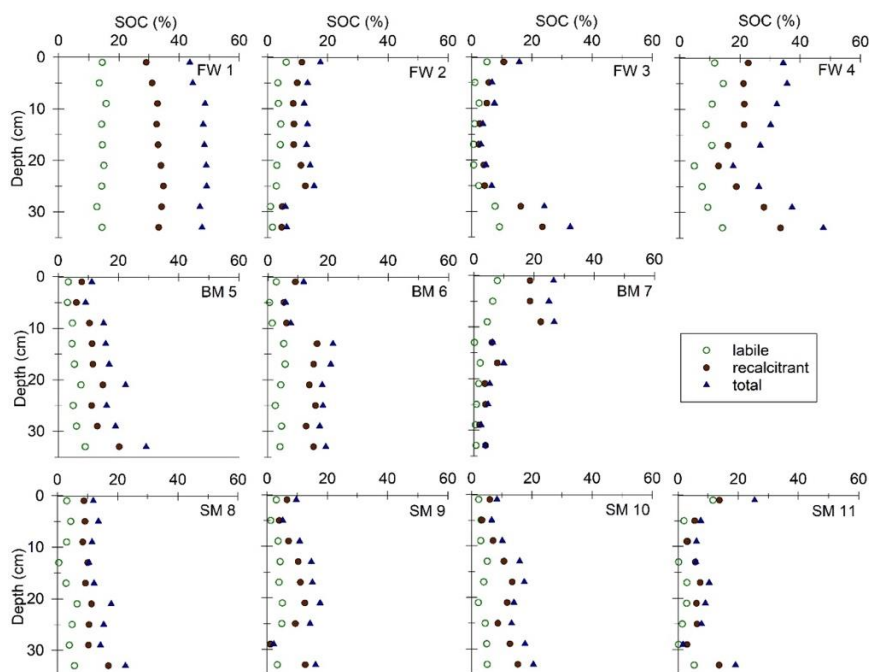


Figure 1.5. Total, refractory, and labile organic carbon concentration (%) profiles in marshes of Barataria Bay, Louisiana. The top row represents data from freshwater marshes ( $n = 4$ ), middle row, brackish marshes ( $n = 3$ ) and bottom row are from salt marshes ( $n = 4$ ).

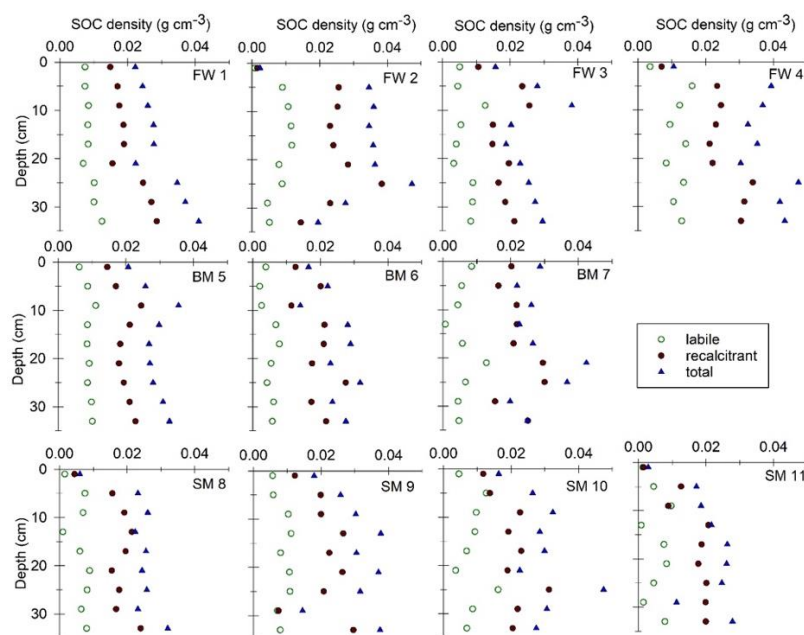


Figure 1.6. Total, refractory, and labile organic carbon density profiles in marshes of Barataria Bay, Louisiana. The top row represents data from freshwater marshes ( $n = 4$ ), middle row, brackish marshes ( $n = 3$ ) and bottom row are from salt marshes ( $n = 4$ ).

particular trend. Due to the correlation between RC density and SOC density ( $R^2 = 0.90$ ,  $p < 0.01$ ; Figure 1.4), the RC density profiles for all freshwater marshes strongly emulate the SOC density. However, the brackish and salt marshes were more variable, without showing a consistent trend, from RC being as low as 47% of SOC in SM11 to 97% of SOC in BM7. LC density was relatively low and less variable with depth (Figure 1.6).

SOC accumulation rate averaged  $128 \pm 12 \text{ g m}^{-2} \text{ yr}^{-1}$  with no significant difference between marsh types ( $F_{2, 10} = 0.04$ ;  $p > 0.05$ ; Table 1.1). Overall, RC accumulation averaged  $73.45 \pm 0.01\%$  of the total organic carbon accumulation rate. RC accumulation rate was similar among wetland types averaging  $93 \pm 8 \text{ g m}^{-2} \text{ yr}^{-1}$  ( $F_{2, 10} = 0.12$ ;  $p > 0.05$ ). Similarly, there was no significant difference between wetland type for LC accumulation rates, which averaged  $34 \pm 4 \text{ g m}^{-2} \text{ yr}^{-1}$  ( $F_{2, 10} = 0.03$ ;  $p > 0.05$ ).

### 1.3.2. Organic carbon sources

Freshwater marshes were dominated by  $C_3$  plants, including *Panicum hemitomon*, *Sagittaria lancifolia*, and *Alternanthera philoxeroides* that had at least 30% cover in one or more of the sites. Soil  $\delta^{13}\text{C}$  values were between  $-26.76$  and  $-30.42\text{‰}$ , which is similar to the  $\delta^{13}\text{C}$  plant tissue values of species present ranging between  $-25.2$  and  $-29.3 \text{‰}$  (Figure 1.7; Chmura and Aharon 1995). Brackish marshes were mostly composed of  $C_4$  plants, with the exception of one site that had *Schoenoplectus americanus* and *Juncus roemerianus*, accounting for 45% cover combined, and two other sites that had  $< 8\%$  *S. americanus*. The isotope mixing model predicted soil  $\delta^{13}\text{C}$  values between  $-11$  and  $-20\text{‰}$  (Table 1.2) in the brackish marsh, however soil  $\delta^{13}\text{C}$  was generally more depleted ranging between  $-17$  and  $-27\text{‰}$ . The salt marsh also had a mix

of C<sub>3</sub> and C<sub>4</sub> species. *Juncus roemerianus*, a C<sub>3</sub> species, was present in 3 out of 4 sites and accounted for 41 to 48% cover at these locations. In the salt marsh, the isotope mixing model predicted values between -12 and -22‰, which was similar to the actual values measured between -16 and -24‰.

Table 1.2. Predicted values of  $\delta^{13}\text{C}$  calculated from percent cover, and measured values of  $\delta^{13}\text{C}$  from soils depths down to 35 cm, in brackish and salt marshes only in Barataria Bay, Louisiana.

Wetland Type	Site ID	Predicted $\delta^{13}\text{C}$ (‰)		Measured $\delta^{13}\text{C}$ (‰)	
		Max	Minimum	Max	Minimum
Brackish	BM 5	-18.3	-20.4	-17.8	-27.1
	BM 6	-11.4	-13.4	-17.6	-23.2
	BM 7	-13.8	-16.0	-19.3	-26.7
Salt	SM 8	-12.1	-15.5	-16.0	-20.3
	SM 9	-18.9	-22.1	-17.4	-24.4
	SM 10	-17.1	-20.4	-18.8	-22.9
	SM 11	-16.6	-19.8	-15.7	-24.0

Depth profiles of  $\delta^{13}\text{C}$  show greater variability in brackish and salt marshes than the freshwater marshes (Figure 1.7). The recalcitrant sample was always more depleted than the bulk sample suggesting that the accumulation of lignin depleted the  $\delta^{13}\text{C}$  values (Benner et al. 1987). Depletion of the recalcitrant sample relative to the bulk sample was greater in the salt marsh than freshwater marsh by 0.6‰ ( $F_{2, 89} = 3.31$ ;  $p < 0.05$ ). My hypothesis that greater change from bulk to recalcitrant soil in  $\delta^{13}\text{C}$  may indicate a higher LC content is not supported by the data because the freshwater marsh had a higher LC: RC ratio than the salt marsh.

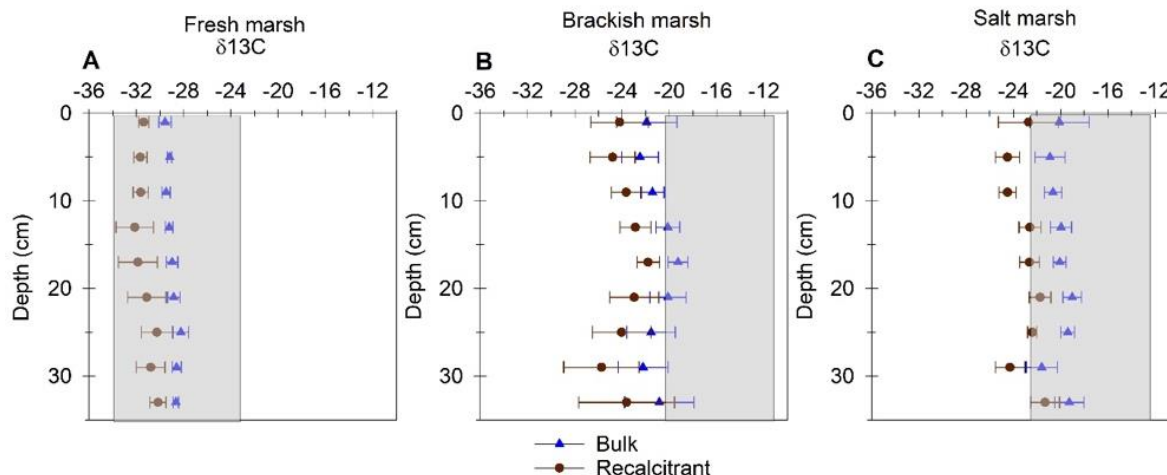


Figure 1.7.  $\delta^{13}\text{C}$  for the bulk (pre-hydrolysis) and recalcitrant (post-hydrolysis) samples for (a) freshwater ( $n = 4$ ), (b) brackish ( $n = 3$ ) and (c) salt ( $n = 4$ ) marshes in Barataria Bay, Louisiana. Region in gray for the freshwater marsh is the range of expected C3 values. The gray region in the brackish and salt marsh is the predicted range based on the isotope mixing model used by Chmura (1987).

### 1.3.3. Relationships between carbon and environmental parameters

#### *Aboveground biomass*

Live and total aboveground biomass and stem density varied significantly among marsh types, but dead aboveground did not vary by marsh type (Table 1.3). Live aboveground biomass was highest in the salt marshes, two times higher than the freshwater marshes and about 10% higher than the brackish marshes ( $F_{2, 32} = 4.28$ ;  $p < 0.05$ ). Stem density doubled from the freshwater to the salt marshes and doubled again between the salt marshes and the brackish marshes ( $F_{2, 26} = 6.57$ ;  $p < 0.01$ ). There were no significant relationships between aboveground vegetation parameters and SOC, RC, and LC concentrations or stocks.

Table 1.3. Above- and belowground biomass in fresh, brackish, and salt marshes in Barataria Bay, Louisiana measured in fall of 2016, ( $n = 3$ ,  $\pm$  standard error). Belowground biomass was the sum to 32 cm. Different letters represent values of individual plant parts that are significantly different ( $p < 0.05$ ) based on one-way ANOVA, and Tukey's honestly significant post-hoc test.

	Fresh	Brackish	Salt	F value	p value
Stem density (# m <sup>-2</sup> )	113 $\pm$ 17 <sup>B</sup>	554 $\pm$ 127 <sup>A</sup>	296 $\pm$ 51 <sup>AB</sup>	6.57	<0.01
<b>Aboveground biomass (g m<sup>-2</sup>)</b>					
Live	849 $\pm$ 103 <sup>B</sup>	1330 $\pm$ 199 <sup>AB</sup>	1473 $\pm$ 186 <sup>A</sup>	4.28	0.02
Dead	151 $\pm$ 22	401 $\pm$ 161	126 $\pm$ 51	2.05	0.14
Total	1000 $\pm$ 112 <sup>B</sup>	1731 $\pm$ 253 <sup>A</sup>	1600 $\pm$ 188 <sup>A</sup>	5.09	0.01
<b>Belowground biomass (g m<sup>-2</sup>)</b>					
Live	730 $\pm$ 460	1056 $\pm$ 1018	1713 $\pm$ 242	0.75	0.5
Dead	1390 $\pm$ 361	2726 $\pm$ 787	3424 $\pm$ 1149	1.22	0.34
Macro-OM	1106 $\pm$ 281 <sup>B</sup>	2502 $\pm$ 414 <sup>A</sup>	1653 $\pm$ 245 <sup>AB</sup>	4.67	0.05
Total	3227 $\pm$ 383	6285 $\pm$ 1234	6791 $\pm$ 1359	2.6	0.14

#### *Belowground biomass*

Total, live and dead biomass were approximately 50% lower in freshwater marshes, compared to brackish and salt marshes, however no significant differences occurred due to high variability ( $p > 0.05$ ; table 1.3; Figure A.2). Depth profiles of all biomass types (live, dead, and macro-organic matter) were similar between marsh types (i.e., no interaction between marsh type and depth;  $p > 0.05$ ). Macro-organic matter was highest in the brackish marshes and about 50% lower in the freshwater marshes ( $F_{2, 10} = 4.67$ ;  $p = 0.05$ ).

There were only a few significant relationships between belowground biomass (live, dead, macro-organic matter and total) and carbon concentration (%) (Table 1.4;

Figure 1.8). Dead biomass explained about 50% of the variation in soil carbon (SOC, RC, and LC) concentration in the freshwater marsh, but there was no relationship in the brackish or salt marshes ( $p > 0.05$ ). Macro-organic matter had a strong positive relationship with soil carbon (SOC, RC and LC) in the brackish marsh ( $p < 0.01$ ), but not in the freshwater or salt marshes ( $p > 0.05$ ). Similarly, total belowground biomass had a significant positive relationship to soil carbon concentration (SOC, RC, and LC) only in the brackish marsh ( $p < 0.05$ ).

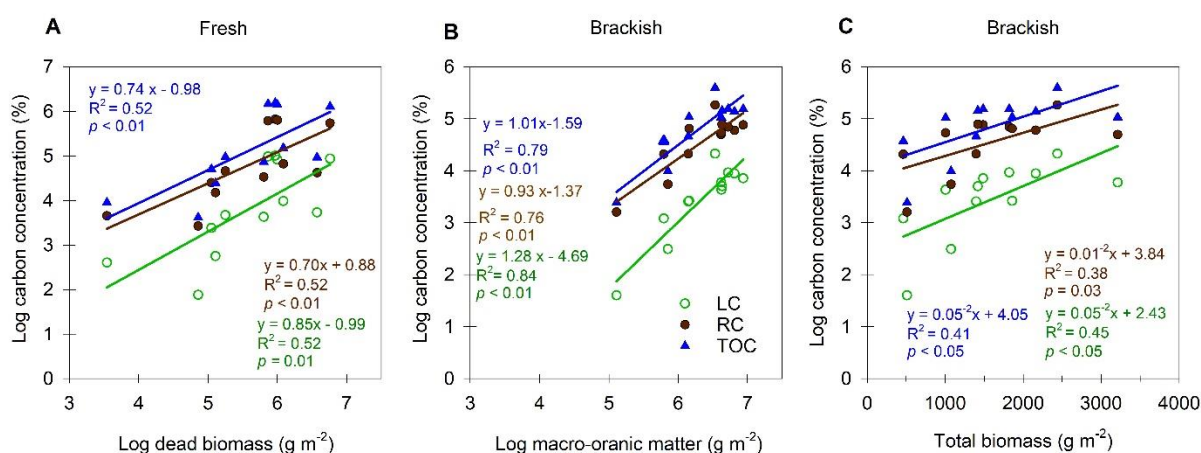


Figure 1.8. Significant relationships between belowground biomass stock ( $\text{g m}^{-2}$ ) and TOC, RC, and LC concentration in freshwater (A), brackish (B and C) marshes in Barataria Bay, LA. Carbon concentration and dead and macro organic matter biomass have been plotted with log transformed data.

Relationships between belowground biomass and SOC stocks ( $\text{g m}^{-2}$ ) varied from that of SOC concentrations (Table 1.4; Figure 1.9). In the freshwater marsh, RC and SOC stocks were negatively related to macro-organic matter ( $p = 0.01$ , and  $p = 0.02$ , respectively). The opposite pattern occurred across salt marsh sites, where LC ( $p = 0.02$ ); RC ( $p = 0.02$ ); and SOC ( $p = 0.02$ ) stocks were positively related to macro-organic matter. In the brackish marsh, LC stock was positively related to live and total belowground biomass ( $R^2 = 0.55$ ;  $p < 0.01$ ;  $R^2 = 0.56$ ;  $p < 0.01$ , respectively).



Table 1.4. Statistical report from regression analyses testing the relationships between TOC, RC, and LC concentrations (%) and SOC stocks (g m<sup>-2</sup>) and belowground biomass (g m<sup>-2</sup>) in fresh, brackish and salt marshes in Barataria Bay, Louisiana.

