



Sources and chemical stability of soil organic carbon in natural and created coastal marshes of Louisiana

Megan Kelsall^{a,b,*}, Tracy Quirk^a, Carol Wilson^c, Gregg A. Snedden^d

^a Department of Oceanography and Coastal Sciences, School of the Coast and Environment, Louisiana State University, Baton Rouge, LA 70803, USA

^b PRIME AE Group Inc., 5521 Research Park Drive, Suite 300, Baltimore, MD 21228, USA

^c Department of Geology and Geophysics, Louisiana State University, Baton Rouge, LA 70803, USA

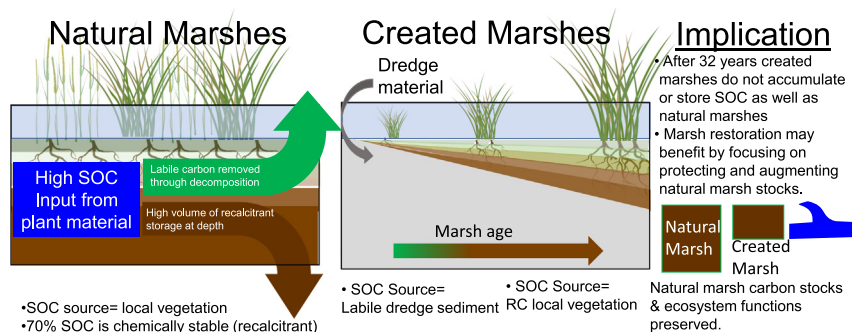
^d U.S. Geological Survey, Wetland and Aquatic Research Center, Baton Rouge, LA 70803, USA



HIGHLIGHTS

- Soil carbon source reflected local vegetation in natural marshes.
- All marsh salinities had similar labile and recalcitrant carbon accumulation rates.
- Recalcitrant carbon was the dominant form of soil organic carbon in all marshes.
- Created marsh soil carbon became increasingly recalcitrant with age up to 32 years.
- Soil carbon accumulation rates in created marshes were lower than natural marshes.

GRAPHICAL ABSTRACT



ARTICLE INFO

Editor: Fang Wang

Keywords:

Recalcitrant carbon
Labile carbon
Created marshes
Salinity gradient
Carbon source
Gulf coastal wetlands

ABSTRACT

Coastal marshes are globally important for sequestering carbon, yet sea-level rise and anthropogenic stressors can reduce their capacity as carbon sinks. Marsh restoration can offset a portion of carbon loss through the degradation of natural marshes, but potential differences in the sources and stability of soil organic carbon (SOC) between created and natural marshes may affect their function as a long-term carbon sink. Here, we examine the sources and chemical stability of SOC in natural and created marshes across the Gulf coast of Louisiana, USA. Marshes were examined along an estuarine salinity gradient in a former interdistributary basin of the Mississippi River Delta and in six created marshes across a 32-year chronosequence and a natural reference marsh ($n = 6$) in the Chenier Plain. Carbon source was assessed using $\delta^{13}\text{C}$ analysis and chemical stability was determined through an acid hydrolysis digestion that removed labile carbon (LC). Soil $\delta^{13}\text{C}$ values suggested that the local vegetation dominated SOC in all natural marshes although brackish marshes had a mix of sources and degradation of SOC. Recalcitrant carbon (RC) was $72.2 \pm 0.5\%$ of SOC across fresh, brackish and saline marshes. The depth-averaged RC accumulation rate was almost three times greater than LC accumulation rate, yet both contributed significantly to accretion and long-term SOC accumulation ($124\text{--}132\text{ g m}^{-2}\text{ y}^{-1}$ in natural marshes). RC and SOC accumulation rate increased with mineral sediment accumulation rate. For the created marshes, SOC became increasingly recalcitrant due to an increase in in-situ plant inputs, but accumulation rates were lower than the natural marshes. Overall, this study illustrates that natural marshes have a large stock of RC from the vegetation while dredge sediment created marshes have no plant-derived carbon initially, which accumulates slowly thereafter. Restoration practices may be improved by preserving and augmenting these deteriorating but carbon-rich natural marshes.

* Corresponding author at: PRIME AE Group Inc., 5521 Research Park Drive, Suite 300, Baltimore, MD 21228, USA.
E-mail address: mkelsall@primeeng.com (M. Kelsall).

<http://dx.doi.org/10.1016/j.scitotenv.2023.161415>

Received 22 September 2022; Received in revised form 28 December 2022; Accepted 2 January 2023

Available online 5 January 2023

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1. Introduction

Approximately half of global soil carbon burial occurs in blue carbon ecosystems, such as 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 have limited soil carbon sequestration capacity (Stewart et al., 2007), coastal wetlands accumulate OM and mineral sediment with sea level rise (Reed, 1995), such that soil organic carbon (SOC) storage capacity can increase over time (Chmura et al., 2003). However, sea level rise, climate change and anthropogenic stressors are causing changes in spatial cover of fresh, brackish, and salt marshes (Torio and Chmura, 2013), and overall loss of marsh area that will impact ecosystem functions including long-term carbon storage (Herbert et al., 2018). Marsh restoration and creation have the potential to mitigate some of the lost carbon sequestration capacity (Craft et al., 1999; Craft et al., 2003; Burden et al., 2013), yet “new” carbon that accumulates in created marshes may differ in source and stability (i.e., recalcitrance) from that in natural marshes with implications for long-term carbon storage.

Organic carbon in marsh soil originates from local marsh macrophytes and algae as well as from deposits of phytoplankton, terrestrial organic matter, and previously eroded marsh carbon (Van de Broek et al., 2018; Chmura et al., 1987; Chmura and Aharon, 1995; Craft et al., 1988; DeLaune and Lindau, 1987; Hopkinson et al., 2018). While process-based models of carbon accumulation generally assume that all of the soil carbon is produced in situ by marsh plants, $\delta^{13}\text{C}$ signatures indicate that marsh soil is comprised of a mix of carbon sources including algal, local vegetation, and allochthonous carbon deposited on the marsh surface (Ember et al., 1987; Gebrehiwet et al., 2008; Drexler et al., 2020; Leorri et al., 2018). In some coastal wetlands, allochthonous deposits can dominate the SOC pool, leading to overestimation of the carbon sequestration capacity and offsets for carbon emissions (Leorri et al., 2018; Van de Broek et al., 2018). The relative abundance of SOC may also vary along estuarine salinity gradients from freshwater to salt marshes due to differences in plant species litter quality (e.g., litter: nitrogen ratio), SOC stabilization mechanisms, and relative contribution of fresh locally-produced OM vs. deposited- which may be highly recalcitrant as it has already been degraded elsewhere (Stagg et al., 2017; Van de Broek et al., 2018). Few studies have examined the relative contribution of soil carbon sources in marsh restoration sites (except see Drexler et al., 2020), despite the emerging importance for calculating carbon offsets for restoration. For example, natural marsh soils of the Nisqually River Delta in Washington were dominated by autochthonous plant carbon while restoration sites had predominately allochthonous sources of deposited carbon (Drexler et al., 2020) implying that greater wetland land area would need to be restored to offset emissions. The dominant carbon sources may also shift with marsh age in restoration sites with an increase in the influence of local vegetation over time (Anderson et al., 2005; Craft et al., 2003; Muench and Elsey-Quirk, 2019).

Carbon stability may also differ between natural and created marsh soils and may change over time corresponding to changes in the carbon source and marsh age. Recalcitrant carbon (RC), is abundant in grasses such as *Spartina* spp., include lignin, waxes, fats, and resins that are more resistant to decay (Trumbore et al., 1990; Rovira and Vallejo, 2002; Dungait et al., 2012). Labile carbon (LC) compounds, abundant in algae and present in plants and root exudates, are easily consumed by microbes and include simple sugars, proteins, amino acids, and polysaccharides. Numerical models have shown that RC is vital to long term carbon storage in coastal marshes (Mudd et al., 2009), however, studies indicate variability in the chemical stability (LC:RC ratios) of marsh soil carbon (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; McClellan et al., 2021). This is not surprising, as anaerobic conditions as well as physiochemical interactions with mineral particles can limit oxidation and preserve LC (e.g., Rovira and Vallejo, 2003; Lehmann et al., 2007). For example, in salt marshes of the mid-Atlantic, United States, LC accumulation rate ranged between 37 and

238 $\text{g m}^{-2} \text{y}^{-1}$ and accounted for much of the variability in total SOC accumulation rates (Unger et al., 2016). Further, LC accumulation rate was positively related to mineral sediment accumulation rate suggesting that LC was deposited contemporaneously with mineral sediment and/or sedimentation led to LC preservation due to anaerobic conditions and/or physiochemical protection by mineral interactions (Unger et al., 2016; Rovira and Vallejo, 2003). Distinguishing SOC sources and their relative stability is becoming increasingly important as coastal wetland restoration is the focus of the first global greenhouse gas offset market where carbon credits are purchased in support of wetland restoration in return for offsets (e.g., one carbon credit/offset = one metric ton of carbon dioxide). The limited information on drivers of SOC in coastal wetlands is a barrier to fully developing a market for climate mitigation (Dencer-Brown et al., 2022).

The purpose of this study is to examine the sources and chemical stability of SOC in natural and created marshes along the Gulf Coast of Louisiana. We hypothesize that: (1) in natural marshes the majority of SOC will originate from the local vegetation with recalcitrant SOC increasing from freshwater to saline marshes due to a decrease in litter quality; (2) in created marshes, SOC source will shift from algal to plant becoming more recalcitrant with age; and (3) LC accumulation, although a smaller fraction, will increase with mineral sediment input (sensu Dodla et al., 2012; Unger et al., 2016).

2. Methods

2.1. Study locations

This study spans two coastal regions of Louisiana— the Mississippi River Deltaic Plain and the Chenier Plain (Fig. 1). The deltaic plain of Louisiana formed during the Holocene as the Mississippi River emptied in the Gulf of Mexico through a series of delta cycles initiated by the Mississippi River switching courses (Coleman et al., 1998). Wetlands of the Chenier Plain region in western Louisiana also formed during the Holocene from longshore transport and deposition of Mississippi River sediment (Jankowski et al., 2017, and references therein). A series of high and low sediment periods resulted in ridges ranging in age from <300 to 2800 years old (Byrne et al., 1959; Gould, 1959), which act as a partial hydrologic barrier to the north. Although these marshes have been present for thousands of years, natural and anthropogenic stressors are causing them to become more saline or convert to open water. Human activities have modified wetland processes in both the Barataria Basin Delta and Chenier Plain regions of Louisiana. Approximately 3000 km of levees along the Mississippi River prevents the river from changing course and has reduced natural overbank sedimentation by approximately 90 % since 1850 (Kesel, 2003). Marshes of Barataria Basin and the Chenier Plain are experiencing increasing flooding and saltwater intrusion resulting in conversion to open water due to a combination of factors including: a reduction in sediment deposits, canal installation (DeLaune et al., 1983; Sasser et al., 1986), subsurface fluid withdrawal leading to rapid land-surface subsidence (Bernier et al., 2011; Morton et al., 2002), and hurricanes (Barras, 2005). In an effort to reduce wetland loss in the Chenier Plain, the Army Corps of Engineers and Sabine National Wildlife Refuge began creating marshes in 1983 using sediment from the maintenance dredging of the Calcasieu Ship Channel. Created marshes were formed by hydraulically pumping dredge material into open water areas bounded by low levees, which were eventually breached or removed. Salt and brackish marsh vegetation naturally colonized the marshes with a vegetation cover similar to natural reference marshes generally within five years (Edwards and Proffitt, 2003).

Soil cores were collected from natural marshes of Barataria Basin in the deltaic plain along a salinity gradient in freshwater, brackish, and salt marshes and in created and natural brackish marshes of the Chenier Plain. The six created marsh were chosen because they represent a 32-year age sequence in close proximity. The average salinity for the fresh, brackish and saline marshes were 0.5, 5.8, and 11.0 ppt respectively according to CRMS data for the sites for the last 10 years prior to soil

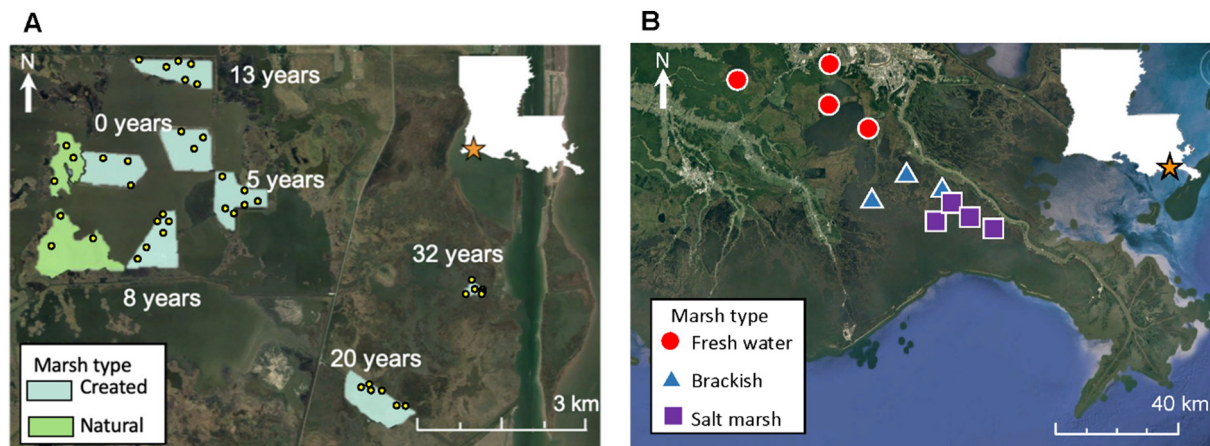


Fig. 1. Map of soil core locations in the created and natural brackish marshes of the Chenier Plain (A) and in freshwater, brackish, and salt marshes in the Barataria Basin, deltaic plain of Louisiana, United States. Yellow and black circles are core locations in the Chenier Plain ($n = 6$ per marsh) and red circles, blue triangles and purple squares are core locations in freshwater ($n = 4$), brackish ($n = 3$), and salt marshes ($n = 4$), respectively, within Barataria Basin.

collection. In the created marshes salinity was 15 ppt in the 5, 8 and 13 year old marsh, 22 ppt in the 32 year old marsh and 11 ppt in the NR marsh, no data was available for the 20-year-old marsh (Abbott et al., 2019).