		Carbon fraction		Live		Dead		Macro-OM		Total	
Carbon concentration (%)		R <sup>2</sup>	p value	R <sup>2</sup>	p value	R <sup>2</sup>	p value	R <sup>2</sup>	p value	R <sup>2</sup>	p value
Freshwater	TOC		<i>n.s.</i>	0.52	<0.01		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
	RC		<i>n.s.</i>	0.52	<0.01		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
	LC		<i>n.s.</i>	0.51	<0.01		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
Brackish	TOC		<i>n.s.</i>		<i>n.s.</i>	0.79	<0.01	0.41	0.02		
	RC		<i>n.s.</i>		<i>n.s.</i>	0.76	<0.01	0.38	0.03		
	LC		<i>n.s.</i>		<i>n.s.</i>	0.84	<0.01	0.46	0.02		
Salt	TOC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
	RC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
	LC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>
<b>Carbon stock (g m<sup>-2</sup>)</b>											
Freshwater	TOC		<i>n.s.</i>		<i>n.s.</i>	0.42	0.02		<i>n.s.</i>		
	RC		<i>n.s.</i>		<i>n.s.</i>	0.50	0.01		<i>n.s.</i>		
	LC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		
Brackish	TOC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		
	RC		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		<i>n.s.</i>		
	LC	0.55	<0.01		<i>n.s.</i>		<i>n.s.</i>	0.56	<0.01		
Salt	TOC		<i>n.s.</i>		<i>n.s.</i>	0.33	0.02		<i>n.s.</i>		
	RC		<i>n.s.</i>		<i>n.s.</i>	0.31	0.02		<i>n.s.</i>		
	LC		<i>n.s.</i>		<i>n.s.</i>	0.31	0.02		<i>n.s.</i>		

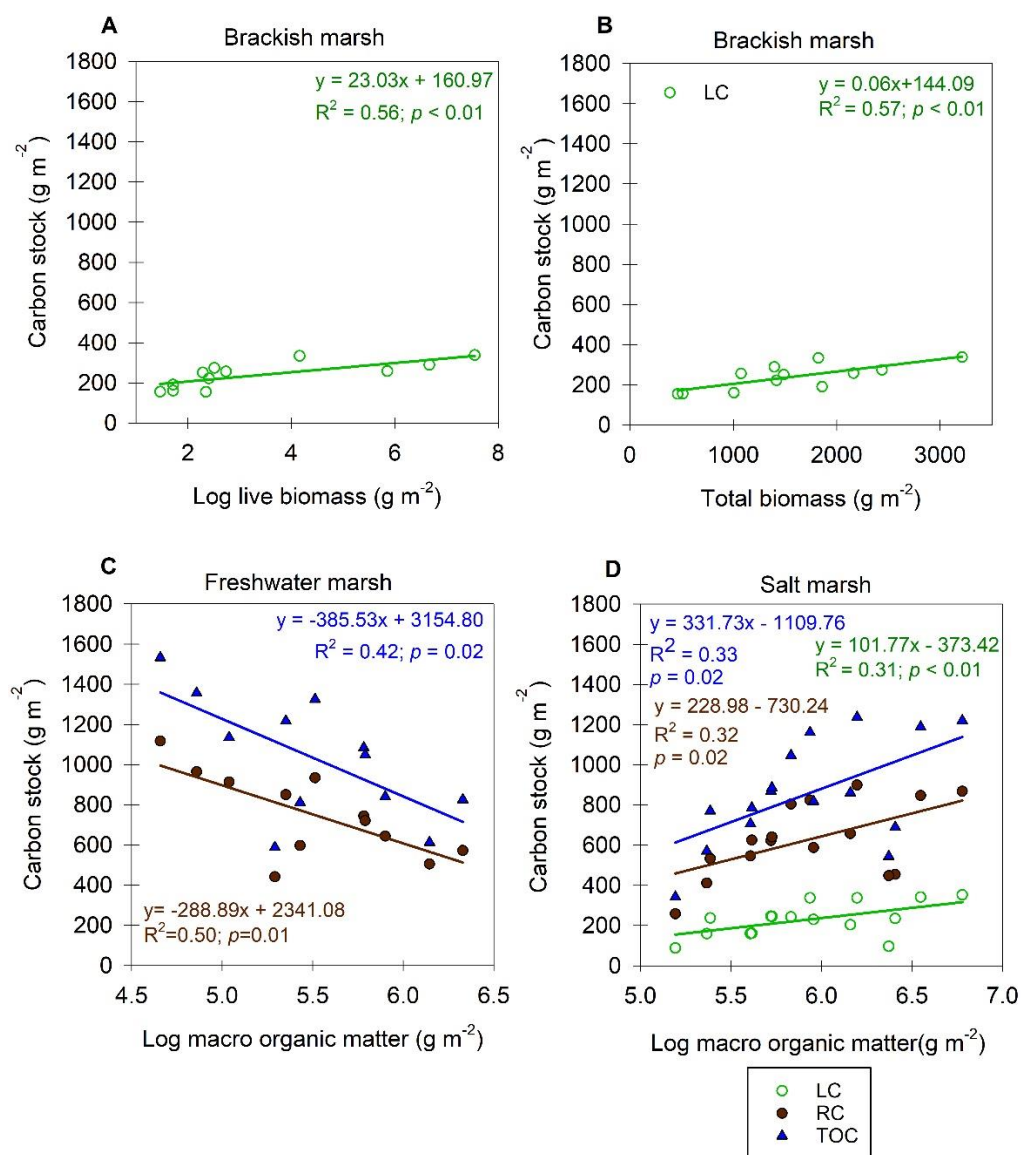


Figure 1.9. Significant relationships between belowground biomass stocks ( $\text{g m}^{-2}$ ) for 8 cm sections to 32cm against carbon stock ( $\text{g m}^{-2}$ ) summed to the same depth intervals, by wetland type in Barataria Bay, LA. Top row shows brackish marshes for (a) log live biomass against LC stock (b) total biomass against LC stock. The bottom row shows log transformed data of macro-organic matter in (c) the freshwater marsh for RC and total carbon stock, and (d) the salt marsh for all carbon stocks.

### Mineral sediment

Relationships between mineral density and carbon concentration and density differed depending on the carbon fraction (SOC, RC or LC), and wetland type. There was a strong negative relationship between mineral sediment concentration and SOC

( $R^2 = 0.90$ ;  $p < 0.01$ ), RC ( $R^2 = 0.83$ ;  $p < 0.01$ ), and LC ( $R^2 = 0.84$ ;  $p < 0.01$ )

concentration. There was also a highly variable negative relationship between mineral sediment density and LC density across freshwater marshes ( $R^2 = 0.07$ ;  $p < 0.05$ ) and brackish marsh ( $R^2 = 0.11$ ;  $p < 0.05$ ), but not the salt marshes ( $p > 0.05$ ). Mineral sediment accumulation rates were similar across wetland types ( $p = 0.17$ ) largely due to the high mineral density in the area of the Davis Pond outfall (FW3). If this location is excluded as relatively unrepresentative of freshwater marshes in the region, salt marshes had higher rates of mineral sediment accumulation than freshwater marshes ( $F_{2,9} = 7.01$ ;  $p < 0.02$ ). There was no relationship between mineral sediment stock and organic carbon (SOC, RC, and LC) accumulation rates. Interestingly, the site at the Davis Pond Diversion had mineral stocks about three times higher than the other freshwater marshes but did not have significantly different rates of LC, RC or SOC accumulation.

## 1.4. Discussion

### 1.4.1. Comparison of wetland types and SOC patterns

Studies have examined SOC concentration and accumulation rates in marshes across salinity gradients to evaluate their relative role in sequestering carbon and understand the different processes along the estuary influencing organic carbon inputs, loss rates, and preservation mechanisms. Additionally, sea-level rise, subsidence, storms, hurricanes and human disturbance can cause salinity increases and wetland loss (Turner 1997, Schoolmaster et al. 2018) affecting the capacity of these marshes to sequester carbon and function as carbon sinks. In U.S. estuaries, SOC concentrations average two times higher in the freshwater marshes than the salt marshes, with the

brackish marshes in between or slightly higher than freshwater marshes (table 1.5). My values of SOC concentration across wetland types (11-25%) were very similar to those of other studies in the Gulf of Mexico (8-26%) (Smith et al. 1983, Craft 2007), but were two times higher compared to Georgia tidal marshes (4.6-10.8%) (Craft 2007, Loomis and Craft 2010). SOC density, which is a more robust measure of carbon quantity (Chmura et al. 2003), was higher in either the freshwater marshes or brackish marshes and generally more similar across the salinity gradient than SOC concentration in this study. Similarity of carbon density indicates that carbon content by volume of soil is similar, while carbon concentrations differ due to heavy fine grain mineral sediment dilutes the carbon concentration by mass of soil (Chmura et al. 2003). However, when comparing SOC density to other studies, SOC density in brackish marshes in the present study was 17 to 37% lower than in brackish marshes in the Gulf of Mexico from other studies (Smith et al. 1983, Craft 2007). Carbon concentrations between these studies and ours had been similar, indicating that locations in this study had lower bulk density from less mineral sediment compared to other locations. In contrast, our study had higher SOC concentrations than Georgia marshes, but the SOC densities were similar (Loomis and Craft 2010). The Georgia marshes were river-dominated with the potential for greater mineral sediment (Loomis and Craft 2010) that diluted their SOC concentration by mass, making densities similar to those in the present study.

Overall, SOC accumulation rates in brackish and salt marshes in this study were about 50% lower than the U.S. average, and along the Gulf Coast, and the freshwater marshes were about 20% lower (Table 1.5). Freshwater marshes in this study had very similar SOC accumulation rate ( $124 \text{ g m}^{-2} \text{ yr}^{-1}$ ), to freshwater marshes in Georgia ( $123$

Table 1.5. SOC concentrations, densities, and accumulation rates from freshwater, brackish and salt marshes within the United States.

Location	Carbon Measurement	Freshwater	Brackish	Salt	
Barataria Bay	Concentration (%)	25 ± 2	14 ± 1	12 ± 1	This Study
Barataria Bay		26 ± 1	24 ± 3	9 ± 2	Smith (1983)
Gulf Coast		22 ± 2	19 ± 2	4 ± 2	Craft (2007)
Georgia coast		11 ± 1	8 ± 1	6 ± 1	Loomis & Craft (2010)
SE Atlantic coast		13 ± 2	22 ± 4	7 ± 1	Craft (2007)
US Average		17 ± 2	20 ± 2	9 ± 1	Craft (2007)
Barataria Bay	Density (mg cm <sup>-3</sup> )	30 ± 1	27 ± 1	25 ± 1	This Study
Barataria Bay		26 ± 1	31 ± 1	24 ± 1	Smith (1983)
Gulf Coast		21 ± 1	41 ± 1	23 ± 16	Craft (2007)
Georgia coast		28 ± 1	26 ± 1	23 ± 1	Loomis & Craft (2010)
SE Atlantic coast		40 ± 1	55 ± 1	66 ± 1	Craft (2007)
US Average		36 ± 1	48 ± 1	38 ± 1	Craft (2007)
Barataria Bay	Accumulation rate (g m <sup>-2</sup> yr <sup>-1</sup> )	123 ± 27	126 ± 28	132 ± 23	This Study
Barataria Bay		224 ± 1	296 ± 3	183 ± 12	Smith (1983)
Gulf Coast		140 ± 30	290 ± 60	310 ± 90	Craft (2007)
Georgia coast		124 ± 10	93 ± 17	40 ± 7	Loomis & Craft (2010)
SE Atlantic coast		160 ± 30	230 ± 40	70 ± 20	Craft (2007)
US Average		140 ± 20	240 ± 30	190 ± 40	Craft (2007)

g m<sup>-2</sup> yr<sup>-1</sup>). However brackish marshes were about 1.3 times higher, and salt marshes were 3 times higher in this study. High relative sea level rise in our study (Penland and Ramsey 1990) may be the cause to higher accumulation rates (Mudd et al. 2009) in brackish and salt marshes compared to the Georgia marshes. Similar SOC accumulation rates across wetland types in Barataria Bay, in this study appear to be the

result of high carbon concentration in freshwater marshes but higher mineral sediment in brackish and salt marshes increasing accumulation rates. A review of SOC accumulation rates showed that in the Gulf of Mexico, SOC accumulation rates in freshwater marshes were about half of that in salt marshes despite similar SOC densities (Craft 2007), therefore lower SOC accumulation rate in freshwater marshes are associated with their lower accretion rates compared to salt marshes. In contrast, SOC accumulation rates in Georgia freshwater marshes were about three times higher than in salt marshes in the Ogeechee, Altamaha, and Satilla river systems (Loomis and Craft 2010) attributed to lower decomposition rates in freshwater as compared to brackish and salt marshes where sulfate can stimulate reduction (Craft 2007). Variability of SOC accumulation rates by region show the importance of differing carbon inputs and preservation mechanisms by site.

#### 1.4.2. Carbon stability across the salinity gradient

The LC fraction of SOC was low across all marshes similar to the fraction measured by Dodla et al. (2012) in Barataria Bay brackish and salt marshes (27.5%). However, the LC fraction was slightly, but significantly, higher in the freshwater marshes ( $27.8 \pm 0.6\%$ ) than brackish marshes ( $24.6 \pm 0.8\%$ ) in this study. Higher LC in freshwater marshes compared to brackish and salt marshes is likely due to a combination of factors. While higher decomposition rates can occur with higher salinities due to increased sulfate reduction (Chambers et al. 2013, Van de Broek et al. 2018), local vegetation in salt and brackish marshes tend to be of lower quality indicated by higher lignin: N ratios, and more resistant to decay (Odum 1988). Salt marshes therefore have less LC input and higher LC decay, therefore had lower LC: RC ratios

compared to freshwater marshes. LC and RC density were both higher in freshwater marshes than the brackish and salt since SOC was 10 to 20% higher in the freshwater marshes. Although marshes SOC density in the mid-Atlantic was higher (31 - 49 mg cm<sup>-3</sup>) than this study (24 - 29 mg cm<sup>-3</sup>), RC densities were similar ranging between 9 and 34 mg cm<sup>-3</sup> in the mid-Atlantic and 17 - 21 mg cm<sup>-3</sup> in this study. Since RC is the fraction that is believed to be stored for longer periods of time, it suggests that despite soils in Barataria Bay having lower SOC density, the potential for long term carbon storage is similar.

#### *Carbon sources*

SOC source reflected predicted  $\delta^{13}\text{C}$  values based on local vegetation in the freshwater and salt marshes but were more depleted than predicted at certain depths of the brackish marshes. Depletion of  $\delta^{13}\text{C}$  values in brackish and salt marshes has previously been observed by (DeLaune 1986, Chmura et al. 1987, Craft et al. 1988). Depletion of soil  $\delta^{13}\text{C}$  from vegetation values could be due to benthic algae being a significant component of the SOC. Algae  $\delta^{13}\text{C}$  range between -12 to -23‰ (Smith and Epstein 1971), and average -21‰ (Sullivan and Moncreiff 1990). In Barataria Bay, Chmura (1987) used the maximum production value of algae, with the most depleted value into their isotope mixing model, and it predicted a  $\delta^{13}\text{C}$  of -14.1‰, which is more enriched than any of their measured soil values (Chmura 1987), or in this study. Low ratios of algae to total production values in marshes around the Gulf of Mexico, compared to the east coast and west coast (Sullivan and Currin 2002) make algae less likely to be a significant contributor to soil SOC, especially since algae are not only labile, but exposed at the surface to consumers compared to the protection at depth in

marsh soil (Chmura et al. 1987). Allochthonous SOC deposited on the marsh may also contribute to  $\delta^{13}\text{C}$  depletion. However, Barataria Bay is hydrologically isolated due to the levees of the Mississippi and the carbon flow between the Gulf of Mexico and Barataria Basin is mainly from salt marshes of the lower basin (Feijtel 1985). It is hypothesized that allochthonous SOC in brackish and salt marshes originates from within the basin (Chmura 1987). Decomposition of OM results in an accumulation of lignin, which is more depleted in  $\delta^{13}\text{C}$  than the bulk plant tissue (Benner 1987). Soils, therefore, can reflect a more recalcitrant  $\delta^{13}\text{C}$  signature than the original plant material. This explains why the recalcitrant sample is more depleted than the original bulk sample, because it contains a higher fraction of lignin. Freshwater marsh soils had similar  $\delta^{13}\text{C}$  values to in-situ vegetation, potentially due to the stability of  $\delta^{13}\text{C}$  in  $\text{C}_3$  plants compared to  $\text{C}_4$  plants. For example, the ash and smoke of burned  $\text{C}_3$  and  $\text{C}_4$  grasses were depleted in  $\delta^{13}\text{C}$  only in  $\text{C}_4$  vegetation (Das et al. 2010). This further supports the observation by Craft (1988), that the soils of  $\text{C}_3$  *Juncus* did not show depletion of  $\delta^{13}\text{C}$  compared to plant material, while  $\text{C}_4$  species such as *S. alterniflora* or *S. patens* did, even though *Juncus* has a comparable quantity of lignin to the *Spartina* species. Brackish and salt marshes had variable  $\delta^{13}\text{C}$  signatures with depth. Previous studies have suggested that depletion of  $\delta^{13}\text{C}$  with depth in brackish and saline marshes may be the result of switching to a freshwater environment (Chmura 1990), however changes in  $\delta^{13}\text{C}$  may also indicate the invasion of *Juncus* during time periods (depths) with more depletion, and the area composed of species such as *S. patens* or *D. spicata* during periods of  $\delta^{13}\text{C}$  enrichment. When using  $\delta^{13}\text{C}$  to determine changes in salinity regime, pollen analysis could be used to verify results (Chmura 1990).



#### 1.4.3. Environmental factors influencing carbon storage

There were no significant relationships between any aboveground biomass and rates of SOC, RC or LC accumulation in marshes of Barataria Bay, Louisiana. This is in contrast to findings where stem density was an important factor influencing SOC accumulation in created brackish marshes of LA (Abbott 2017) and aboveground biomass has been shown to be positively related to rates of LC and SOC accumulation in salt marshes in the mid-Atlantic (Elsey-Quirk and Unger 2018).