2.2. Field sampling

Barataria Basin- Soil cores were collected in freshwater ($n = 4$), brackish ($n = 3$), and salt ($n = 4$) marshes in Barataria Basin in the spring and fall of 2016. One site was located near the Davis Pond Freshwater Diversion, installed in 2002. The diversion is an outlet to the Mississippi River that promotes ecological health by reintroducing the flooding that occurred before the levees were built, thus reintroducing nutrients, sediment and freshwater to these wetlands (DeLaune et al., 2013). The freshwater marshes were the most diverse with species richness ranging between 11 and 25 species, followed by brackish marshes with 7–9 species and salt marshes with only 3–5 species. Core locations varied in distance from the closest water body (bay, channel, or freshwater diversion) from 0 to 2000 m from the shoreline (Shrull, 2018; Supplementary Table S1). Soil properties, radiometric dating methodology, accretion rates and results are reported in Shrull (2018) and Snedden (2018). Briefly, cores were extracted using an aluminum push core device 60.8 cm long. Soil cores were 10.16 cm in diameter and a minimum of 35 cm deep. Following collection, cores were frozen to ensure no internal mixing or mold growth, and that the stratigraphy remained intact. Aboveground plant biomass was measured between August and October 2016 in three 0.25 m² plots per core location (33 plots total) within Barataria Basin (Snedden, 2018).

Chenier Plain- Six created marshes aged 0, 5, 8, 13, 20 and 32 y and a nearby natural reference (NR) marsh were sampled. The two oldest marshes, created in 1983 and 1996, were higher in elevation by 20–35 cm and adjacent to Calcasieu Ship Channel east of Highway 27 (which runs north to south, parallel to the Calcasieu Ship Channel) allowing unimpeded diurnal tidal flushing (Abbott et al., 2019). The younger created marshes and the NR marshes were west of Highway 27 and water control structures in Hog Island Gully, which dampen tidal flushing to approximately 25 % of tides experienced in the ship channel (Abbott et al., 2019). Six soil cores were collected to 35 cm depth in each of the six created marshes and adjacent NR marshes in spring 2016 (Fig. 1; Abbott et al., 2019). The dredge material was distinguished from marsh peat accumulation by its light gray color and high bulk density. The 0-year-old marsh contained no vegetation and 100 % dredge material with negligible marsh carbon accumulation. Each soil core was randomly located in the marsh interior and hand-extracted using a 35 cm polycarbonate tube with a diameter of 6.35 cm with a sharpened bottom edge to minimize compaction. Cores were transported back to the lab horizontally and stored cold until processed. In

August 2016, aboveground biomass was collected in six plots adjacent to soil core collection locations in each marsh. Quadrat size was dependent on species, 0.01 m² was used for *Spartina patens* and *Distichlis spicata* dominated plots, which have very high densities, 0.1 m² for *Spartina alterniflora* and 0.25 m² for *Bolboschoenus robustus* dominated plots; all were normalized to 1.0 m² for calculations (reported in Abbott et al., 2019).

2.3. Soil analysis

All soil cores were sliced into 2 cm depth intervals and processed for soil bulk density, percent loss on ignition (LOI), and SOC measurements (reported in Shrull, 2018; Abbott et al., 2019; Table S2). Briefly, each soil depth was dried to a constant weight at 60 °C for soil bulk density (Cheng et al., 2007; Gao et al., 2016); a subsample was combusted at 550–560 °C for 4–5 h for loss on ignition (LOI; Heiri et al., 2001), and a subsample was analyzed for total SOC concentration using a Costech 1040 CHNOS Elemental Combustion System. This subsample was fumed with 12 M HCl for 6 h prior to measurement to remove carbonates (Harris et al., 2001). The mineral density was determined by the mass remaining after LOI, which was used to calculate the mineral accumulation rate similarly to carbon accumulation rates described below.

2.3.1. Carbon sources

$\delta^{13}\text{C}$ was measured on both the bulk SOC sample and the post-hydrolysis recalcitrant sample (see section 2.3.3 below) using an elemental analyzer interfaced to a continuous flow isotope ratio mass spectrometer (IRMS) by University of California, Davis Laboratory. The long term standard deviation for ^{13}C samples is 0.2 ‰.

An isotope mixing model used by Chmura et al. (1987) was used to predict a range of soil $\delta^{13}\text{C}$ values based on present vegetation. Isotope values for vegetation were obtained from Chmura and Aharon (1995), Chmura et al. (1987) and Smith and Epstein (1971). Species representation was estimated using aboveground biomass for the Chenier Plain marshes and percent cover for the Barataria Basin marshes obtained from CPRA (2019). In the 20 and 32-year-old created marsh and the NR marsh, there were species for which reported $\delta^{13}\text{C}$ values were unavailable. In those cases, an estimate based on maximum and minimum $\delta^{13}\text{C}$ values for the photosynthetic pathway (C_3 vs C_4) was used (Smith and Epstein, 1971). The freshwater marshes were diverse without a dominant species present, and literature values for $\delta^{13}\text{C}$ could not be found for a majority of species. It was assumed that these freshwater species were the typical range of C_3 species which is supported by DeLaune (1986), Chmura et al. (1987)

and Chmura and Aharon (1995). A simple weighted mixing model used for the predicted soil $\delta^{13}\text{C}$ value based on the vegetation present was:

Predicted $\delta^{13}\text{C}$ soil

$$= \frac{\sum_{i=1}^n (\% \text{aboveground biomass } i \text{ or } \% \text{cover } i) (\delta^{13}\text{C})}{\sum_{i=1}^n (\% \text{aboveground biomass or } \% \text{cover})}$$

2.3.2. Carbon stability

Labile and recalcitrant SOC fractions (LC and RC, respectively) were measured every other depth interval down to 35 cm for natural marsh cores and for created marshes to the depth of the dredge sediment horizon which was distinguishable by the light color from the organic matter. RC was isolated using acid hydrolysis digestion methods described by Unger et al. (2016). Briefly, 10 mL of deionized water and 20 mL of 6 M HCl was added to 1.0 g of dry soil in a glass vial. Samples were then placed in a digestion block for 18 h at 150 °C. Following hydrolysis, samples were centrifuged at 2500 rpm for 10 min and the supernatant immediately decanted. The sample was then dried at 60 °C until a constant weight was reached. Both the bulk sample (pre-hydrolysis) and residual fraction (post-hydrolysis) were analyzed for elemental carbon using the same procedure described above for SOC. The fraction of recalcitrant SOC was determined from the formula:

$$\begin{aligned} \frac{\text{RC}}{\text{SOC}} &= \frac{\text{C post-hydrolysis (g)}}{\text{total SOC (g)}} \\ &= \frac{\text{soil mass post-hydrolysis (g)} \times (\text{post-hydrolysis C}\%)}{100} \\ &= \frac{\text{soil mass pre-hydrolysis (g)} \times \left(\frac{\text{total SOC (\%)}}{100} \right)}{100} \end{aligned}$$

The fraction of labile SOC was calculated as the difference between total carbon and the RC fraction.

The percent of the RC in soil mass was calculated using the formula:

$$\begin{aligned} \text{RC}\% &= \frac{\text{C mass post-hydrolysis (g)}}{\text{soil mass pre-hydrolysis (g)}} \\ &= \frac{\text{soil mass post-hydrolysis (g)} \times (\text{post-hydrolysis C}\%)}{100} \\ &= \frac{\text{soil mass pre-hydrolysis (g)}}{\text{soil mass pre-hydrolysis (g)}} \end{aligned}$$

The percent of the LC in soil mass was calculated as the difference between total SOC% and RC%.

A strong linear relationship between SOC and LC or RC concentration ($R^2 > 0.62$; Fig. S1), allowed us to interpolate RC and LC values for alternate depth intervals not analyzed directly using the equation of best fit for each estuary. Carbon density was calculated by multiplying the carbon concentration by the bulk density for LC, RC and SOC. Accretion rates previously calculated and reported in Shrull (2018) and Abbott et al. (2019) were used for new calculations of LC and RC, and mineral accumulation rates here. Accumulation rates for each carbon fraction were calculated using the equation below:

$$\varphi = \frac{\sum_i (C \times d_i) \times 10^4}{a}$$

where φ represents the carbon (labile or refractory) accumulation rates ($\text{g m}^{-2} \text{y}^{-1}$), C represents carbon or mineral density (g cm^{-3}), d_i represents depth (cm), and a represents the age of the marsh (created marshes) or number of years since the ^{137}Cs -peak (natural marshes). Carbon stocks are the sum of carbon (g m^{-2}) to the same depth of either the dredge material or the ^{137}Cs -peak in natural marshes. In the NR marshes of the Chenier Plain, the ^{137}Cs peak was not evident (Abbott et al., 2019) and therefore regional accretion and SOC accumulation rates from Smith (2012) were used, and LC and RC accumulation rates were calculated based on the average percentages of LC and RC of our NR marshes.

2.4. Data analysis

Data were transformed either by log, logit, square root, cubic or quadratic root when necessary to improve data normality and homogeneity of variance. A nested one-way Analysis of Variance (ANOVA) was used to account for the effect of the estuary (Chenier Plain or Barataria Basin), marsh type (freshwater, brackish, and salt marsh), and marsh age on LC, RC and SOC, density, accumulation rate, and $\delta^{13}\text{C}$ (bulk and recalcitrant $\delta^{13}\text{C}$ were run together to test for differences before and after hydrolysis). ANOVA was used to test the difference among marshes in biomass categories including aboveground biomass and stem density. Tukey's Honestly Significant Difference test was used for post-hoc multiple comparisons.

Regression analyses were used to examine the relationships between response variables (LC, RC, and SOC density, stock, and accumulation rate), against predictor variables (accretion rate, aboveground biomass, and mineral sediment density, stock, and accumulation rate). The relationship between SOC, RC, and LC stocks and mineral stocks and marsh accretion rates were analyzed using regression analysis. All statistical analyses were conducted using JMP version 14.2 (SAS institute Inc).

3. Results

3.1. SOC and RC sources

The freshwater marshes in Barataria Basin were dominated by C_3 plants, including *Panicum hemitomon*, *Salix nigra*, and *Alternanthera philoxeroides*, which comprised at least 30 % cover at one or more of the sites (Table 1). Brackish and salt marshes were composed of a mix of C_3 and C_4 plants. *Schoenoplectus americanus* and *Juncus roemerianus*, both C_3 plants, accounted for 40 to 50 % cover in one of the three brackish marshes and three of the four salt marshes. The remaining marshes were dominated by C_4 vegetation, including *S. patens*, *S. alterniflora*, and *D. spicata*.

Freshwater marshes had bulk soil $\delta^{13}\text{C}$ values between -28 and -30 ‰ that were relatively uniform with depth (Fig. 2A). These values fell within the predicted range based on the C_3 species present and results of the mixing model indicating that the SOC is from the plants growing in the marsh (Fig. 3). Brackish and salt marsh soil $\delta^{13}\text{C}$ values were much higher and more variable with depth than those of fresh marshes likely due to greater mixing of organic matter from C_3 and C_4 species (Table 1; Fig. 2A). Fresh and salt marsh bulk $\delta^{13}\text{C}$ samples were all within range of the values predicted by the isotope mixing model, whereas approximately 75 % of the samples in the brackish marsh were below predicted values suggesting that decomposition or mixing of organic matter sources depleted the $\delta^{13}\text{C}$ signature (Fig. 3).

The brackish natural reference (NR) marsh in the Chenier Plain had a similar soil $\delta^{13}\text{C}$ signature to the saline marshes in Barataria Basin at the surface, but was more depleted with depth, closer to the $\delta^{13}\text{C}$ signature of freshwater marshes (Fig. 2B). Averaged across depths, the $\delta^{13}\text{C}$ signature of the NR fell within the predicted range, but the depletion with depth reflects a shift in the dominant source of carbon from C_3 fresh marsh plants to more C_4 salt tolerant plants (Figs. 2B and 3). The newly created marsh (i.e., age "0"), comprised of only dredge material, had a depleted $\delta^{13}\text{C}$ signature, lower than predicted from the *S. alterniflora* vegetation present (Figs. 2B and 3). This isotopically light dredge material became heavier from local organic matter inputs as marshes aged. Thus, older created marshes were more enriched at the surface and depleted with depth (Fig. 2B).

Following hydrolysis, the recalcitrant $\delta^{13}\text{C}$ soils were more depleted than the bulk soil signifying a higher concentration of lignin in the hydrolyzed sample (Fig. 3; Benner et al., 1987). Depletion of the recalcitrant sample relative to the bulk sample was similar across natural marshes and much greater in created marshes (Fig. 3). Young, created marshes had a greater depletion than older marshes due to the labile nature of carbon in the dredged material and RC that is increasingly plant-derived in older created marshes.

Table 1

Plant species cover and aboveground biomass in study locations of marshes in Barataria Basin and the Chenier Plain, Louisiana. Vegetation data in Barataria Basin was from the Coastwide Reference Monitoring System data (CPRA, 2019).

Basin	Marsh	Species	Plant Type	Percent cover range (%)	Total aboveground biomass (g m ⁻²)
Barataria Basin	Fresh	<i>Eleocharis macrostachya</i>	C ₃	18–20	1000 ± 112
		<i>Sagittaria lancifolia</i>	C ₃	18–29	
		<i>Panicum hemitomon</i>	C ₃	64–69	
		<i>Baccharis halimifolia</i>	C ₃	12	
		<i>Mikania scandens</i>	C ₃	14	
		<i>Polygonum punctatum Elliott</i>	C ₃	15–17	
		<i>Colocasia esculenta</i>	C ₃	24	
		<i>Salix nigra Marshall</i>	C ₃	35	
		<i>Typha latifolia</i>	C ₃	13	
		<i>Alternanthera philoxeroides</i>	C ₃	30–35	
	Brackish	<i>Spartina patens</i>	C ₄	27–56	1731 ± 253
		<i>Schoenoplectus americanus</i>	C ₃	39	
		<i>Spartina alterniflora</i>	C ₄	11	
		<i>Distichlis spicata</i>	C ₄	12	
Salt	<i>Spartina alterniflora</i>	C ₄	26–90	1600 ± 188	
	<i>Juncus roemerianus</i>	C ₃	41–48		
Chenier Plain	0	<i>Spartina alterniflora</i>	C ₄	100	1193 ± 1067
	5	<i>Spartina alterniflora</i>	C ₄	0–100	2535 ± 457
		<i>Distichlis spicata</i>	C ₄	0–100	
	8	<i>Spartina alterniflora</i>	C ₄	100	1470 ± 363
	13	<i>Distichlis spicata</i>	C ₄	98–100	2102 ± 230
		<i>Spartina alterniflora</i>	C ₄	100	
	20	<i>Distichlis spicata</i>	C ₄	52–93	1508 ± 82
		<i>Spartina patens</i>	C ₄	26	
		<i>Borrichia frutescens</i>	C ₃	22	
	32	<i>Spartina alterniflora</i>	C ₄	100	1774 ± 335
		<i>Distichlis spicata</i>	C ₄	41–62	
		<i>Spartina patens</i>	C ₄	15–49	
		<i>Borrichia frutescens</i>	C ₃	10	
	NR	<i>Distichlis spicata</i>	C ₄	23–59	1489 ± 338
<i>Bolboschoenus robustus</i>		C ₃	40–76		