Belowground biomass contributes directly to SOC pools and may be important in influencing the quantity and accumulation rate of SOC, particularly in high platform organic marshes (Elsey-Quirk and Unger 2018) that have less mineral sedimentation than ramped marshes (FitzGerald et al. 2008). In the present study, live biomass was related to LC stock in the brackish marsh. Interestingly, live biomass composed the smallest portion of the total belowground biomass in the brackish marsh, averaging about 17% of belowground biomass. This fraction of live biomass was less than the freshwater (22%) and salt (25%) marshes. The slopes of the relationship between LC stock and live and total biomass are relatively shallow, indicating that it takes a relatively large change in live biomass to increase the LC stock (e.g., 10 g of live biomass for every 1 g of LC stock per square meter). Relationships between dead biomass and macro-organic matter were inconsistent between marshes and within SOC type, showing the importance of environmental differences, and plant communities have different relationships. Unlike the study in Chinese mudflats, comparing belowground biomass of only *S. alterniflora* and SOC, RC and LC stock all had similar, positive and exponential relationships with belowground biomass (Gao et al. 2016). Freshwater

marshes exhibited a negative relationship between RC and SOC stocks and macro-organic matter. Higher macro-organic matter biomass may contribute to higher porosity and a lower C density and SOC stock. The salt marsh had a positive relationship with macro-organic matter and all carbon stocks, suggesting here that the macro-organic matter is higher in density, potentially associating with mineral sediment. Overall, the only relationship with total biomass and carbon stock, was the LC stock in the brackish marsh. This is in contrast to the finding in the Mid-Atlantic where RC density was strongly related to belowground biomass across all marshes (Elsey-Quirk and Unger 2018), suggesting that large biomass fractions do not contribute as much to carbon stocks in Barataria Bay as it did in the mid-Atlantic.

Previous research has demonstrated the importance of mineral sediment in preserving LC through sorption or aggregates, preventing decomposition in both terrestrial environments (Rovira and Vallejo 2003, Mikutta et al. 2006), and salt marshes (Unger et al. 2016, Van de Broek et al. 2018). However, marshes in this study had a negative relationship with mineral density and the fraction of LC to SOC, suggesting this is not a mechanism that promotes LC preservation in these study sites. Rather RC was associated with mineral sediment in freshwater and brackish marshes, indicating that the SOC associated with sediment is older, relatively degraded, and deposited in the marsh from elsewhere (Leorri et al. 2018), potentially from the Mississippi River especially in sites near the Davis Pond Diversion . There was no relationship between mineral stock and accumulation rates of LC, RC or SOC in any marsh type, which differs from findings in the mid-Atlantic salt marshes, where LC accumulation rates were positively related to mineral sedimentation rate (Unger, 2016).

## CHAPTER 2. SOURCES AND CHEMICAL STABILITY OF SOIL ORGANIC CARBON ACROSS A CHRONOSEQUENCE OF CREATED MARSHES IN THE CHENIER PLAIN, LOUISIANA

### 2.1. Introduction

Approximately half of global carbon burial in soil is suggested to occur in blue carbon ecosystems, including mangroves, seagrass beds, and coastal marshes (Paul et al. 1997, Mcleod et al. 2011), where high rates of organic matter (OM) input and slow rates of decomposition lead to efficient organic carbon storage (Reddy and DeLaune 2008). Unlike terrestrial soils that may become carbon-saturated over time (Stewart et al. 2007), coastal wetlands accrete OM and mineral sediment with sea level rise (Reed 1995), such that carbon storage can increase over time (Chmura et al. 2003). Additionally, organic carbon may be preserved for millennia in anaerobic marsh soils, compared to forest soils, where carbon turnover may be only decades or centuries (Orson et al. 1987, Nellemann et al. 2010).

Areal coverage of coastal marshes is declining rapidly, putting their many ecosystem services at risk of being lost (Mcleod et al. 2011). Approximately 50% of salt marsh area in the United States has been lost since the 1700s (Watzin and Gosselink 1992), and this loss continues at a rate of 1 - 2% per year globally (Duarte et al. 2008). Coastal Louisiana, which contains about 40% of the total coastal wetland area in the United States is experiencing 80% of the wetland loss (Williams 1995) due to a combination of anthropogenic and natural factors (Britsch and Dunbar 1993, Turner 1997). Thus, carbon sequestration capacity is also lost. Marsh creation has the potential to mitigate some of the lost carbon sequestration capacity (Craft et al. 1999, Craft et al. 2003, Burden et al. 2013), yet it is not well established how carbon stocks

develop over time in created marshes and whether the carbon accumulating is vulnerable to decomposition (i.e. chemically labile). The dominant sources of soil organic carbon (SOC) in created marshes may influence the relative chemical stability, which has been suggested to have a significant influence on long-term soil carbon in coastal marshes (Mudd et al. 2009).

Few studies have examined soil carbon storage and accumulation rates in created marshes in general. Previous studies have shown that that SOC accumulation rates are highly variable in created marshes (Table 2.1), but can be similar to natural reference marshes within a year (Craft et al. 2003). Factors that influence SOC, such as productivity and decomposition take between 5 and 15 years to develop in created

Table 2.1. Soil organic carbon accumulation rates (CAR) in created, naturally colonized marshes in the U.S.

<b>Site</b>	<b>Age</b>	<b>Created CAR (g m<sup>-2</sup> yr<sup>-1</sup>)</b>	<b>Natural CAR (g m<sup>-2</sup> yr<sup>-1</sup>)</b>	<b>Source</b>
Sabine NWR, LA	5	52.24 ± 12.25	101.27 ± 15.22	Abbott et al. (2017)
Sabine NWR, LA	8	66.67 ± 12.29	101.27 ± 15.22	Abbott et al. (2017)
Sabine NWR, LA	13	75.77 ± 5.62	101.27 ± 15.22	Abbott et al. (2017)
Sabine NWR, LA	20	52.01 ± 6.91	101.27 ± 15.22	Abbott et al. (2017)
Sabine NWR, LA	32	66.96 ± 6.17	101.27 ± 15.22	Abbott et al. (2017)
DOT, NC	1	99	30	Craft et al. (2003)
Consultant, NC	3	39	51	Craft et al. (2003)
Port, NC	8	27	28	Craft et al. (2003)
Swansboro, NC	11	18	115	Craft et al. (2003)
Snow's Cut, NC	28	39	77	Craft et al. (2003)
Sapelo Island, Georgia	42	80	71	Craft et al. (2001)

marshes (Proffitt and Young 1998, Craft et al. 2003). Despite these relatively short time periods, carbon stocks can take between 70 and 100 years to reach equivalency to natural reference marshes to 30 cm depth (Craft et al. 2003, Burden et al. 2013). Most of the carbon development will occur at the surface where OM will accumulate first, therefore, to understand how created marshes build carbon instead of examining stock to 30 cm, analyzing SOC density at different depths will be more informative showing environmental factors that increase SOC storage (Table 2.2). For example, in the Chesapeake Bay, comparing a 5 year old marsh, and a natural reference, SOC density was similar between 0 and 2 cm, but three times higher between 14 and 16 cm in the natural reference (Havens et al. 1995). Location within the tidal frame has also been

Table 2.2. Total soil organic carbon (SOC) concentration (%) and density ( $\text{mg cm}^{-3}$ ) in created and natural salt marshes in the U.S.

Location	Age (yr)	Sample depth	SOC % created	SOC % natural	SOC density ( $\text{mg cm}^{-3}$ ) created	SOC density ( $\text{mg cm}^{-3}$ ) natural	Source
Sarah's Creek, VA	5	0-2	-	-	10	14	Havens et al. (1995)
Sarah's Creek, VA	5	14-16	-	-	5	16	Havens et al. (1995)
Tollesbury Essex, UK- high marsh	15	0-2	$2.2 \pm 0.4$	$9.7 \pm 2.4$	22	31	Burden et al. (2013)
Tollesbury Essex, UK- low marsh	15	0-2	$2.4 \pm 0.7$	$2.2 \pm 0.3$	11	14	Burden et al. (2013)
Pine Knoll, NC	21	0-10	$1.0 \pm 0.6$	$1.4 \pm 0.2$	10	14	Craft et al. (1999)
Snow's Cut, NC	25	0-10	$4.0 \pm 0.9$	$9.5 \pm 0.9$	22	30	Craft et al. (1999)
Sapelo Island, Georgia	42	0-10	$4.7 \pm 0.2$	$3.8 \pm 0.4$	13	14	Craft et al. (2001)

shown to be an important factor in influencing SOC density, where surface sediments of the low marsh was similar to the natural reference marsh after 15 years, but the high marsh had lower SOC density than the natural reference marsh (Burden et al. 2013). High marsh platforms tend to be highly organic, due to less inundation and less mineral sediment, therefore natural marshes have higher carbon contents (FitzGerald et al. 2008) therefore it takes longer for high marsh platforms to develop the same organic matter content. Understanding how carbon, and other ecosystem properties develop in different marsh zonations, such as low and high marsh should be used to prioritize how created marshes are built to maximize value of created marshes.

SOC is dependent on rates of input and loss of carbon (DeBusk and Reddy 1998, Jobbágy and Jackson 2000, Lützow et al. 2006), which is influenced by carbon source, stability, and environmental factors. Organic carbon is produced as biomass during photosynthesis when plants and algae fix atmospheric carbon dioxide. Over time, tissues die and become fragmented and decompose, breaking down through a variety of biological, physical, and chemical transformation processes (Reddy and DeLaune 2008, Lehmann and Kleber 2015). The rate of decomposition is partly determined by the lability of the SOC, or, conversely, the chemical recalcitrance of the organic matter (OM), and physio-chemical protection from microbes (Rovira and Vallejo 2002). The labile organic carbon (LC) pool is relatively small and by definition has the fastest turnover time (Krull et al. 2003, McLauchlan and Hobbie 2004), with a mean residence time of a few years to several decades (Cheng et al. 2007). The recalcitrant organic carbon (RC) pool is larger and has a slower turnover time (Krull et al. 2003, McLauchlan and Hobbie 2004), with a mean residence time of hundreds to thousands

of years (Cheng et al. 2007). LC consists of molecules such as carbohydrates and polysaccharides, while RC tissues are composed of cellulose, lignin, humic acids, and phenolic compounds (Benner et al. 1987, Reddy and DeLaune 2008). However, recent advances in our understanding of OM decay processes have demonstrated that molecular structure and chemical recalcitrance alone does not control stability (Schmidt et al. 2011). Rather, environmental conditions and biological processes are of utmost importance. Conditions present in coastal wetlands, in particular, promote the accumulation, burial, and preservation of detritus in various stages of physical and chemical decay (Reddy and DeLaune 2008), potentially preserving LC at depth where anaerobic conditions and associations with mineral substrates may limit oxidation (Unger et al. 2016). The relative fraction of LC and RC is important because climate change may cause environmental conditions to change, and SOC may be released; the LC may be vulnerable to oxidation, while RC may be more resistant to decay.

Few studies have measured SOC stability in coastal marshes, despite its importance for long-term SOC storage (Mudd et al. 2009), and the relatively large variation among marshes (Unger et al. 2016). In the Mississippi River Delta, for example, the LC fraction of SOC averaged 27.5% in the brackish and salt marshes (Dodla et al. 2012). In mid-Atlantic, USA salt marshes, LC accounted for up to 80% of SOC (Unger et al. 2016). The variation in SOC, which ranged from 100 to 331 g m<sup>-2</sup> yr<sup>-1</sup>, was mostly driven by differences in LC accumulation, while RC accumulation rate was relatively constant averaging 78 ± 5 g m<sup>-2</sup> yr<sup>-1</sup> (Unger et al. 2016). LC accumulation rate was directly and positively related to the amount of mineral sediment in the soil. It was hypothesized that marshes with higher rates of mineral sedimentation had either higher

inputs of LC and/or more efficient preservation of LC over time (Unger et al. 2016). The size of the watershed has also been implicated in influencing the allochthonous inputs of SOC, a major factor influencing RC concentration. Marshes within larger watersheds tend to have a greater fraction of RC that is deposited within their marshes, compared to marshes within smaller watersheds have little deposition and most SOC is derived from in-situ production. (Leorri et al. 2018). Therefore, the fraction of labile and recalcitrant carbon in coastal marshes may depend on the major source of carbon, whether produced in-situ by algae or plants, or deposited from allochthonous watershed or marine sources.

In created marshes, the sources of SOC may change as the marsh develops and may influence its chemical stability. Young created marshes without dense vegetation may be dominated by edaphic algal carbon (Zedler 1980). Algae have a lower concentration of recalcitrant compounds compared to salt marsh vegetation with 7.1% cellulose, 16.3% hemicellulose, and 1.5% lignin (Ververis et al. 2007), compared to 30% cellulose, 41% hemicellulose, and 6.3% lignin in *Spartina alterniflora* – reclassified as *Sporobolus alterniflorus* (Benner et al. 1987). In addition, algae are produced at the surface, where exposure facilitates decomposition or consumption by larger fauna, leading to less preservation of this carbon source in the soil column (Chmura and Aharon 1995). Vegetation succession may influence relative SOC stability as species can differ in lignin content (Table 2.3; Odum 1988). In created marshes of Louisiana, it can only take 1-2 years for *S. alterniflora* to naturally colonize and 5 years to reach full coverage (Proffitt and Young 1998), thereafter increasing their soil carbon contribution. Depending on elevation in the tidal frame and drainage factors, other species may



become dominant over time such as *Spartina patens*, or *Distichlis spicata* at mid to high elevations (Edwards and Proffitt 2003).

$\delta^{13}\text{C}$  isotope values have been used to determine the dominant SOC sources in marsh soils. Differences in photosynthetic pathway between  $\text{C}_3$  and  $\text{C}_4$  plants result in a

Table 2.3. Lignin content of leaves and roots of *Spartina patens* and *Spartina alterniflora*

	Species	Litter Type	Lignin Content (%)	Live or dead	Study
Louisiana	<i>S. patens</i>	leaves	7.8 - 9.2	live	Stagg et al. (2018)
Maryland	<i>S. patens</i>	leaves	14.2	dead	Ball and Drake (1997)
Louisiana	<i>S. patens</i>	roots	14.8 - 18.6	live	Stagg et al. (2018)
Maryland	<i>S. patens</i>	roots	27.0 - 30.4	live	Saunders et al. (2006)
Louisiana	<i>S. alterniflora</i>	leaves	5.5 - 5.7	live	Stagg et al. (2018)
Georgia	<i>S. alterniflora</i>	leaves	15.1	live	Maccubbin & Hodson (1980)
Georgia	<i>S. alterniflora</i>	leaves	13.1	live	Hodson et al. (1984)
Georgia	<i>S. alterniflora</i>	leaves	14.4	dead	Hodson et al. (1984)
Massachusetts	<i>S. alterniflora</i>	leaves	11.7	unknown	Wilson et al. (1985)
Massachusetts	<i>S. alterniflora</i>	leaves	11.0 - 12.0	dead	Wilson et al. (1985)
Unknown	<i>S. alterniflora</i> (tall form)	leaves	4.3	unknown	Benner et al. (1987)
Unknown	<i>S. alterniflora</i> (short form)	leaves	6.1	unknown	Benner et al. (1987)
Louisiana	<i>S. alterniflora</i>	roots	13.7-15.2	live	Stagg et al. 2018
Unknown	<i>S. alterniflora</i> (short form)	roots	9.3	unknown	Benner et al. (1987)
Georgia	<i>S. alterniflora</i> (tall form)	rhizomes	12.2	unknown	Hodson et al. (1984)
Georgia	<i>S. alterniflora</i> (short form)	rhizomes	19.3	unknown	Hodson et al. (1984)

different range of  $\delta^{13}\text{C}$  values.  $\text{C}_3$  plants, which tend to occur in terrestrial or fresh water environments have  $\delta^{13}\text{C}$  values that range from -23 to -34‰, while  $\text{C}_4$  plants, which tend to dominate salt marshes have  $\delta^{13}\text{C}$  values that range from -6 to -19‰ (Smith and Epstein 1971, Chmura and Aharon 1995). Edaphic algae have  $\delta^{13}\text{C}$  values in between those of  $\text{C}_3$  and  $\text{C}_4$  plants averaging -21‰ (Sullivan and Moncreiff 1990). Edaphic algae production can be a significant proportion of carbon fixed, ranging from 8 to 33% of vascular plant production in the Atlantic and Gulf coasts (Sullivan and Moncreiff 1990), and as much as 140% in marshes in California with less vegetation and more sunlight (Zedler 1980).

As created marshes develop, both the source and stability of carbon may change over time. In Jiangsu Province, China, the  $\delta^{13}\text{C}$  values indicated that bare mudflats were dominated by algae while SOC in the *S. alterniflora* marsh was more derived from local vegetation (Gao et al. 2016). Additionally, LC, RC, and SOC were significantly higher in the *S. alterniflora* marsh than the bare mudflat. LC in the marsh was largely derived directly from *S. alterniflora* plants, while the RC resembled  $\text{C}_3$ , terrestrial plants (Gao et al. 2016), suggesting that the RC may have been allochthonous and deposited in the marsh, rather than produced in situ. After initial colonization, vegetation succession will alter carbon inputs due to changes in litter quality, which may alter the LC to RC ratio, and their respective accumulation rates.

The purpose of this study is to investigate sources of labile and refractory carbon accumulating in created marshes across a 32-year chronosequence, as compared to an adjacent natural marsh. Algae do well in high light conditions and respond quickly to environmental conditions (Zedler 1980), and therefore, are predicted to colonize created

marshes first. However, the potential for decomposition of surface algae is also high, since they lack lignin and other recalcitrant compounds (Ververis et al. 2007).

Vegetation colonization facilitates sediment deposition and accretion, which may promote greater burial of LC. Therefore, I hypothesize that the relative magnitude of LC and RC will increase over time, as vegetation establishes and i) contributes both labile and refractory biomass to the soil directly, and ii) indirectly traps organic particles and facilitates burial of carbon to anoxic depths prior to decomposition. It is also predicted that young marshes dominated by algal carbon will have a higher fraction of LC than RC, and older marshes that have more vegetation will have a higher RC proportion than LC (Figure 2.1). The dominant plant species may play a role in the relative magnitude of LC and RC. Elevation is also predicted to influence carbon accumulation through effects of flooding on productivity, decomposition, deposition and preservation of carbon. Specifically, LC accumulation rate is predicted to decrease with increasing marsh elevation due to less flooding and more exposure to oxygen, promoting decomposition and less burial by sediment.

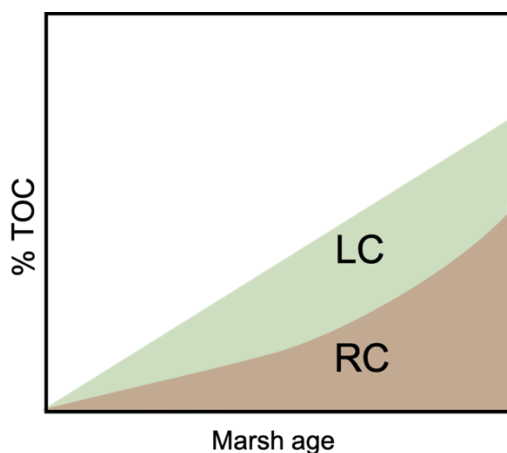


Figure 2.1, Conceptual model of hypothesized labile organic carbon (LC) and recalcitrant organic carbon (RC) of marsh derived (surface) carbon in created marshes.

## 2.2. Methods

### 2.2.1. Study area

A 32-year chronosequence of created brackish marshes was sampled in Sabine National Wildlife Refuge (SNWR), located in the Chenier Plain in southwest Louisiana. Many anthropogenic modifications have occurred in the region since the 1800s. The dredging and deepening of the Calcasieu Ship Channel has altered the natural hydrology and allowed salt water intrusion into previously fresh and intermediate marshes (DeLaune et al. 1983). Additionally, subsurface fluid withdrawal in the 1950's lead to rapid land-surface subsidence (Bernier et al. 2011), which combined with salt water intrusion has led to extensive marsh conversion to open water. In response, the Army Corps of Engineers and the National Wildlife Refuge system created marshes beginning in 1983 in the SNWR by pumping channel dredge material into open water areas bounded by low levees, which were eventually breached or removed to allow natural hydrology to return. Salt marsh vegetation naturally colonized the marshes (Edwards and Proffitt 2003). The focus of this study is six created salt marshes aged 0, 5, 8, 13, 20, and 32 years (representing a space-for-time substitution), and two adjacent natural reference marshes (Figure 2.2).

In a previous study, Abbott (2017) found that aboveground biomass was similar among these created marshes ( $p > 0.05$ ), but the natural and 13 year-old created marsh had higher stem densities associated with *D. spicata* and *S. patens*, than the 5-year-old marsh, which was dominated by *S. alterniflora*. Marsh age and environmental conditions influenced differences in plant community composition among marshes ( $p < 0.01$ ). Younger sites are dominated by *S. alterniflora* at all elevations, while older sites are

dominated by *S. alterniflora* at lower elevations and *D. spicata*, *Borrchia frutescens* and *Batis maritima* at higher elevations (Abbott 2017).

Soil parameters were also found to vary among marshes. The 20 and 32-year-old created marshes were adjacent to the Calcasieu Ship Channel and had higher bulk densities and lower OM content. The 0-year-old marsh had no vegetation to contribute OM, and therefore reflects soil properties of the dredge sediment. Carbon stock linearly increased through time in the created marshes, however carbon accumulation rates (CAR) were highly variable among created marshes (between 16 and 99 g C m<sup>-2</sup>yr<sup>-1</sup>) the only significant difference was between the 20 year old marsh and the natural reference marsh, the later was about two times higher. The most significant predictor variable of SOC accumulation rate was stem density, however it only explained 20% of variation (Abbott 2017).

#### 2.2.2. Field sampling

Six sites in each of the six created marshes and between the two natural marshes (n = 42) were randomly established for soil, vegetation, and environmental sampling (Figure 2.2; Abbott 2017). Although all marshes were relatively close together, spatial location did influence environmental characteristics, with the older marshes located adjacent to the Calcasieu Ship Channel being exposed and described as exterior, while the younger marshes (<13 years), and natural reference marshes were more protected and described as interior marshes.

For every site (n=42) one soil core was hand-extracted at each location using a 35 cm polycarbonate tube with a diameter of 6.35 cm with a sharpened bottom edge to minimize compaction (n = 6). Cores were transported back to the lab horizontally.

Elevation (NAVD88) of each core location was surveyed using Leica Real Time Kinematic GS14 with horizontal and vertical accuracies were 1 and 3 cm, respectively. In August 2016, aboveground biomass, stem density and shoot height measurements were made in each of the six plots in each marsh. Quadrat size was dependent on species, 0.01 m<sup>2</sup> was used for *S. patens* and *D. spicata* dominated plots, 0.1 m<sup>2</sup> for *S. alterniflora* and 0.25 m<sup>2</sup> for *Bolboschoenus robustus* dominated plots; all were normalized to 1.0 m<sup>2</sup> for calculations. Standing biomass was sorted into live and dead, and dried to a constant weight at 60°C to obtain aboveground biomass. Live shoot height of *S. patens*, *D. spicata*, *S. alterniflora* and *B. robustus* was measured. Differences in aboveground biomass was originally analyzed in Abbott (2017), and the influence of these characteristics on LC and RC accumulation rates was analyzed in this study.

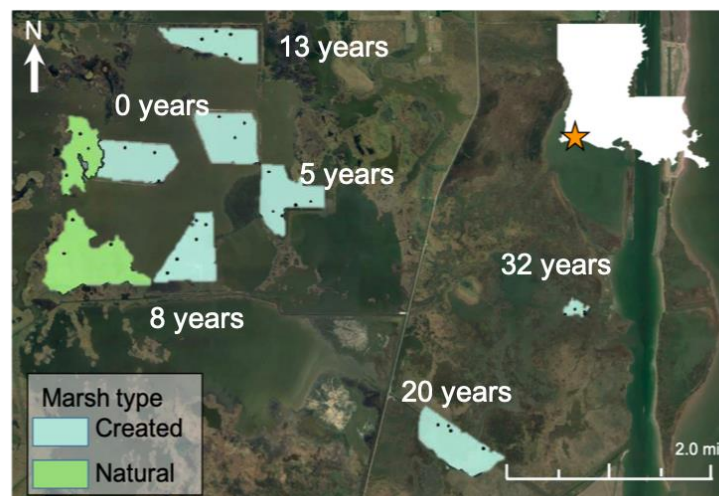


Figure 2.2. Map of created and natural marshes in the Sabine National Wildlife Refuge, Chenier Plain, Louisiana. Dots are core locations (n=6 per marsh age). The natural reference marshes and created marshes 13 years and younger are considered interior, and the 20 and 32-year-old marsh located closer to the ship channel are termed exterior.

### 2.2.3. Soil processing and analysis

Soil cores were sectioned into 2 cm depth intervals and dried to a constant weight at 60°C. Bulk density was calculated as the dry mass per unit volume. Sub-samples for total organic carbon (SOC) and acid hydrolysis were ground and homogenized using a Wiley™ soil mill. SOC content was determined by weighing 10 to 12 µg of each sample into silver foil capsules. To remove inorganic carbon, samples were fumigated either by adding 30µL of DI water and fuming with 12M HCl for 6 hours (Harris et al. 2001), or fumed with 12M HCl for 24 hours, then dried, packed in tin capsules and sealed. Samples were run on the Costech 1040 CHNOS Elemental Combustion System. SOC data were processed and presented in Abbott (2017).

Fractionation of LC and RC can be conducted through a variety of chemical, thermal, and biological processes (McLauchlan and Hobbie 2004, Dodla et al. 2012). The assumption is that a labile fraction of SOC susceptible to extraction is also susceptible to microbial degradation (McLauchlan and Hobbie 2004, Dodla et al. 2012). RC separated by acid hydrolysis, similar to the method employed in the present study, when dated using  $^{14}\text{C}$  was found to be an average of 1200 years older than the bulk SOC, providing support for the idea that the pool of carbon remaining following hydrolysis is older than the pool removed (Paul et al. 2006). A comparison of molecules before and after acid hydrolysis using solid-state  $^{13}\text{C}$  Nuclear Magnetic Resonance (NMR) spectroscopy provided evidence that labile compounds such as carbohydrates and amino acids were removed during the hydrolysis procedure, while chemically recalcitrant compounds remained after hydrolysis (Silveira et al. 2008).

Labile and refractory C was measured for every other 2 cm depth section starting at the surface (0 - 2 cm) using selected cores (Table A.2). Following Unger et al. (2016), 1.0 g of soil sample was placed into a glass vial, and 10 ml of deionized (DI) water and 20 ml of 6M HCL were added. Samples were then placed on a digestion block at 150 °C for 18 hours, followed by centrifugation at 2500 revolutions per minute for 10 minutes. The supernatant was immediately decanted, and the remaining recalcitrant material was then dried at 60°C until a constant weight was reached. This residual fraction was measured for SOC by weighing 10 µg of sample into silver capsules, fumigating with 12 M HCl for 24 hours (Hedges and Stern 1984), then analyzing using a Costech 1040 CHNOS Elemental Combustion System. The percent of SOC that was refractory was calculated as:

$$RC\% \text{ of SOC} = \frac{C_{\text{post hydrolysis}} (g)}{SOC \text{ original} (g)} = \frac{\text{Mass post hydrolysis} (g) * (\text{post hydrolysis } C\%) / 100}{\text{Mass pre hydrolysis} (g) * \left(\frac{TOC (\%)}{100}\right)} \quad (1)$$

Where the mass post hydrolysis is the mass measured after hydrolysis, and post hydrolysis C% is the elemental carbon concentration measured on the recalcitrant sample after hydrolysis. The mass pre hydrolysis was the exact amount of 1 g of sample measured, and TOC% is the elemental C percent measured on bulk sample before hydrolysis.

The percent of organic carbon that was labile was calculated as:

$$LC\% \text{ of SOC} = 100 - RC\% \text{ of SOC} \quad (2)$$

The percent of the total soil mass that was refractory carbon (RC %) was calculated as:

$$RC\% = SOC\% * \frac{RC\% \text{ of SOC}}{100} \quad (3)$$

Where SOC% is the elemental carbon percent of the bulk sample before hydrolysis.

The percent of the total soil mass that was labile carbon (LC %) was calculated as:



$$LC\% = SOC(\%) * \frac{LC\% \text{ of SOC}}{100} \quad (4)$$

LC, RC, and SOC accumulation rates in created marshes were calculated by summing the respective carbon density above the dredge layer and dividing by the known age of the marsh. In the natural marsh,  $^{210}\text{Pb}$ , and  $^{137}\text{Cs}$  did not result in reliable age estimates, and therefore, literature values for the region (Smith 2012) were used to calculate CAR and mineral accumulation rates. The average fraction of LC and RC of SOC from the natural marsh values was multiplied by each total SOC accumulation rate to obtain an approximate LC and RC accumulation rate. Loss on Ignition (LOI) was performed to determine organic matter and mineral sediment fractions by burning samples at 550 °C for 4 hours, and the percent weight lost is OM and the percent remaining mineral sediment (Abbott 2017). Mineral accumulation rate ( $\text{g m}^{-2} \text{yr}^{-1}$ ) was calculated as  $1 - \% \text{LOI}$  multiplied by the bulk density, accretion rate ( $\text{cm yr}^{-1}$ ) and 10,000 to convert from  $\text{cm}^2$  to  $\text{m}^2$ .

#### 2.2.4. Statistical Analysis

Data were transformed using logit, square root, cubic or quadratic root when necessary to meet assumptions of normality and homogeneity of variance. One-way Analysis of Variance (ANOVA) was used to determine the effects of marsh age on soil properties, and carbon types. Soil properties included bulk density, OM content (%), organic matter volume (%), and mineral sediment volume (%). Carbon types included SOC, RC and LC, concentration, density and accumulation rate. When a significant difference was found, Tukey's Honestly Significant Difference post-hoc test was used. Analysis of Covariance (ANCOVA) was used to determine the effect of marsh age and

elevation on SOC, RC and LC concentration, density, and accumulation rates. ANOVA was also used to test the effect of dominant vegetation within marshes on SOC, RC, and LC, concentration, density and accumulation rates. Because all vegetation types were not present in all marshes, it was not possible to test for interactions between dominant species and marsh age, only difference in carbon types within a marsh with multiple plant species. One-way ANOVA was used to determine the effect of marsh age on  $\delta^{13}\text{C}$  values at different depths. Depth categories were surface (0 - 4 cm), mid depth (4 - 8 cm), and > 8 cm, with the deepest core being the natural marsh at 30 cm. When the ANOVA showed significant differences Tukey's Honestly Significant Difference post-hoc test was used to show differences. Regression analyses were used to examine the relationships between SOC, LC, RC concentration and density between all response variables (LC, RC, and SOC concentration, density, and accumulation), and predictor variables (marsh elevation, mineral sediment, stem density, aboveground biomass).

## 2.3. Results

### 2.3.1. Carbon concentrations and densities

Across marshes, both RC and LC concentrations were positively related to SOC concentration (RC,  $R^2=0.94$ ; LC,  $R^2 = 0.82$ : Figure 2.3) with the RC fraction comprising  $69.3 \pm 1.3\%$  of SOC. The RC fraction was higher in the natural marsh than the 5, 20, and 32-year-old marshes ( $F_{6, 184} = 4.61$ ;  $p < 0.01$ ). SOC concentration increased with created marsh age in the interior marshes from  $1.3 \pm 0.1\%$  in the newly created 0-year-old marsh to  $6.6 \pm 1.5\%$  in 13-year-old marsh. Similarly, RC and LC concentrations increased from the 0- to the 13-year-old marsh. The 20 and 32-year-old, exterior marshes had lower SOC concentrations with  $2.6 \pm 0.3$  and  $3.4 \pm 0.3\%$ , respectively.

The RC concentration in the 20-year-old marsh was statistically lower than the 13-year-old marsh, but the LC concentration was not. The natural marsh had over two times the SOC concentration than the created marshes averaging  $16.7 \pm 0.8\%$  compared an average of  $3.9 \pm 0.4\%$  in created marshes ( $F_{6,6} = 62.3$ ;  $p < 0.01$ ; Table 2.4). The concentration of RC in the bulk soil was four times higher in the natural marsh ( $12.4 \pm 0.5\%$ ) than the created marshes averaging  $2.8 \pm 0.5\%$ . The LC concentration was also four times higher in the natural marsh ( $4.3 \pm 0.3\%$ ) compared to the created marshes ( $1.1 \pm 0.1\%$ ). SOC depth profiles in the created marshes exhibited a consistent trend with approximately twice the SOC concentration at the surface compared to directly above the dredge horizon (appendix, Figure A.3). RC comprised most of the SOC and therefore, had a similar depth profile, while the LC profile was slightly more uniform across all depths. In the natural marsh, SOC density generally increased with depth from  $16 \pm 3 \text{ mg cm}^{-3}$  at the surface to  $51 \pm 9 \text{ mg cm}^{-3}$  at 27 cm. Carbon density in created marshes, however, did not have a consistent trend with depth (see appendix; Figure A.4). RC density profiles mirrored the SOC density as RC density had a positive linear relationship with SOC density ( $R^2 = 0.95$ ; Figure 2.3). SOC density was three times higher in the natural marsh, averaging  $35 \pm 2 \text{ mg cm}^{-3}$  compared to the created marshes averaging  $13.7 \pm 0.5 \text{ mg cm}^{-3}$ . RC density was over two times higher in the natural marsh ( $27 \pm 2 \text{ mg cm}^{-3}$ ) than the created marshes. ( $10 \pm 4 \text{ mg cm}^{-3}$ ). LC density was also about two times higher in the natural reference marsh ( $9 \pm 1 \text{ mg cm}^{-3}$ ) compared to the created marshes ( $4.1 \pm 0.2 \text{ mg cm}^{-3}$ ). Unlike carbon concentrations, which tended to be higher the 13-year-old marsh, average SOC and RC density were

similar among created marshes (table 2.4). LC density, however, was 40% higher in the 20 and 32-year-old marshes than the 8-year-old marsh.