3.2. SOC stability and accumulation rates

3.2.1. SOC concentration

Across marshes, RC and LC comprised an average of 73 and 27 % of SOC, respectively. SOC was more labile in the created marshes than in the NR marshes of the Chenier Plain (32 ± 2 vs. 24 ± 1 %; $p < 0.01$) but not significantly different from the natural marshes in Barataria Basin. Interestingly, LC increased with SOC concentration following a quadratic function in both created and natural Chenier Plain marshes while LC and RC increased linearly with an increase in SOC in marshes of Barataria Basin (Fig. S2 A, B). At very low carbon concentration, the 0-year-old marsh, comprised completely of dredge material, had a high percentage of LC which declined sharply with marsh age as organic carbon content increased. At SOC concentration between ~10 and 25 %, Chenier Plain NR marshes had more RC than all the Barataria Basin marshes yet when SOC concentrations were > 30 %, the carbon was more labile in Chenier Plain marshes (Fig. S2 C, D).

SOC concentrations ranged from an average of 1 % in the newly created marsh to 26 % in freshwater marshes in Barataria Basin (Table S3). The created marshes had lower SOC, RC, and LC concentrations than natural marshes and the freshwater marshes in Barataria Basin had significantly greater SOC, RC, and LC concentrations than the other marshes in this study (Table S3).

3.2.2. SOC density

SOC density ranged from an average of 9.5 mg cm⁻³ in the newly created marsh to 32.3 mg cm⁻³ in the NR marsh (Fig. 4; Table S3). Generally, the pattern in SOC density with depth followed the pattern of RC rather than LC density (Fig. 4). LC density was relatively low and uniform across depths, ranging from 0.4 to 17.6 mg cm⁻³. RC accounted for over 70 % of the total SOC density with similar patterns across Chenier Plain and Barataria Basin marshes (Fig. 5A). Averaged over the depth profile, SOC, RC, and LC density were greatest in the NR marshes in the Chenier Plain and freshwater marshes in Barataria Basin (Fig. 5A). SOC and RC density

were lower in the created marshes than in the natural marshes, but LC density in the 5- and 20-year-old created marshes were similar to brackish and salt marshes in Barataria Basin (Fig. 5A; Table S3).

3.2.3. SOC stocks

RC stocks averaged 4564 ± 358 g m⁻² in the upper 50 cm of natural marshes across both regions of Louisiana (Fig. 5B). RC stock above the dredge material in created marshes averaged much lower, 633 ± 88 g m⁻². RC increased with created marsh age from <200 g m⁻² in 5-year-old marshes to an average of 1518 g m⁻² in the 32-year-old marsh. This is equivalent to an increase of approximately 46 g RC m⁻² y⁻¹, similar to the calculated rates of RC accumulation within marshes (Fig. 5B). RC stocks comprised an average of 70 % of the total SOC stock, and LC stocks comprised about 30 %. This percentage was very consistent across marshes.

3.2.4. SOC accumulation rate

SOC accumulation rates ranged from an average of 52 g m⁻² y⁻¹ in the 5-year-old created marsh to 132 g m⁻² y⁻¹ in the Barataria Basin salt marsh (Fig. 5C, Table S3). SOC accumulation rates were significantly greater in the salt marsh and natural reference marshes than in the 5- and 20-year-old created marshes ($p < 0.0001$; Fig. 5C). The patterns were similar for RC accumulation rates. On average, RC accumulation rates in the created marshes were about 57 % of the total SOC accumulation. LC accumulation was also similar across marshes but was significantly lower, approximately half, in the 8-year-old marsh than in the NR marsh.

LC and RC stocks both contribute linearly to the SOC accumulation rate. For a given stock of LC or RC in the soil, SOC accumulation rates tended to be greater in Chenier Plain than Barataria Basin marshes especially where accumulation rates were high (Fig. S3), but the rate of increase of SOC accumulation unit of RC stock was similar for both basins. For example, for a LC stock of approximately 2000 SOC g m⁻², accumulation rates in the Chenier Plain natural marshes averaged 50 g m⁻² y⁻¹ greater than in Barataria Basin marshes (Fig. S3 A).

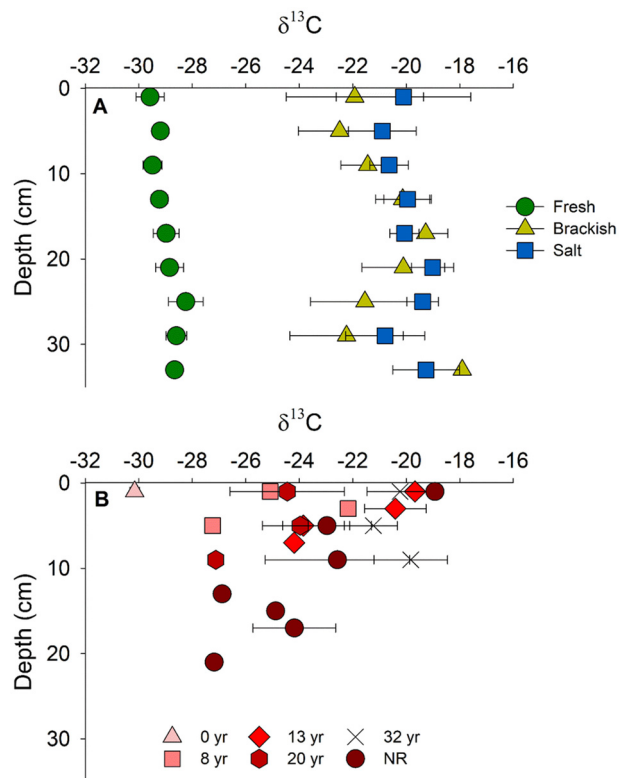


Fig. 2. $\delta^{13}\text{C}$ depth profiles for freshwater, brackish and salt marshes in Barataria Basin (A), and created and natural reference (NR) marshes of the Chenier Plain (B), Louisiana. Data points are means and error bars standard error. $\delta^{13}\text{C}$ was only measured in the organic matter for created marshes, and the 0-year-old marsh represents the dredge material. No samples were measured for the 5-year-old marsh.

3.3. Relationships to environmental factors and contribution to accretion

SOC, RC, and LC accumulation rates were not related to vegetation properties (i.e., aboveground biomass or stem height). In the natural marshes, both SOC and RC accumulation rates increased with an increasing mineral sediment accumulation rate (Fig. 6). LC accumulation rate was low and consistent ($35 \pm 3 \text{ g m}^{-2} \text{ y}^{-1}$) across a range of mineral sediment accumulation rates from 37 to $1803 \text{ g m}^{-2} \text{ y}^{-1}$. Interestingly, the freshwater diversion at Davis Pond introduced a high sediment load to one of the freshwater marshes ($1529 \text{ vs. } > 400 \text{ g m}^{-2} \text{ y}^{-1}$), yet rates of LC, RC, and SOC accumulation rates were similar to the other freshwater marshes studies in the Barataria Basin.