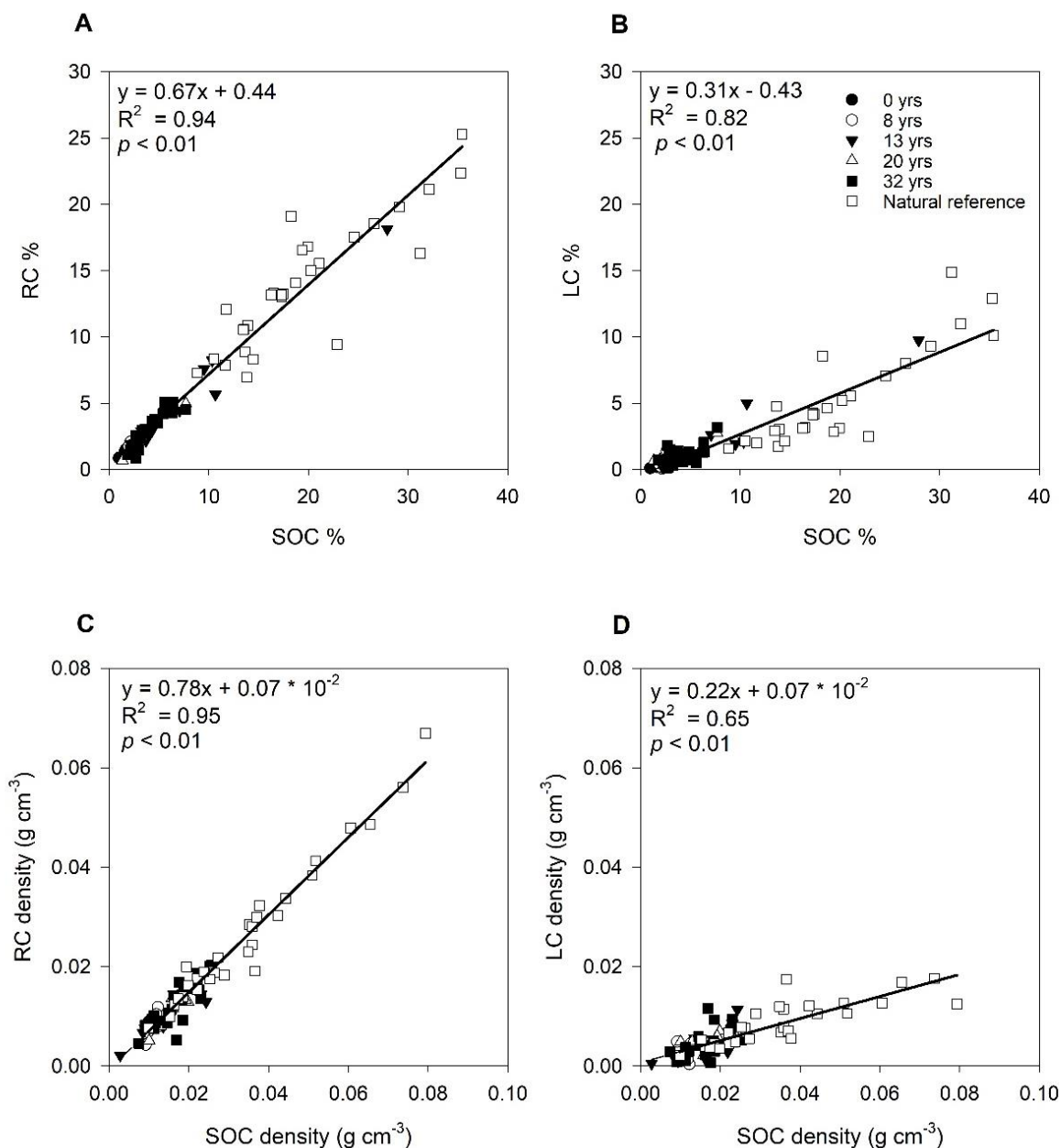


Figure 2.3 Relationships between the percentage (A and B) and density (C and D) of total soil organic carbon (SOC) and labile (LC) and recalcitrant (RC) carbon in freshwater, brackish, and marshes of Barataria Bay, Louisiana.

### 2.3.2. Organic carbon accumulation rates

SOC accumulation rates were not significantly different among created marshes of different ages and were generally similar to the natural reference marshes. One exception was the 20-year-old marsh, which had 50% lower SOC accumulation rates than the natural marsh ( $52 \pm 7 \text{ g m}^{-2} \text{ yr}^{-1}$  and  $101 \pm 15 \text{ g m}^{-2} \text{ yr}^{-1}$ , respectively;  $p = 0.03$ ). RC accumulation rate was also lower in the 5- and 20-year-old marshes than the natural marshes ( $p = 0.01$ ). LC accumulation rate averaged  $20 \pm 1 \text{ g m}^{-2} \text{ yr}^{-1}$  and did not significantly differ across marshes ( $p > 0.05$ ). In the lowest elevation 8-year-old marsh, RC and SOC accumulation rates were positively related to elevation (RC,  $R^2 = 0.80$ ;  $p = 0.01$ ; SOC,  $R^2 = 0.97$ ;  $p < 0.01$ ).

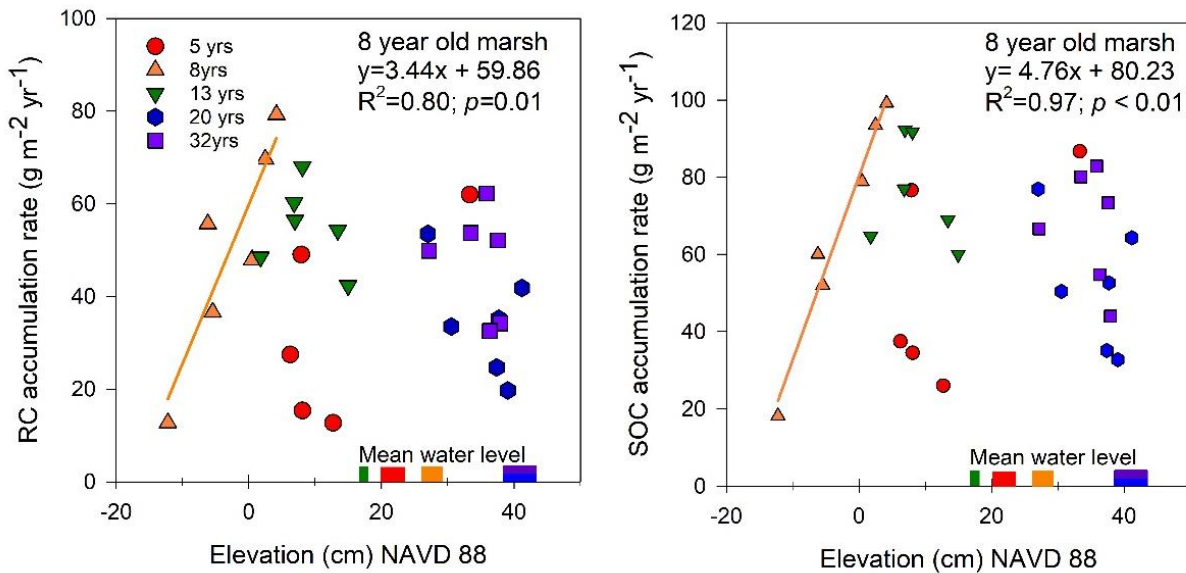


Figure 2.4. Relationship between marsh elevation cm in NAVD 88 and (a) RC accumulation rate and (b) SOC accumulation rate of created marshes in the Chenier Plain, Louisiana. The bars along the x axis represents the range between mean low and high water for each created marsh (Abbott 2017).

Table 2.4. Soil properties averaged above the dredge horizon layer in created marshes, the surface for the 0-year-old marsh and to 23 cm for natural marshes. Values are means  $\pm$  standard error (n = 6). Values represented by different letters are significantly different ( $p < 0.05$ ) based on one-way ANOVA and Tukey's post-hoc test.

	Created marsh age (years)								
Parameter	0	5	8	13	20	32	Natural Reference	F value	p value
Bulk density (g cm <sup>-3</sup> )	0.72 $\pm$ 0.11 <sup>ABC</sup>	0.55 $\pm$ 0.08 <sup>ABC</sup>	0.31 $\pm$ 0.05 <sup>CD</sup>	0.34 $\pm$ 0.04 <sup>CD</sup>	0.65 $\pm$ 0.04 <sup>A</sup>	0.50 $\pm$ 0.02 <sup>B</sup>	0.27 $\pm$ 0.02 <sup>D</sup>	20.1	<0.01
OM content (%)	7.86 $\pm$ 0.43 <sup>BC</sup>	8.72 $\pm$ 0.64 <sup>BC</sup>	16.64 $\pm$ 3.2 <sup>BC</sup>	19.58 $\pm$ 3.2 <sup>B</sup>	10.42 $\pm$ 0.71 <sup>C</sup>	12.02 $\pm$ 0.53 <sup>BC</sup>	36.61 $\pm$ 1.8 <sup>A</sup>	38.09	<0.01
OM vol (%)	4.48 $\pm$ 0.53 <sup>ABC</sup>	4.05 $\pm$ 0.43 <sup>ABC</sup>	3.12 $\pm$ 0.31 <sup>C</sup>	4.12 $\pm$ 0.29 <sup>BC</sup>	5.50 $\pm$ 0.25 <sup>AB</sup>	4.92 $\pm$ 0.13 <sup>B</sup>	6.73 $\pm$ 0.36 <sup>A</sup>	9.85	<0.01
Mineral vol (%)	23.29 $\pm$ 3.96 <sup>A</sup>	19.33 $\pm$ 2.79 <sup>AB</sup>	10.41 $\pm$ 1.78 <sup>BC</sup>	11.29 $\pm$ 1.39 <sup>BC</sup>	22.31 $\pm$ 1.63 <sup>A</sup>	16.52 $\pm$ 0.76 <sup>A</sup>	7.18 $\pm$ 0.89 <sup>C</sup>	50.27	<0.01
<b>Carbon concentration (%)</b>									
SOC	1.26 $\pm$ 0.12 <sup>D</sup>	2.07 $\pm$ 0.33 <sup>BCD</sup>	5.36 $\pm$ 1.36 <sup>BC</sup>	6.55 $\pm$ 1.48 <sup>B</sup>	2.61 $\pm$ 0.31 <sup>CD</sup>	3.40 $\pm$ 0.26 <sup>BCD</sup>	16.69 $\pm$ 0.83 <sup>A</sup>	62.33	<0.01
RC	0.87 $\pm$ 0.19 <sup>D</sup>	1.34 $\pm$ 0.30 <sup>CD</sup>	4.04 $\pm$ 1.02 <sup>BC</sup>	4.67 $\pm$ 1.0 <sup>B</sup>	1.78 $\pm$ 0.23 <sup>CD</sup>	2.47 $\pm$ 0.22 <sup>BCD</sup>	12.37 $\pm$ 0.54 <sup>A</sup>	65.35	<0.01
LC	0.4 $\pm$ 0.11 <sup>C</sup>	0.73 $\pm$ 0.03 <sup>BC</sup>	1.33 $\pm$ 0.35 <sup>BC</sup>	1.88 $\pm$ 0.51 <sup>B</sup>	0.83 $\pm$ 0.10 <sup>BC</sup>	0.94 $\pm$ 0.08 <sup>BC</sup>	4.32 $\pm$ 0.32 <sup>A</sup>	38.09	<0.01
<b>Carbon density (mg cm<sup>-3</sup>)</b>									
SOC	8.57 $\pm$ 1.33 <sup>B</sup>	10.88 $\pm$ 2.22 <sup>B</sup>	10.7 $\pm$ 1.44 <sup>B</sup>	14.07 $\pm$ 1.30 <sup>B</sup>	14.56 $\pm$ 0.91 <sup>B</sup>	15.31 $\pm$ 0.83 <sup>B</sup>	35.40 $\pm$ 1.86 <sup>A</sup>	33.63	<0.01
RC	6.69 $\pm$ 1.71 <sup>B</sup>	6.94 $\pm$ 1.81 <sup>B</sup>	8.04 $\pm$ 1.22 <sup>B</sup>	10.21 $\pm$ 0.94 <sup>B</sup>	9.92 $\pm$ 0.82 <sup>B</sup>	10.84 $\pm$ 0.75 <sup>B</sup>	26.81 $\pm$ 1.47 <sup>A</sup>	31.72	<0.01
LC	1.89 $\pm$ 0.41 <sup>BC</sup>	3.94 $\pm$ 0.55 <sup>BC</sup>	2.67 $\pm$ 0.41 <sup>C</sup>	3.86 $\pm$ 0.57 <sup>BC</sup>	4.93 $\pm$ 0.28 <sup>B</sup>	4.46 $\pm$ 0.32 <sup>B</sup>	8.59 $\pm$ 0.45 <sup>A</sup>	20.69	<0.01

(table cont'd)

	Created marsh age (years)							
	5	8	13	20	32	Natural Reference	F value	P value
<b>Accretion rate (cm yr<sup>-1</sup>)</b>	0.48 ± 0.08 <sup>AB</sup>	0.63 ± 0.06 <sup>A</sup>	0.54 ± 0.03 <sup>AB</sup>	0.35 ± 0.03 <sup>B</sup>	0.44 ± 0.03 <sup>AB</sup>	0.52 <sup>AB</sup>	4.48	<0.01
<b>Carbon accumulation rate (g m<sup>-2</sup> yr<sup>-1</sup>)</b>								
SOC	52.2 ± 12.3 <sup>AB</sup>	66.9 ± 12.3 <sup>AB</sup>	75.7 ± 6.6 <sup>AB</sup>	52.01 ± 6.91 <sup>B</sup>	66.96 ± 6.17 <sup>AB</sup>	101.23 ± 15.23 <sup>A</sup>	2.87	0.03
RC	33.3 ± 9.6 <sup>B</sup>	50.3 ± 9.7 <sup>AB</sup>	54.9 ± 3.7 <sup>AB</sup>	34.73 ± 4.94 <sup>B</sup>	47.44 ± 4.77 <sup>AB</sup>	76.01 ± 11.43 <sup>A</sup>	3.59	0.01
LC	18.9 ± 3.3 <sup>A</sup>	16.7 ± 4.3 <sup>A</sup>	20.8 ± 3.3 <sup>A</sup>	17.28 ± 2.08 <sup>A</sup>	19.53 ± 2.31 <sup>A</sup>	25.26 ± 3.80 <sup>A</sup>	0.98	0.45

### 2.3.3. Organic carbon sources

$\delta^{13}\text{C}$  was more depleted in the soil compared to reference values for in-situ vegetation in the created marshes.

Vegetation in the created marshes was dominated by C<sub>4</sub> plants including *Spartina alterniflora* and *Spartina patens* with  $\delta^{13}\text{C}$  tissues ranging from -12.1 to -15.5‰ (Chmura and Aharon, 1995). Soil  $\delta^{13}\text{C}$  values in created marshes ranged from -30.3‰ in the 0-year-old marsh to -17.8 ‰ in the 32-year-old marsh across depths, and thus more depleted than the dominant vegetation. *Bolboschoenus robustus* was a dominant species in the natural marsh, comprising 60 ± 10% of the total aboveground biomass across plots. *Bolboschoenus robustus* is a C<sub>3</sub> species which  $\delta^{13}\text{C}$  values range between -24 and -34 ‰ (Smith and Epstein 1971). The range of  $\delta^{13}\text{C}$  values for the natural marsh soils (-27.1 and -17.5 ‰) reflect a

mix of C<sub>3</sub> and C<sub>4</sub> plants. There were no differences of  $\delta^{13}\text{C}$  with depth in any of the marshes (Figure 2.5). Recalcitrant soil had  $\delta^{13}\text{C}$  values that were more depleted than bulk soil ranging from -18.9‰ to -39.6‰, likely associated with the accumulation of lignin that is more depleted in  $\delta^{13}\text{C}$  (Benner et al. 1987) after hydrolysis. The  $\delta^{13}\text{C}$  depletion following hydrolysis was much greater in the created marshes than the natural marsh, averaging 8 and 2‰ depletion, respectively, indicating that the LC removed in the natural marsh is more similar to the RC remaining in the natural marsh.

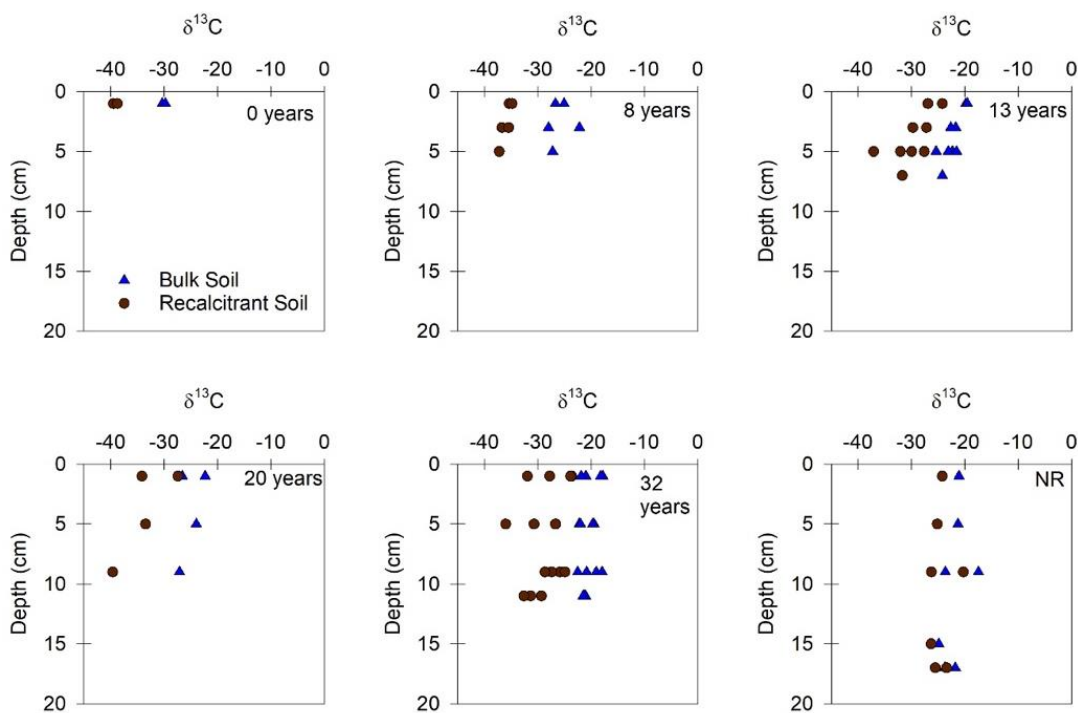


Figure 2.5. Depth profiles of  $\delta^{13}\text{C}$  in created and natural reference (NR) marshes in Sabine National Wildlife Refuge, Louisiana. Blue triangles represent the bulk soil (before hydrolysis), and the brown circles represent the recalcitrant sample (post-hydrolysis).