Marsh accretion rate was positively related to both SOC and mineral sediment stock across Barataria Basin marshes but not in the Chenier Plain marshes (Fig. 7). Both LC and RC stocks were positively related to accretion rate in the Barataria Basin (Fig. 7A). In the created marshes, carbon stocks were relatively low and variable. The mineral sediment component in the created marshes is partly dredge sediment from marsh creation that has been incorporated into the marsh peat through movement by macrofauna and tides therefore the sediment stock includes deposition as well as relict dredged material. Thus, organic and mineral contributions to accretion are decoupled across the 32-year time span of marsh creation.

4. Discussion

4.1. Recalcitrant plant carbon dominates soil carbon in the transgressive Mississippi deltaic marshes

Allochthonous deposits of organic matter can be a major source of soil carbon in some coastal wetlands (Leorri et al., 2018; Van de Broek et al., 2018; Saintilan et al., 2013). While in this study we cannot distinguish

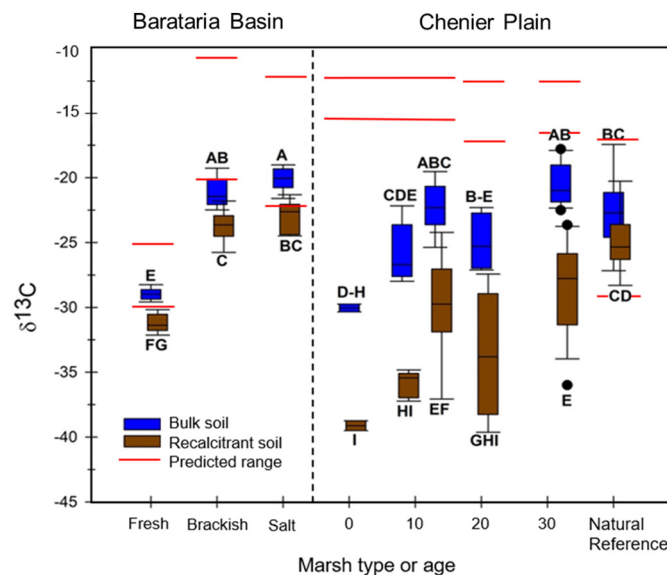


Fig. 3. Depth-averaged soil $\delta^{13}\text{C}$ values in freshwater, brackish, and salt marshes in Barataria Basin and created and natural marshes of the Chenier Plain pre- and post-hydrolysis. Boxes represent the 25 and 75 % quartile data, the middle bar represents the median, bars represent the range, and the dots represent outlier data. Values represented by the same letters are not significantly different ($p < 0.05$) based on Tukey's HSD post hoc test. An isotope mixing model used by Chmura et al. (1987) was used to predict high and low soil $\delta^{13}\text{C}$ values (shown in red) based on dominant vegetation. Isotope values for vegetation were obtained from Chmura et al. (1987), Chmura and Aharon (1995) and values not reported in this paper were found in Smith and Epstein (1971).

old, eroded marsh carbon from in situ produced carbon, ^{13}C signatures indicate natural marsh soil carbon along the Gulf coast of Louisiana is primarily derived from local vegetation and is highly recalcitrant. RC comprised a high and consistent percentage ($72.2 \pm 0.5 \%$) of SOC across marshes similar to the findings in other studies in Louisiana (Dodla et al., 2012; McClellan et al., 2021). Previous studies in the Barataria Basin, Louisiana have attributed much of the soil carbon to the local vegetation (DeLaune, 1986; Chmura et al., 1987), while eroded marsh carbon may be an additional source undetected by ^{13}C signatures. Here, we found that $\delta^{13}\text{C}$ signatures of RC were more depleted than non-hydrolyzed soil indicating that further decomposition and concentration of lignin is possible (Benner et al., 1987) with an average of 28 % labile carbon. As compared to other studies (e.g., Unger et al., 2016), LC was low and uniform across all marshes and with depth, thus controls on rates of marsh SOC accumulation are largely a function of controls of marsh plant productivity and degradation processes.

Carbon dynamics in marshes of the Mississippi Delta differ from that in U.S. mid-Atlantic estuaries. RC accumulation rates were higher in Louisiana natural marshes ($88 \text{ to } 98 \text{ g m}^{-2} \text{ y}^{-1}$) than in mid-Atlantic marshes ($63 \text{ to } 95 \text{ g m}^{-2} \text{ y}^{-1}$; Unger et al., 2016; Table S4). Greater marsh plant productivity (Turner, 1976; Kirwan et al., 2009) and lower litter quality (Zhang et al., 2021) at lower latitudes may contribute to greater RC. However, LC accumulation rates of $33 \text{ to } 36 \text{ g m}^{-2} \text{ y}^{-1}$ in this study were at the very low end of rates in mid-Atlantic marshes ($37 \text{ to } 238 \text{ g m}^{-2} \text{ y}^{-1}$; Unger et al., 2016). The LC accumulation rates in the mid-Atlantic were strongly associated with mineral sedimentation. Greater decomposition of LC at lower latitudes along with profound differences in estuary type (e.g., deltas and drowned estuaries) may be contributing to these differences in marsh soil carbon stability.

4.2. Carbon stability was similar across fresh, brackish and salt marshes

Contrary to our hypothesis that salt and brackish marshes have more RC compared to freshwater marshes, we found that the chemical stability of

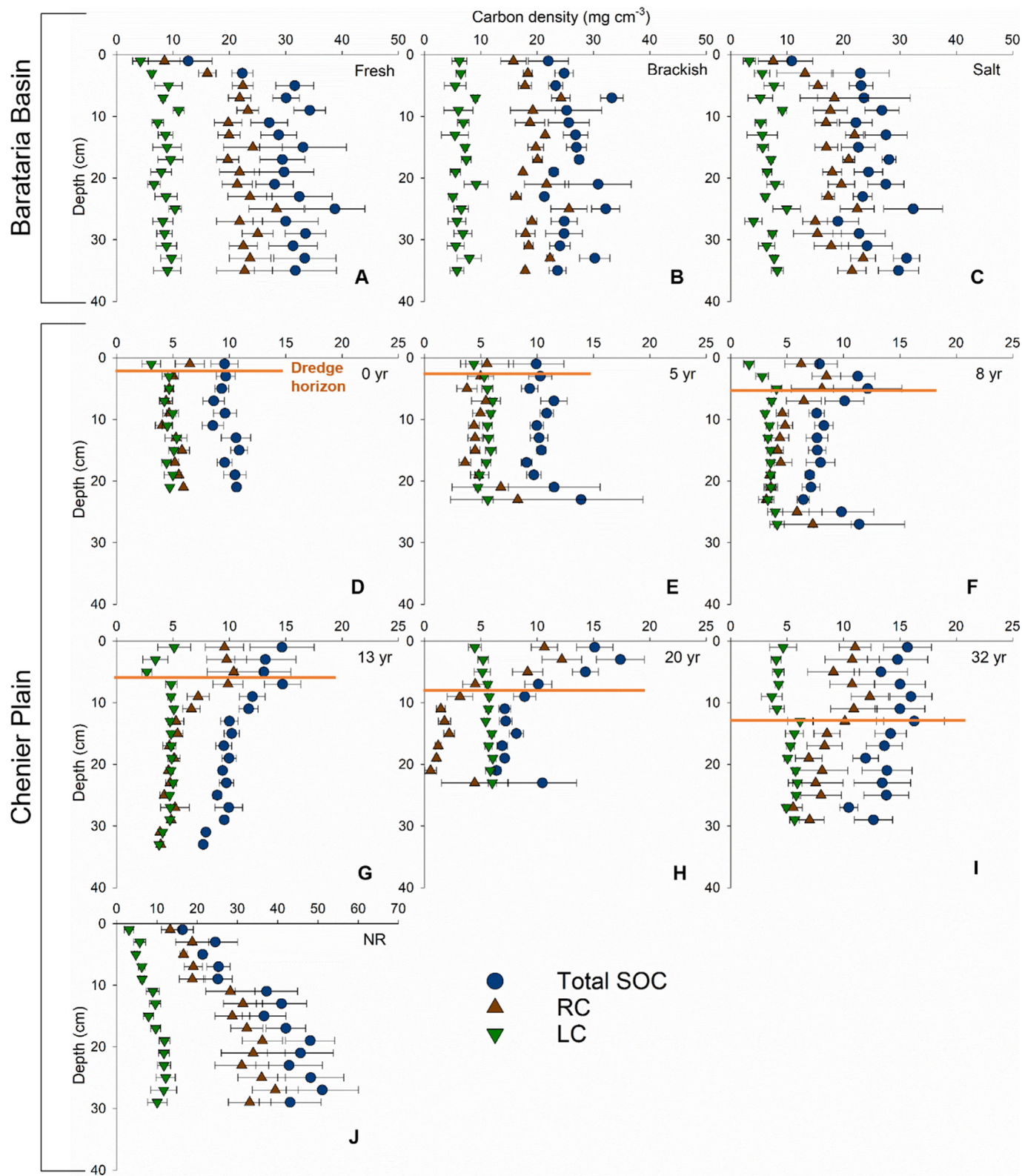


Fig. 4. Total, refractory, and labile soil organic carbon (SOC, RC, and LC, respectively) density profiles for fresh, brackish, and salt marshes in Barataria Basin and created and natural brackish marshes in the Chenier Plain region of Louisiana. Data points are the averages with standard error bars. The transition depth between marsh peat and dredge material (i.e., dredge horizon) in the created marsh is represented by an orange line. Note differences in x-axis scale.

SOC was similar among marsh types, despite distinct differences in the local vegetation. Freshwater marsh soils had $\delta^{13}\text{C}$ signatures that reflected C_3 plant species, whereas brackish and salt marshes had signatures dominated by C_4 plants. Freshwater marsh SOC concentrations were approximately

two times higher than in brackish and salt marshes. Additionally, the freshwater marshes had approximately half the mineral sediment per area of brackish and salt marshes (Shrull, 2018). Despite having lower mineral sediment inputs and lower soil bulk densities than salt marshes (Table S2), SOC

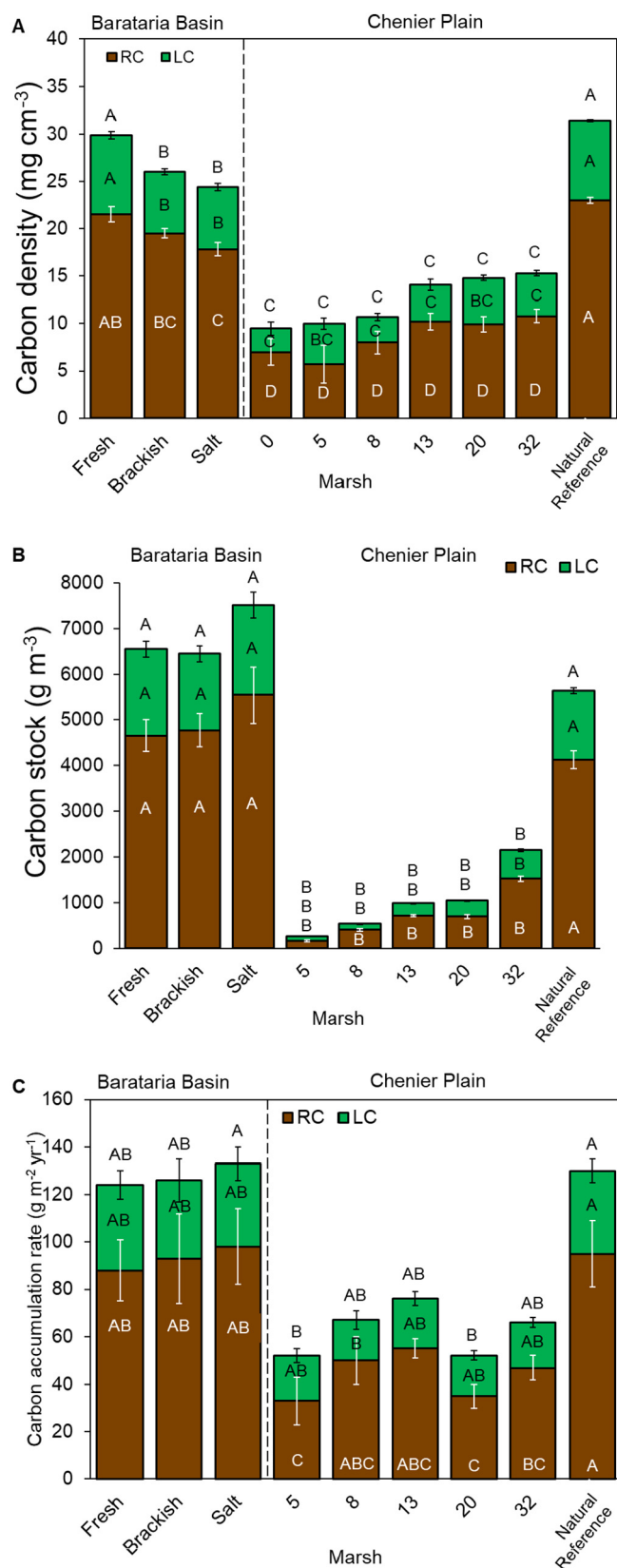


Fig. 5. Labile carbon (LC), and recalcitrant carbon (RC) density (A), carbon stocks (B), and accumulation rates (C) in marshes of Barataria Basin and created and natural marshes of the Chenier Plain. Values represented by the same letters are not significantly different ($p < 0.05$) by a nested ANOVA.