Overall,  $\delta^{13}\text{C}$  values increased with marsh age, except in the 20-year-old marsh where SOC was more depleted than the 13-year-old marsh ( $F_{5, 43} = 14.39$ ;  $p < 0.01$ ; Figure 2.6). The 0-year-old marsh, comprised of dredge material, was the only created marsh where  $\delta^{13}\text{C}$  values were more depleted from the natural reference marsh.  $\delta^{13}\text{C}$  values of the dredge sediment in the 0 year old marsh were  $-29.9 \pm 0.2$  ‰, and this value is most similar to either marine or terrestrial petroleum (Park and Epstein 1961). Since shoestring deposits of the Chenier Plain are oil-producing (Byrne et al. 1959), and dredge material may also contain old oil spill material if it is material from a navigational canal connecting oil and gas pipelines, making petroleum this is the most likely source as opposed some of the other similar values of fern and common wheat (Park and Epstein 1961).

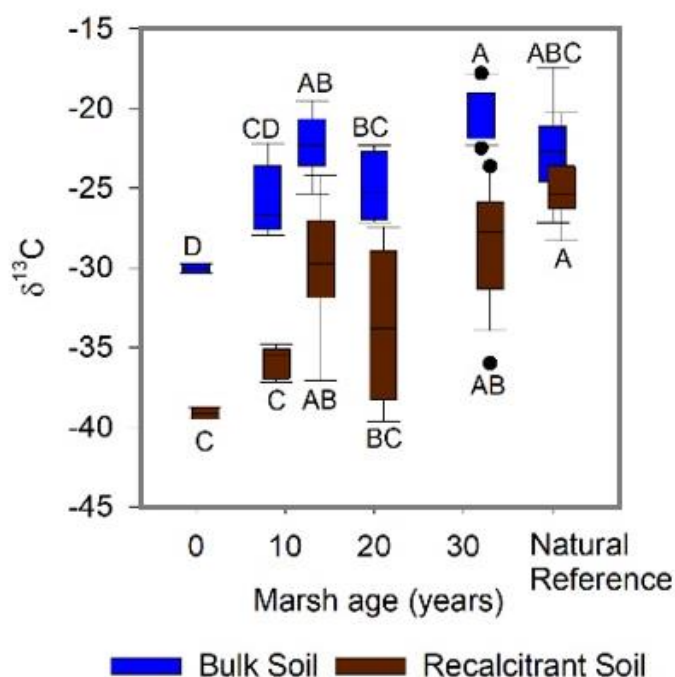


Figure 2.6.  $\delta^{13}\text{C}$  values in soils of created and natural marshes pre- and post-hydrolysis. Values represented by the same letters are not significantly different ( $p < 0.05$ ) from one-way ANOVA followed by Tukey's post hoc test.

### 2.3.4 Relationship between soil carbon stability and environmental parameters

#### Vegetation

The dominant plant species did not significantly affect the chemical stability of soil carbon across marshes ( $p > 0.05$ ). Because all species were not present in all marshes, the effect of plant species within marshes was tested where possible. In the 20 year-old marsh, SOC and RC density were higher in soils where *D. spicata* was dominant than in soils where *B. maritima* occurred (RC,  $F_{2,5} = 10.34$ ;  $p < 0.05$ ; SOC,  $F_{2,5} = 10.61$ ;  $p < 0.05$ ). Across created marshes, both RC and SOC accumulation rates were positively, but weakly related to stem density (RC,  $R^2 = 0.14$ ,  $p < 0.05$ ; SOC,  $R^2 = 0.16$ ,  $p < 0.05$ ; Figure 2.7). SOC, RC, and LC accumulation rates were not related to above ground biomass ( $p > 0.05$ ).

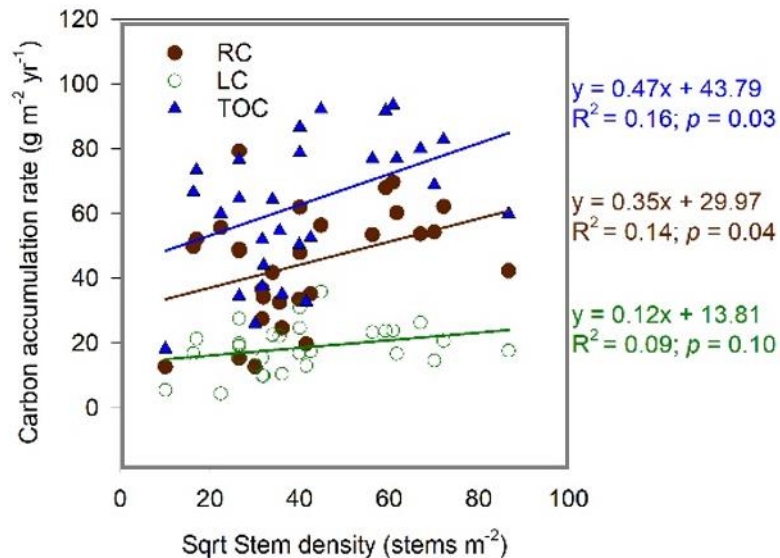


Figure 2.7. The relationship between stem density and total soil organic carbon (SOC), recalcitrant carbon (RC) and labile carbon (LC) in created marshes of southwest Louisiana.

## Mineral sediment

Relationships between mineral sediment density and SOC were only present in select marshes with a relatively high mineral sediment density (i.e., 20 and 32-year-old marshes with  $0.58 \pm 0.04$  and  $0.44 \pm 0.02$  g cm<sup>-3</sup>, respectively). Mineral sediment density was positively related to the proportion of SOC that was labile (20 -year-old,  $R^2 = 0.34$ ;  $p < 0.01$  and 32-year-old,  $R^2 = 0.36$ ;  $p < 0.01$ ; Figure 2.8), indicating that the preservation mechanisms of LC with mineral sediment were only present when there was a certain threshold of mineral sediment.

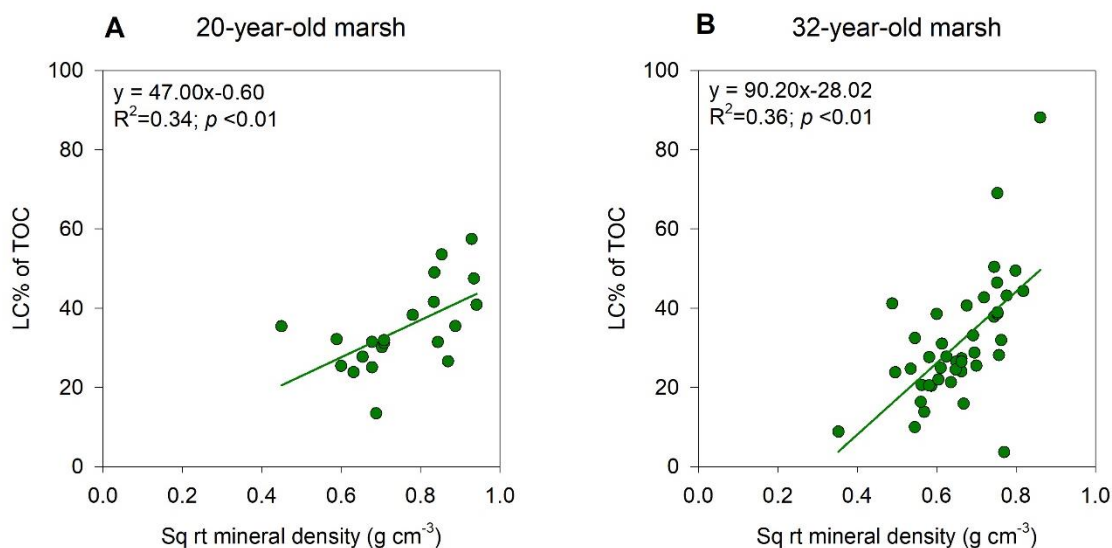


Figure 2.8. Relationships between mineral density and the labile carbon (LC) concentration of total organic carbon (SOC) in (a) the 20-year-old, and (b) the 32-year-old created marshes. Transformed mineral density data are displayed.

## 2.4. Discussion

### 2.4.1. Soil carbon stability with created marsh age

The development of ecosystem properties in created marshes is dependent upon environmental factors influencing sediment delivery, primary production, and microbial

activity, which may determine carbon accumulation rates and stocks (Craft et al. 2003, Burden et al. 2013). We have two distinct environments in created marshes in this study, the interior, young created marshes (0 - 13 years), and the older, exterior marshes (20 and 32 years). Differences in proximity to the ship channel and hydrology, along with variations in marsh elevations and associated vegetation types, complicate direct evaluations of the progression of ecosystem properties over time in created marshes. The natural reference marsh was located in the interior area and, therefore, is similar to the interior marshes and may not reflect conditions of the exterior.

The SOC density values in the created marshes were within a similar range of other created and restored marshes (Havens et al. 1995, Craft et al. 1999, Burden et al. 2013). The LC fraction of SOC ( $27.6 \pm 0.8\%$ ) was very similar to values found in Louisiana natural marshes (24-28 %) in Barataria Bay (Chapter 1, Dodla et al. 2012). A higher labile fraction of SOC was found in the 5, 20, and 32-year-old marsh indicating that age is not the main factor influencing lability.

Only a handful of studies have examined the chemical stability of SOC in coastal marshes, but there is extensive literature, on this subject in inland wetlands and terrestrial systems. Recalcitrant organic carbon comprised an average of  $71.6 \pm 0.8\%$  of SOC across created and natural marshes in the present study, which was comparable to upland and inland wetland systems of Georgia and Alabama where RC fraction of SOC ranged from 68 to 82% (Silveira et al. 2008). Similarly, RC fraction of SOC in clay loam, silt loam, and sandy loam soils in Minnesota ranged between 66 and 71% (Xu et al. 1997). However agricultural fields, forests, and grasslands were found to have a lower amount of RC per unit of SOC ranging between 28 and 60% (Paul et al.

2006). This study found that RC was linearly related to SOC, which has also been found in several other studies (Paul et al. 2006, Dodla et al. 2012). The slopes of the line between SOC and RC differs among studies, indicating that the recalcitrant proportion of SOC also differs. However, our RC fraction is higher than terrestrial that were cultivated, forest and grassland averaging 44.4% RC of SOC (Paul et al. 2006). Although carbon source may be important in influencing LC and RC inputs, environmental conditions have been suggested to be more important in determining the SOC composition in wetlands (Wang et al. 2011). The effect of carbon source may be neutralized by decomposition of LC, regardless of original LC content, such that the RC proportion of litter increases over time. There is an equalization effect, as species high in LC decompose quickly, and species high in RC decompose slowly, and the end RC fraction for both is similar (Rovira and Vallejo 2002). However, in wetlands, preservation mechanisms can protect LC from decomposition, such as or anaerobic conditions (DeBusk and Reddy 1998) or mineral particle interactions (Rovira and Vallejo 2003, Paul et al. 2006), that allow for higher concentrations of LC than otherwise would be expected. In this study, the mechanism of mineral sediment preservation was only found in higher mineral sediment marshes, suggesting that under these conditions, LC is deposited or protected more readily with greater mineral sediment deposition, however, in marshes with lower mineral sediment, other mechanisms become more important in controlling the LC fraction.

Few studies have reported RC and LC accumulation rates in marshes. Natural reference marshes in SNWR had similar rates of SOC accumulation ( $101 \pm 15 \text{ g m}^{-2} \text{ yr}^{-1}$ ) to those in salt marshes of a coastal lagoon in New Jersey ( $100 - 169 \text{ g m}^{-2} \text{ yr}^{-1}$ ) but

were about half of their coastal plain estuary ( $180 - 331 \text{ g m}^{-2} \text{ yr}^{-1}$ ; Unger et al. 2016). RC accumulation rates were similar with  $75 \pm 11.43 \text{ g m}^{-2} \text{ yr}^{-1}$  in this study compared to the range of 63 to  $95 \text{ g m}^{-2} \text{ yr}^{-1}$  in the coastal lagoon, and 72 to  $93 \text{ g m}^{-2} \text{ yr}^{-1}$  in the coastal plain estuary (Unger et al. 2016). LC accumulation rates in this study were lower even in the natural marsh with the highest LC accumulation rate ( $25 \pm 4 \text{ g m}^{-2} \text{ yr}^{-1}$ ) compared to 37 to  $97 \text{ g m}^{-2} \text{ yr}^{-1}$  in the coastal lagoon, and less than a quarter than those found in the coastal plain estuary ( $109 \text{ to } 238 \text{ g m}^{-2} \text{ yr}^{-1}$ ; Unger et al. 2016). Despite the fact that most of our SOC accumulation rates were lower than those found in the mid-Atlantic, our RC accumulation were similar, therefore both regions are accumulating similar amounts of stable carbon.

#### 2.4.2. Relationships between carbon stability and environmental parameters

Factors such as tidal amplitude, average water depth and salinity have been shown to influence decomposition rates (Stagg et al. 2017a) and therefore, can influence the stability and accumulation of SOC (Craft 2007). Elevation played an important role in influencing the RC and SOC accumulation rate in the lowest elevation created marsh, which was the 8-year old marsh. Here, elevation ranged from -12.2 to 4.1 cm, NAVD88, and RC accumulation rates were six times higher at high elevations. At low elevations, inputs of RC through belowground plant production can be limited due to water logging and anaerobic conditions (DeLaune et al. 1983). At the highest elevations of 4.1 cm, NAVD88, the 8-year old marsh had RC accumulation rates of  $79.27 \text{ g m}^{-2} \text{ yr}^{-1}$ . At similar elevations, the RC accumulation rate in the 5-year-old marsh was 50% lower, which was unexplained by aboveground vegetation properties and mineral sediment accumulation rate, which was similar between marshes. Because OM

content is related to the quantity of belowground biomass (Nyman et al. 1990), the 5-year-old marsh may have less developed belowground biomass and therefore, lower rates of RC accumulation than the 8-year-old marsh at similar elevations. This suggests that marsh age and elevation interact to influence RC accumulation rates. Other studies have found that aboveground production can be similar to that in natural reference marshes within 2 growing seasons, while belowground production can take up to 4 growing seasons when marshes are planted with vegetation and potentially longer in marshes that are dependent on natural colonization. While elevation influenced RC and SOC, but not LC accumulation rates in the 8-year-old marsh this is likely due to the high recalcitrance of belowground root material (Elsey-Quirk and Unger 2018), and the high correlation between RC and SOC. It is likely that elevation did not influence LC accumulation rates because elevation differences and marsh age may have more directly influenced RC through the direct effects on root productivity.

Vegetation, and its associated properties plays an important role in the carbon cycle. Abbott (2017) illustrated a weak but positive relationship between SOC accumulation rate and stem density across the same created marshes in this study. High density species include those found at later successional stages and higher elevations such as *S. patens* and *D. spicata* (e.g. 3,900 stem m<sup>-2</sup>). We found that RC accumulation rate was also weakly but positively related to stem density ( $R^2 = 0.14$ ;  $p < 0.05$ ), while LC accumulation rate was not ( $p > 0.05$ ), suggesting that higher stem density species contributes to the input of RC. When directly comparing the chemical stability of SOC between vegetation types, RC and SOC density were significantly higher in areas dominated by high density *D. spicata* ( $1363 \pm 339$  stems m<sup>-2</sup>), compared

to *B. maritima* ( $303 \pm 98$  stems  $m^{-2}$ ). However, there were no significant relationships between LC and dominant plant species. Mineral sediment had a positive relationship with LC accumulation in the older marshes. These results indicate that creating marshes that promote the establishment of *S. patens* and *D. spicata* at higher elevations, and with less mineral sediment will promote the burial of RC for long term SOC burial.

#### 2.4.3. Carbon source with created marsh age

Natural marshes contained over 50% *B. robustus*, a  $C_3$  plant, and  $\delta^{13}C$  values were very similar between the soil and expected values of  $C_3$  vegetation, indicating soil carbon dominated by local vegetation. Soil  $\delta^{13}C$  values in created marshes, however, were always more depleted than  $\delta^{13}C$  values of vegetation obtained from Chmura et al. 1987. The depletion of  $\delta^{13}C$  in soil of created marshes relative to the vegetation may be caused by the dredge sediment, which was much more depleted than all other bulk soil values. Although samples were taken above the dredge horizon layer in this study, mineral particles may have been mixed into the OM layer as it accumulated and tides, and/or fauna move particles vertically. Depletion due to underlying substrate was also found in a transplanted marsh with top soil additions used to create the ideal elevation (Craft et al. 1988).  $\delta^{13}C$  depletion could also be associated with algae being a dominant contributor, which averages  $-21\text{‰}$  (Sullivan and Moncreiff, 1990). Most of the marshes of Sabine are more depleted than this value, so algae alone could not cause the amount of depletion measured, except in the 13, and 32-year-old marsh. Another cause of depletion is decomposition and concentration of lignin, which in *S. alterniflora* is about  $4\text{‰}$  more depleted than bulk vegetation, causes the soil  $\delta^{13}C$  to become more depleted



(Benner 1987). Following hydrolysis, recalcitrant  $\delta^{13}\text{C}$  was always more depleted than bulk soil due to the removal of LC, and the concentration of lignin. However, the difference in bulk and recalcitrant  $\delta^{13}\text{C}$  tended to decrease with age, especially in the natural reference marshes. One possible explanation the depletion is less in natural reference is because it is dominated by  $\text{C}_3$  plants which  $\delta^{13}\text{C}$  of smoke and ash are similar to the original vegetation, while  $\text{C}_4$  plant smoke and ash becomes more depleted (Das et al. 2010).  $\delta^{13}\text{C}$  values tended to increase from the 0-year-old marsh, composed of dredge sediment, to the 32-year-old marsh, and therefore, it appears that the source of SOC becomes more dominated by the in-situ vegetation as marshes age, and influence of dredge sediment.

### Chapter 3. Conclusions

Freshwater, brackish, and salt marshes in Barataria Basin, LA had similar rates of total, labile and recalcitrant organic carbon accumulation averaging  $34 \pm 4 \text{ g m}^{-2}\text{yr}^{-1}$  and  $93 \pm 8 \text{ g m}^{-2}\text{yr}^{-1}$ , respectively. SOC accumulation rate in Barataria Bay is approximately half of the average of the U.S. in brackish and salt marshes, and about 80% of U.S. freshwater marshes (Craft 2007). Approximately 73% of the SOC was chemically recalcitrant across marsh types. Slightly higher LC fractions in freshwater marshes suggest that there may be effects of higher O.M. quality (i.e. higher lability) in freshwater marshes. Carbon isotope analysis indicated that most of the SOC is derived from local vegetation, although variation with depth of  $\delta^{13}\text{C}$  and the presence of  $\text{C}_3$  and  $\text{C}_4$  vegetation in brackish and salt marshes suggest that changes in plant communities, from  $\text{C}_4$  plants such as *S. patens* to  $\text{C}_3$  plants such as *J. roemerianus*, could be relatively large shifts of  $\delta^{13}\text{C}$ , which was previously attributed to changes in depositional

environments of the rapidly accreting marshes (DeLaune 1986, Chmura 1990).

Relationships between SOC and vegetation and mineral sediment were variable across marsh types. Very few studies have examined the chemical reactivity of SOC in marshes and this study contributes to understanding of how SOC accumulates across the salinity gradient.

SOC accumulation rates in created marshes of SNWR were lower than other studies (Craft 2007, Unger et al. 2016), the majority of the organic carbon that is accumulating is recalcitrant or chemically stable, suggesting that it has the potential to contribute to long-term carbon storage. Additionally, SOC and RC stocks are increasing with marsh age. Carbon source appears to be dominated by local vegetation, and the existing carbon from the dredge material. Mineral sediment increased with LC fraction in older created marshes with higher mineral sediment, indicating that the other marshes have other more important factors here. RC and TOC accumulation rate was positively associated with elevation in the lowest elevation created marsh (8 year old) suggesting that greater flooding reduces the recalcitrant biomass contribution to the SOC pool. Results from this study show that to promote stable SOC burial in created marshes, higher elevations for *S. patens* and *D. spicata* should be encouraged, however elevations that are too high, such as those in the 20-year-old marsh may have lower accretion rates compared to other marshes.

## APPENDIX

Table A.1. Sample locations at CRMS sites.

CRMS Site	Sample Name
189	FW1
192	FW2
3169	FW3
3985	FW4
225	BM5
3565	BM6
3617	BM7
173	SM8
224	SM9
237	SM10
4529	SM11

Table A.2. Selection of cores for acid hydrolysis and  $\delta^{13}\text{C}$  analysis from Abbott (2017).

Marsh Age	Species	Number of Cores	Core ID
0	<i>S. alterniflora</i>	1	1
0	No Vegetation	2	1,2
8	<i>S. alterniflora</i>	2	1,4
13	<i>S. alterniflora</i>	2	2,6
13	<i>D. Spicata</i>	2	1,3
20	<i>D. Spicata</i>	2	1,6
32	<i>S. alterniflora</i>	2	1,3
32	<i>D. Spicata</i>	2	4,6
Natural Reference	<i>D. Spicata</i>	3	1,2,3

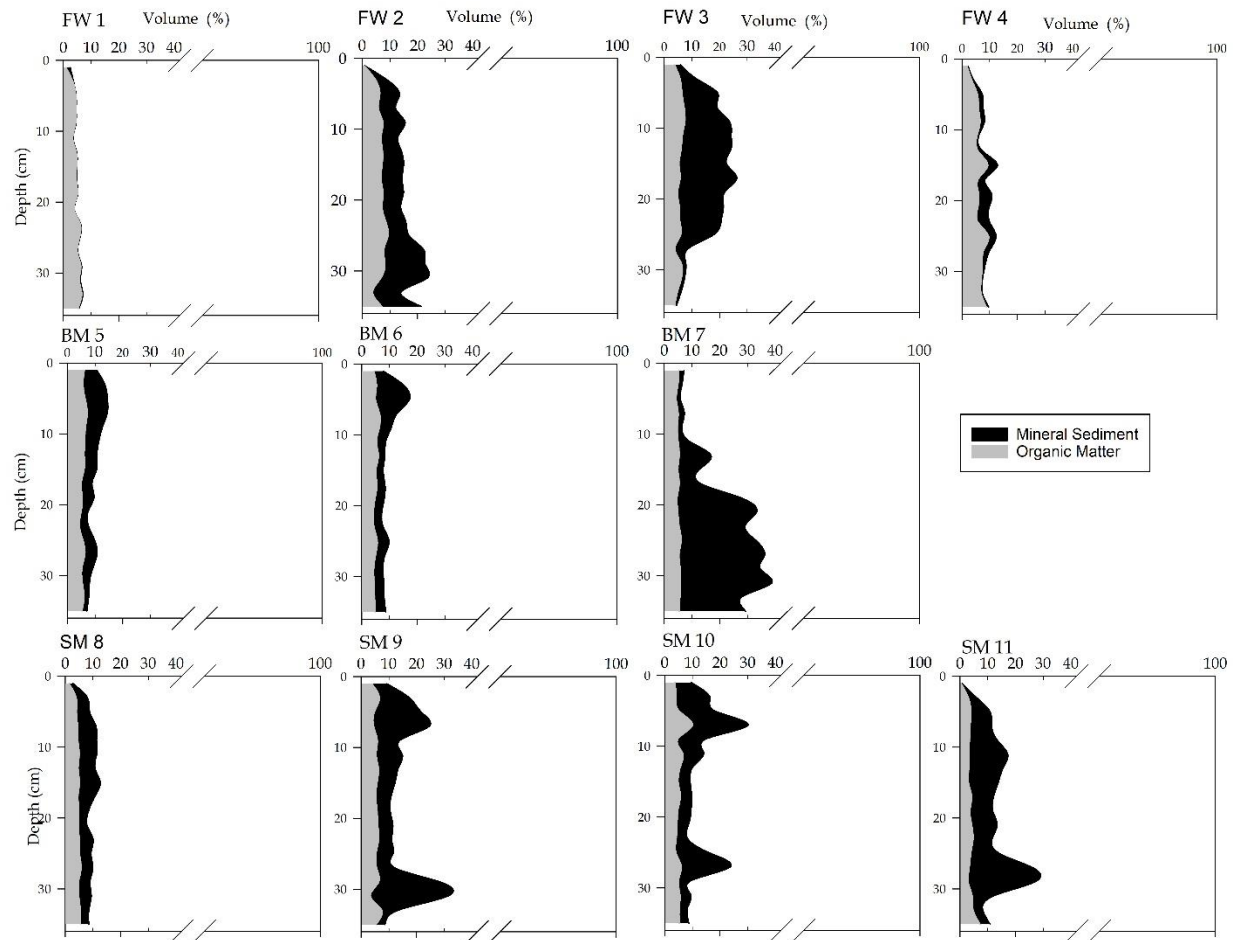


Figure A.1. Percent volume of organic matter, mineral sediment, and pore space in soil cores collected in freshwater (top row), brackish (middle), and salt (bottom row) marshes in Barataria Bay, Louisiana.

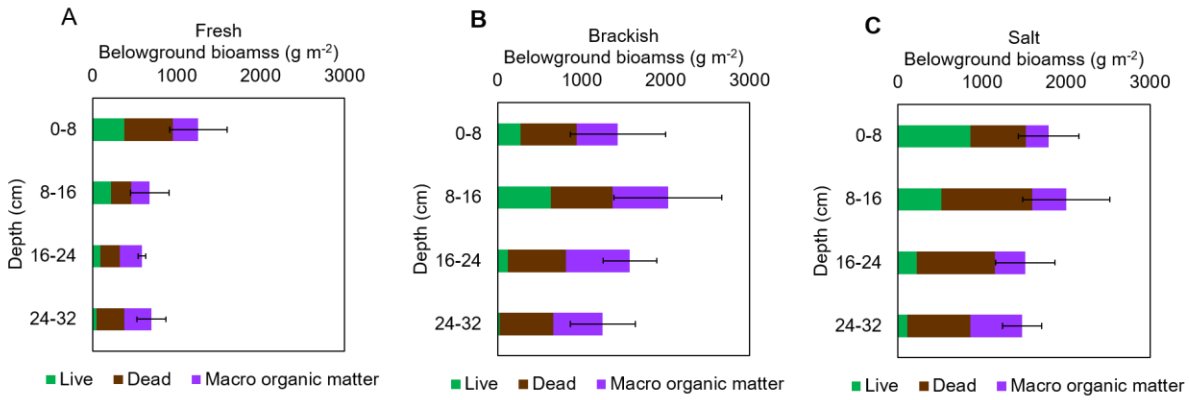


Figure A.2. Belowground biomass depth profiles in freshwater (A), brackish (B), and salt (C) marshes in Barataria Bay, Louisiana (n = 3,  $\pm$  standard error).

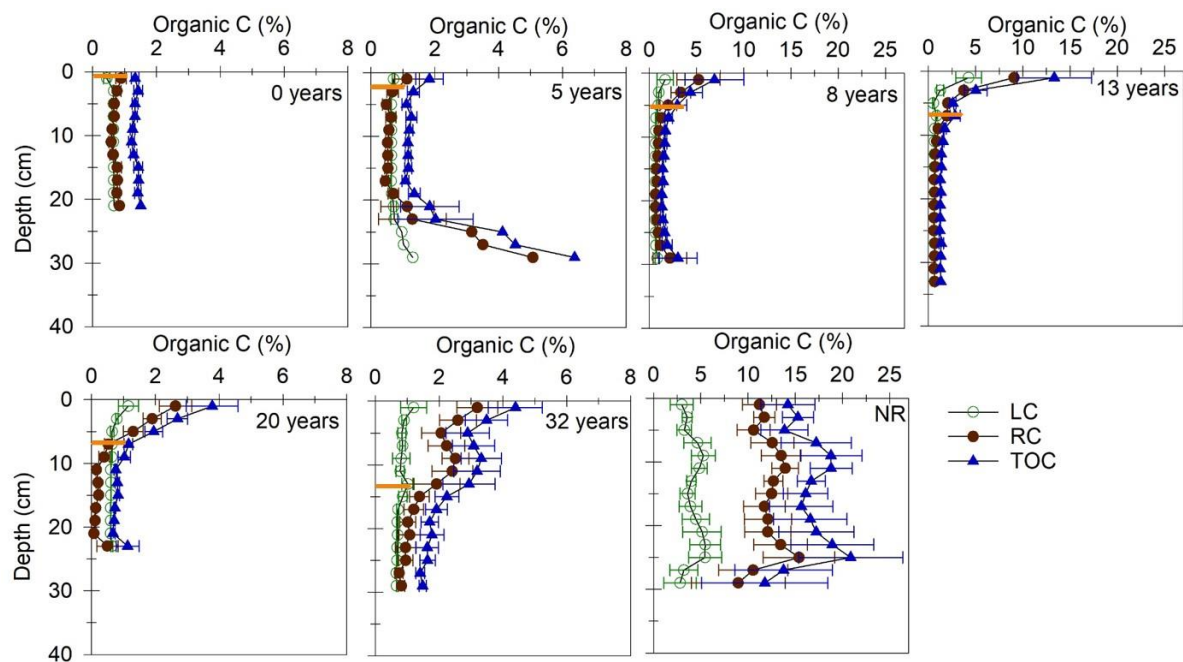


Figure A.3. Organic SOC % depth profiles in created and natural reference (NR) marshes in the Chenier Plain Louisiana. Values are means  $\pm$  standard error ( $n = 6$ ). Orange line represents the average dredge horizon layer for created marshes.

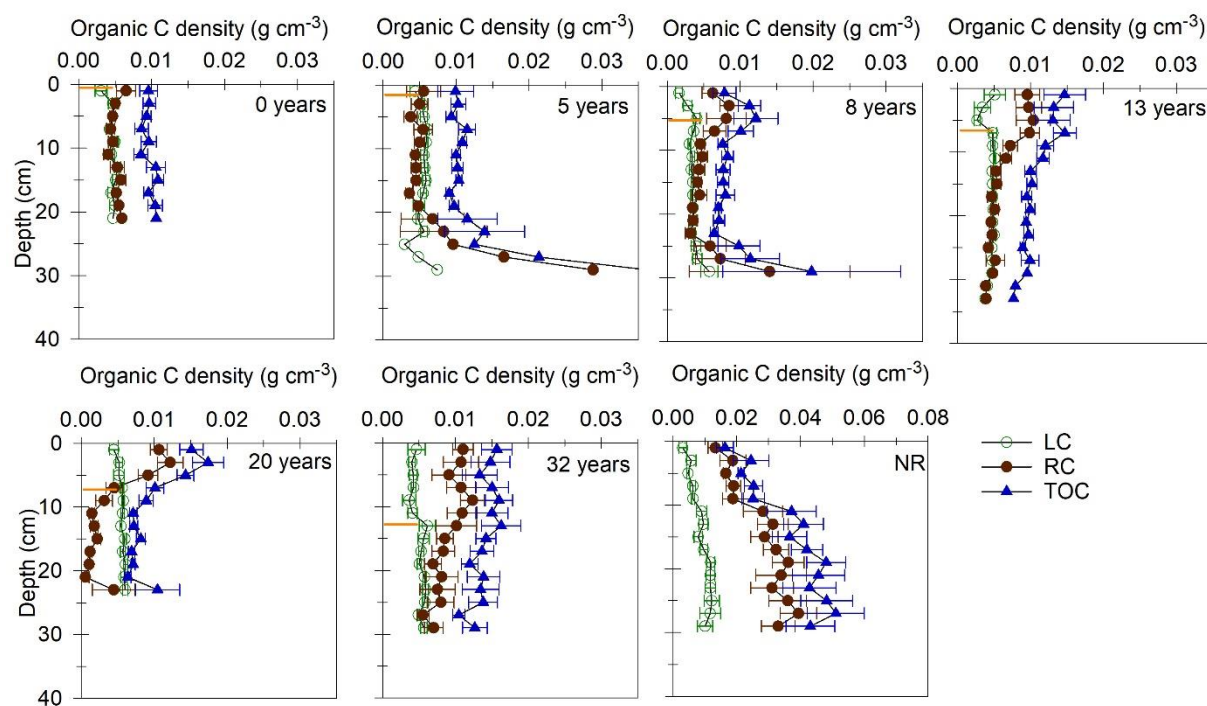


Figure A.4. Organic SOC density profiles for created marshes 0 to 32 years and natural references (NR) in Sabine National Wildlife Refuge, LA. Values are means for 6 cores for each marsh, and error bars are stand error. Orange line represents the average dredge horizon layer for created marshes.



## References

- Baldocchi, D. 1994. A comparative study of mass and energy exchange rates over a closed C3 (wheat) and an open C4 (corn) crop: II. CO<sub>2</sub> exchange and water use efficiency. *Agricultural and forest meteorology* **67**:291-321.
- Benner, R., M. L. Fogel, E. K. Sprague, and R. E. Hodson. 1987. Depletion of <sup>13</sup>C in lignin and its implications for stable carbon isotope studies. *Nature* **329**:708-710.
- Benner, R., A. Maccubbin, and R. E. Hodson. 1984. Anaerobic biodegradation of the lignin and polysaccharide components of lignocellulose and synthetic lignin by sediment microflora. *Applied and Environmental Microbiology* **47**:998-1004.
- Bernier, J. C., R. A. Morton, and K. W. Kelso. 2011. Trends and causes of historical wetland loss, Sabine National Wildlife Refuge, southwest Louisiana. 2331-1258, US Geological Survey.
- Blum, M. D., and H. H. Roberts. 2009. Drowning of the Mississippi Delta due to insufficient sediment supply and global sea-level rise. *Nature Geoscience* **2**:488.
- Boesch, D. F., M. N. Josselyn, A. J. Mehta, J. T. Morris, W. K. Nuttle, C. A. Simenstad, and D. J. Swift. 1994. Scientific assessment of coastal wetland loss, restoration and management in Louisiana. *Journal of Coastal Research*:i-103.
- Britsch, L. D., and J. B. Dunbar. 1993. Land loss rates: Louisiana coastal plain. *Journal of Coastal Research*:324-338.
- Burden, A., R. A. Garbutt, C. D. Evans, D. L. Jones, and D. M. Cooper. 2013. Carbon sequestration and biogeochemical cycling in a saltmarsh subject to coastal managed realignment. *Estuarine, Coastal and Shelf Science* **120**:12-20.
- Byrne, J. V., D. O. LeRoy, and C. M. Riley. 1959. The chenier plain and its stratigraphy, southwestern Louisiana.
- Chambers, L. G., T. Z. Osborne, and K. R. Reddy. 2013. Effect of salinity-altering pulsing events on soil organic carbon loss along an intertidal wetland gradient: a laboratory experiment. *Biogeochemistry* **115**:363-383.
- Chambers, L. G., K. R. Reddy, and T. Z. Osborne. 2011. Short-term response of carbon cycling to salinity pulses in a freshwater wetland. *Soil Science Society of America Journal* **75**:2000-2007.
- Cheng, L., S. Leavitt, B. Kimball, P. Pinter, M. Ottman, A. Matthias, G. Wall, T. Brooks, D. Williams, and T. Thompson. 2007. Dynamics of labile and recalcitrant soil carbon pools in a sorghum free-air CO<sub>2</sub> enrichment (FACE) agroecosystem. *Soil Biology and Biochemistry* **39**:2250-2263.

- Chmura, G., and P. Aharon. 1995. Stable carbon isotope signatures of sedimentary carbon in coastal wetlands as indicators of salinity regime. *Journal of Coastal Research*:124-135.
- Chmura, G., P. Aharon, R. Socki, and R. Abernethy. 1987. An inventory of  $^{13}\text{C}$  abundances in coastal wetlands of Louisiana, USA: vegetation and sediments. *Oecologia* **74**:264-271.
- Chmura, G. L. 1990. Palynological and Carbon-Isotopic Techniques for Reconstruction of Paleomarch Salinity Zones.
- Chmura, G. L., S. C. Anisfeld, D. R. Cahoon, and J. C. Lynch. 2003. Global carbon sequestration in tidal, saline wetland soils. *Global biogeochemical cycles* **17**.
- Couvillion, B. R., H. Beck, D. Schoolmaster, and M. Fischer. 2017. Land area change in coastal Louisiana (1932 to 2016). Report 3381, Reston, VA.
- CPRA, C. P. a. R. A. o. L. 2019. Coastwide Reference Monitoring System-Wetlands Monitoring Data. Coastal Information Management System (CIMS) database.
- Craft, C. 2007. Freshwater input structures soil properties, vertical accretion, and nutrient accumulation of Georgia and US tidal marshes. *Limnology and Oceanography* **52**:1220-1230.
- Craft, C., S. Broome, E. Seneca, and W. Showers. 1988. Estimating sources of soil organic matter in natural and transplanted estuarine marshes using stable isotopes of carbon and nitrogen. *Estuarine, Coastal and Shelf Science* **26**:633-641.
- Craft, C., P. Megonigal, S. Broome, J. Stevenson, R. Freese, J. Cornell, L. Zheng, and J. Sacco. 2003. The pace of ecosystem development of constructed *Spartina alterniflora* marshes. *Ecological applications* **13**:1417-1432.
- Craft, C., J. Reader, J. N. Sacco, and S. W. Broome. 1999. Twenty-five years of ecosystem development of constructed *Spartina alterniflora* (Loisel) marshes. *Ecological applications* **9**:1405-1419.
- CWPPRA. 2018. The Barataria Basin.
- Das, O., Y. Wang, and Y.-P. Hsieh. 2010. Chemical and carbon isotopic characteristics of ash and smoke derived from burning of  $\text{C}_3$  and  $\text{C}_4$  grasses. *Organic geochemistry* **41**:263-269.
- DeBusk, W., and K. Reddy. 1998. Turnover of detrital organic carbon in a nutrient-impacted Everglades marsh. *Soil Science Society of America Journal* **62**:1460-1468.
- DeLaune, R. 1986. The use of  $\delta^{13}\text{C}$  signature of  $\text{C}_3$  and  $\text{C}_4$  plants in determining past depositional environments in rapidly accreting marshes of the Mississippi River deltaic plain, Louisiana, USA. *Chemical Geology: Isotope Geoscience section* **59**:315-320.

- DeLaune, R., R. Baumann, and J. Gosselink. 1983. Relationships among vertical accretion, coastal submergence, and erosion in a Louisiana Gulf Coast marsh. *Journal of Sedimentary Research* **53**:147-157.
- DeLaune, R., and C. Lindau. 1987.  $\delta^{13}\text{C}$  signature of organic carbon in estuarine bottom sediment as an indicator of carbon export from adjacent marshes. *Biogeochemistry* **4**:225-230.
- DeLaune, R., and J. White. 2012. Will coastal wetlands continue to sequester carbon in response to an increase in global sea level?: a case study of the rapidly subsiding Mississippi river deltaic plain. *Climatic Change* **110**:297-314.
- Dodla, S., J. Wang, and R. Delaune. 2012. Characterization of labile organic carbon in coastal wetland soils of the Mississippi River deltaic plain: Relationships to carbon functionalities.
- Duarte, C. M., W. C. Dennison, R. J. W. Orth, and T. J. B. Carruthers. 2008. The Charisma of Coastal Ecosystems: Addressing the Imbalance. *Estuaries and Coasts* **31**:233-238.
- Edwards, K. R., and C. E. Proffitt. 2003. Comparison of wetland structural characteristics between created and natural salt marshes in southwest Louisiana, USA. *Wetlands* **23**:344-356.
- Elsey-Quirk, T., and V. Unger. 2018. Geomorphic influences on the contribution of vegetation to soil C accumulation and accretion in *Spartina alterniflora* marshes. *Biogeosciences* **15**:379-397.
- FitzGerald, D. M., M. S. Fenster, B. A. Argow, and I. V. Buynevich. 2008. Coastal impacts due to sea-level rise. *Annual Review of Earth and Planetary Sciences* **36**.
- Foote, A. L., and K. A. Reynolds. 1997. Decomposition of saltmeadow cordgrass (*Spartina patens*) in Louisiana coastal marshes. *Estuaries* **20**:579-588.
- Gao, J. H., Z. X. Feng, L. Chen, Y. P. Wang, F. Bai, and J. Li. 2016. The effect of biomass variations of *Spartina alterniflora* on the organic carbon content and composition of a salt marsh in northern Jiangsu Province, China. *Ecological Engineering* **95**:160-170.
- Harris, D., W. R. Horwáth, and C. Van Kessel. 2001. Acid fumigation of soils to remove carbonates prior to total organic carbon or carbon-13 isotopic analysis. *Soil Science Society of America Journal* **65**:1853-1856.
- Havens, K. J., L. M. Varnell, and J. G. Bradshaw. 1995. An assessment of ecological conditions in a constructed tidal marsh and two natural reference tidal marshes in coastal Virginia. *Ecological Engineering* **4**:117-141.
- Hedges, J. I., and J. H. Stern. 1984. Carbon and nitrogen determinations of carbonate-containing solids. *Limnology and Oceanography* **29**:657-663.

- Herbert, E. R., P. Boon, A. J. Burgin, S. C. Neubauer, R. B. Franklin, M. Ardón, K. N. Hopfensperger, L. P. Lamers, and P. Gell. 2015. A global perspective on wetland salinization: ecological consequences of a growing threat to freshwater wetlands. *Ecosphere* **6**:1-43.
- Jobbágy, E. G., and R. B. Jackson. 2000. The vertical distribution of soil organic carbon and its relation to climate and vegetation. *Ecological applications* **10**:423-436.
- Kirwan, M., and L. Blum. 2011. Enhanced decomposition offsets enhanced productivity and soil carbon accumulation in coastal wetlands responding to climate change. *Biogeosciences* **8**:987-993.
- Krull, E. S., J. A. Baldock, and J. O. Skjemstad. 2003. Importance of mechanisms and processes of the stabilisation of soil organic matter for modelling carbon turnover. *Functional plant biology* **30**:207-222.
- Laursen, K. R. 2004. The effects of nutrient enrichment on the decomposition of belowground organic matter in a *Sagittaria lancifolia*-dominated oligohaline marsh.
- Lehmann, J., J. Kinyangi, and D. Solomon. 2007. Organic matter stabilization in soil microaggregates: implications from spatial heterogeneity of organic carbon contents and carbon forms. *Biogeochemistry* **85**:45-57.
- Lehmann, J., and M. Kleber. 2015. The contentious nature of soil organic matter. *Nature* **528**:60.
- Leorri, E., A. R. Zimmerman, S. Mitra, R. R. Christian, F. Fatela, and D. J. Mallinson. 2018. Refractory organic matter in coastal salt marshes-effect on C sequestration calculations. *Science of the Total Environment* **633**:391-398.
- Loomis, M. J., and C. B. Craft. 2010. Carbon sequestration and nutrient (nitrogen, phosphorus) accumulation in river-dominated tidal marshes, Georgia, USA. *Soil Science Society of America Journal* **74**:1028-1036.
- Lützw, M. v., I. Kögel-Knabner, K. Ekschmitt, E. Matzner, G. Guggenberger, B. Marschner, and H. Flessa. 2006. Stabilization of organic matter in temperate soils: mechanisms and their relevance under different soil conditions—a review. *European Journal of Soil Science* **57**:426-445.
- McKee, K. L., and E. D. Seneca. 1982. The influence of morphology in determining the decomposition of two salt marsh macrophytes. *Estuaries* **5**:302-309.
- McLauchlan, K., and S. Hobbie. 2004. Comparison of Labile Soil Organic Matter Fractionation Techniques.
- McLeod, E., G. L. Chmura, S. Bouillon, R. Salm, M. Björk, C. M. Duarte, C. E. Lovelock, W. H. Schlesinger, and B. R. Silliman. 2011. A blueprint for blue carbon: toward an improved understanding of the role of vegetated coastal habitats in sequestering CO<sub>2</sub>. *Frontiers in Ecology and the Environment* **9**:552-560.

- Mikutta, R., M. Kleber, M. S. Torn, and R. Jahn. 2006. Stabilization of soil organic matter: association with minerals or chemical recalcitrance? *Biogeochemistry* **77**:25-56.
- Mudd, S. M., S. M. Howell, and J. T. Morris. 2009. Impact of dynamic feedbacks between sedimentation, sea-level rise, and biomass production on near-surface marsh stratigraphy and carbon accumulation. *Estuarine, Coastal and Shelf Science* **82**:377-389.
- Nellemann, C., E. Corcoran, C. M. Duarte, C. De Young, L. E. Fonseca, and G. Grimsdith. 2010. Blue carbon: the role of healthy oceans in binding carbon.
- Neubauer, S. C. 2013. Ecosystem Responses of a Tidal Freshwater Marsh Experiencing Saltwater Intrusion and Altered Hydrology. *Estuaries and Coasts* **36**:491-507.
- Noe, G. B., C. R. Hupp, C. E. Bernhardt, and K. W. Krauss. 2016. Contemporary deposition and long-term accumulation of sediment and nutrients by tidal freshwater forested wetlands impacted by sea level rise. *Estuaries and Coasts* **39**:1006-1019.
- Nyman, J., R. DeLaune, and W. Patrick Jr. 1990. Wetland soil formation in the rapidly subsiding Mississippi River deltaic plain: Mineral and organic matter relationships. *Estuarine, Coastal and Shelf Science* **31**:57-69.
- O'Leary, M. H. 1981. Carbon isotope fractionation in plants. *Phytochemistry* **20**:553-567.
- Odum, W. E. 1988. Comparative ecology of tidal freshwater and salt marshes. *Annual Review of ecology and Systematics* **19**:147-176.
- Orson, R. A., R. S. Warren, and W. A. Niering. 1987. Development of a tidal marsh in a New England river valley. *Estuaries* **10**:20.
- Ouyang, X., and S. Lee. 2014. Update estimates of carbon accumulation rates in coastal marsh sediments.
- Park, R., and S. Epstein. 1961. Metabolic fractionation of C<sup>13</sup> & C<sup>12</sup> in plants. *Plant physiology* **36**:133.
- Paul, E., R. Follett, S. Leavitt, A. Halvorson, G. Peterson, and D. Lyon. 1997. Radiocarbon dating for determination of soil organic matter pool sizes and dynamics. *Soil Science Society of America Journal* **61**:1058-1067.
- Paul, E. A., S. J. Morris, R. T. Conant, and A. F. Plante. 2006. Does the acid hydrolysis-incubation method measure meaningful soil organic carbon pools? *Soil Science Society of America Journal* **70**:1023-1035.
- Penland, S., and K. E. Ramsey. 1990. Relative sea-level rise in Louisiana and the Gulf of Mexico: 1908-1988. *Journal of Coastal Research*:323-342.
- Pezeshki, S., and R. DeLaune. 1991. A comparative study of above-ground productivity of dominant US Gulf Coast marsh species. *Journal of Vegetation Science* **2**:331-338.

- Proffitt, C. E., and J. Young. 1998. Salt marsh plant colonization, growth, and dominance on large mudflats created using dredged sediments. Recent research in coastal Louisiana: natural system function and response to human influence.:218-228.
- Reddy, K. R., and R. D. DeLaune. 2008. Biogeochemistry of wetlands: science and applications. CRC press.
- Reed, D. J. 1995. The response of coastal marshes to sea-level rise: Survival or submergence? *Earth Surface Processes and Landforms* **20**:39-48.
- Roberts, H. H. 1997. Dynamic changes of the Holocene Mississippi River delta plain: the delta cycle. *Journal of Coastal Research*:605-627.
- Rovira, P., and V. R. Vallejo. 2002. Labile and recalcitrant pools of carbon and nitrogen in organic matter decomposing at different depths in soil: an acid hydrolysis approach. *Geoderma* **107**:109-141.
- Rovira, P., and V. R. Vallejo. 2003. Physical protection and biochemical quality of organic matter in mediterranean calcareous forest soils: a density fractionation approach. *Soil Biology and Biochemistry* **35**:245-261.
- Schoolmaster, D. R., C. L. Stagg, L. A. Sharp, T. E. McGinnis, B. Wood, and S. C. Piazza. 2018. Vegetation Cover, Tidal Amplitude and Land Area Predict Short-Term Marsh Vulnerability in Coastal Louisiana. *Ecosystems* **21**:1335-1347.
- Shrull, S. B. 2018. Spatial Trends and Variability of Marsh Accretion Rates in Barataria Basin, Louisiana, USA Using 210Pb and 137Cs Radiochemistry.
- Silveira, M., N. Comerford, K. Reddy, W. Cooper, and H. El-Rifai. 2008. Characterization of soil organic carbon pools by acid hydrolysis. *Geoderma* **144**:405-414.
- Smith, B. N., and S. Epstein. 1971. Two categories of 13C/12C ratios for higher plants. *Plant physiology* **47**:380-384.
- Smith, C., R. DeLaune, and W. Patrick Jr. 1983. Carbon dioxide emission and carbon accumulation in coastal wetlands. *Estuarine, Coastal and Shelf Science* **17**:21-29.
- Smith, K. E. 2012. Paleoecological study of coastal marsh in the Chenier Plain, Louisiana: Investigating the diatom composition of hurricane-deposited sediments and a diatom-based quantitative reconstruction of sea-level characteristics. University of Florida.
- Snedden, G. A. 2018. Soil properties, soil radioisotope activity, and end-of-season belowground biomass across Barataria basin wetlands U.S. Geological Survey data release.
- Stagg, C. L., M. M. Baustian, C. L. Perry, T. J. Carruthers, and C. T. Hall. 2018. Direct and indirect controls on organic matter decomposition in four coastal wetland communities along a landscape salinity gradient. *Journal of Ecology* **106**:655-670.

- Stagg, C. L., D. R. Schoolmaster, K. W. Krauss, N. Cormier, and W. H. Conner. 2017a. Causal mechanisms of soil organic matter decomposition: deconstructing salinity and flooding impacts in coastal wetlands. *Ecology* **98**:2003-2018.
- Stagg, C. L., D. R. Schoolmaster, S. C. Piazza, G. Snedden, G. D. Steyer, C. J. Fischenich, and R. W. McComas. 2017b. A landscape-scale assessment of above-and belowground primary production in coastal wetlands: implications for climate change-induced community shifts. *Estuaries and Coasts* **40**:856-879.
- Stewart, C. E., K. Paustian, R. T. Conant, A. F. Plante, and J. Six. 2007. Soil carbon saturation: concept, evidence and evaluation. *Biogeochemistry* **86**:19-31.
- Sullivan, M. J., and C. A. Currin. 2002. Community structure and functional dynamics of benthic microalgae in salt marshes. Pages 81-106 *Concepts and controversies in tidal marsh ecology*. Springer.
- Sullivan, M. J., and C. A. Moncreiff. 1990. Edaphic algae are an important component of salt marsh food-webs: evidence from multiple stable isotope analyses. *Marine ecology progress series*. Oldendorf **62**:149-159.
- Turner, R. E. 1997. Wetland loss in the northern Gulf of Mexico: multiple working hypotheses. *Estuaries* **20**:1-13.
- Twilley, R. R., S. J. Bentley, Q. Chen, D. A. Edmonds, S. C. Hagen, N. S.-N. Lam, C. S. Willson, K. Xu, D. Braud, and R. H. Peele. 2016. Co-evolution of wetland landscapes, flooding, and human settlement in the Mississippi River Delta Plain. *Sustainability Science* **11**:711-731.
- Unger, V., T. Elsey-Quirk, C. Sommerfield, and D. Velinsky. 2016. Stability of organic carbon accumulating in *Spartina alterniflora*-dominated salt marshes of the Mid-Atlantic US. *Estuarine, Coastal and Shelf Science* **182**:179-189.
- Van de Broek, M., C. Vandendriessche, D. Poppelmonde, R. Merckx, S. Temmerman, and G. Govers. 2018. Long-term organic carbon sequestration in tidal marsh sediments is dominated by old-aged allochthonous inputs in a macrotidal estuary. *Global Change Biology*.
- Ververis, C., K. Georghiou, D. Danielidis, D. Hatzinikolaou, P. Santas, R. Santas, and V. Corleti. 2007. Cellulose, hemicelluloses, lignin and ash content of some organic materials and their suitability for use as paper pulp supplements. *Bioresource Technology* **98**:296-301.
- Vogel, J. C. 1980. Fractionation of the carbon isotopes during photosynthesis. Pages 5-29 *Fractionation of the Carbon Isotopes during Photosynthesis*. Springer.
- Wang, J. J., S. K. Dodla, R. D. DeLaune, W. H. Hudnall, and R. L. Cook. 2011. Soil Carbon Characteristics in Two Mississippi River Deltaic Marshland Profiles. *Wetlands* **31**:157-166.

- Watzin, M. C., and J. G. Gosselink. 1992. The fragile fringe: coastal wetlands of the continental United States. Louisiana Sea Grant College Program.
- Weston, N. B., R. E. Dixon, and S. B. Joye. 2006. Ramifications of increased salinity in tidal freshwater sediments: Geochemistry and microbial pathways of organic matter mineralization. *Journal of Geophysical Research: Biogeosciences* **111**.
- White, D. A., and M. J. Simmons. 1988. Productivity of the marshes at the mouth of the Pearl River, Louisiana. *Castanea*:215-224.
- Williams, S. J. 1995. Louisiana coastal wetlands: A resource at risk. US Geological Survey.
- Xu, J., H. Cheng, W. Koskinen, and J. Molina. 1997. Characterization of potentially bioreactive soil organic carbon and nitrogen by acid hydrolysis. *Nutrient Cycling in Agroecosystems* **49**:267-271.
- Zedler, J. B. 1980. Algal mat productivity: comparisons in a salt marsh. *Estuaries* **3**:122-131.



## VITA

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