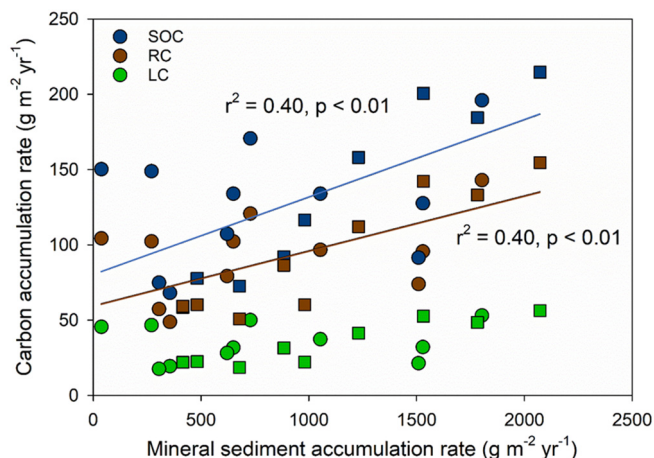


Fig. 6. Relationship between mineral sediment accumulation rate and LC, RC, and SOC accumulation rates in natural marshes of Louisiana. Circles represent natural marshes in Barataria Basin, and squares represent the NR marshes in the Chenier Plain. Data for created marshes are not shown as mineral sediment stocks are compounded by natural tidal sedimentation and anthropogenically dredged sediment placement. Trend lines are for RC and SOC accumulation rates, the LC accumulation rate against mineral sediment accumulation rate did not yield a significant result.

densities in this study were greater in freshwater than brackish and salt marshes (Table S3). Ultimately, accumulation rates of SOC, RC, and LC were similar across the salinity gradient and between marshes in the Barataria Basin and the Chenier Plain. This differs from the broader trend across the Gulf of Mexico, where SOC accumulation rates in freshwater marshes are about half of that in their respective salt marshes. Lower SOC accumulation in freshwater marshes was due to lower accretion rates because SOC densities were similar between the marshes (Craft, 2007). Across the Ogeechee, Altamaha, and Satilla river systems in Georgia, USA, SOC accumulation rates in freshwater marshes were about three times higher than in salt marshes attributed to lower decomposition rates in freshwater as compared to brackish and salt marshes where sulfate (a source of energy for microbial activity) is abundant (Craft, 2007; Loomis and Craft, 2010). In comparison to our study, these marshes had similar SOC accumulation in the freshwater marsh, however they had about 75 % of our SOC accumulation in brackish marshes, and 30 % of our SOC accumulation in salt marshes. Higher SOC accumulation in brackish and salt marshes in our study is likely associated with the higher mineral sedimentation in Gulf Coast marshes compared to southeast marshes (Craft, 2007), and the relationships found between carbon and mineral sediment in this study.

4.3. Soil carbon dynamics differ in the Deltaic and Chenier Plains

Brackish marshes in the Barataria Basin Delta and Chenier plains differed in $\delta^{13}\text{C}$ signatures with depth. Brackish marshes in the Deltaic Plain were comprised of a mix of C_3 and C_4 plants with subtle differences with depth from temporary changes in dominance such as a C_3 plant such as *Juncus* in an otherwise C_4 plant dominated marsh. In contrast, signatures of natural marshes in the Chenier Plain reflect a more recent shift from freshwater-intermediate salinity marshes to a mix of C_3 and C_4 brackish vegetation with species such as *D. spicata* and *B. robustus* being dominant (O'Neil, 1949). These marshes are now more saline, as shown by the shift in $\delta^{13}\text{C}$ signatures from C_3 signatures at depth to C_4 species toward the surface.

Carbon stability at similar levels of carbon concentration were different in the Deltaic and Chenier Plain marshes. Deltaic plain marshes had highly recalcitrant carbon especially when SOC content in the soil was low. In organic rich marsh soil where SOC concentration averaged $>30\%$, Chenier Plain marshes had more LC than Deltaic plain marshes. In the Chenier

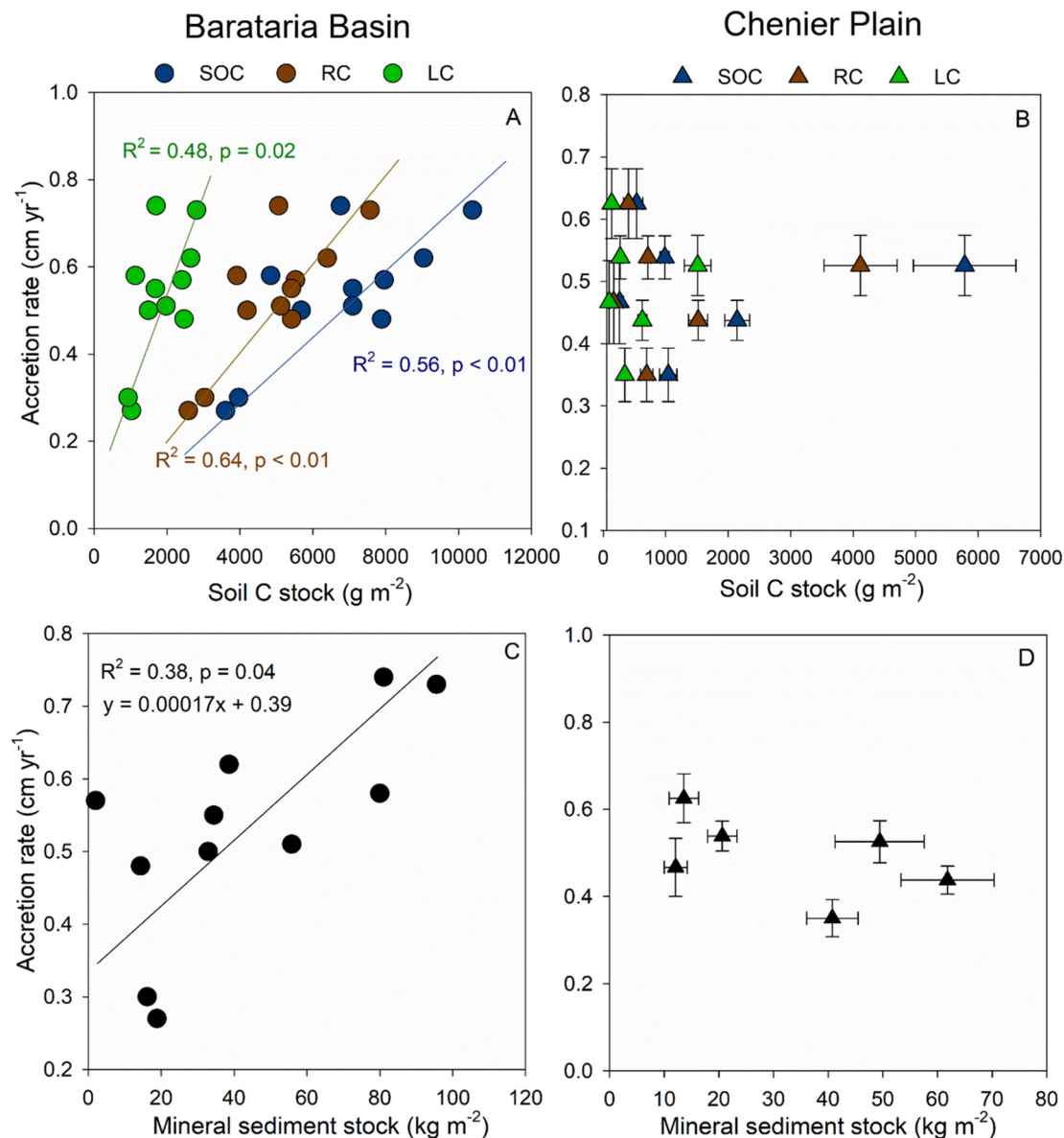


Fig. 7. Relationships between total, refractory, and labile soil carbon stocks, and accretion rates in Barataria Basin (A) and Chenier Plain (B) marshes. Relationships between mineral sediment stock and accretion rate in marshes of Barataria Basin (C) and the Chenier Plain (D). Marsh means and standard errors are shown for Chenier Plain marshes.

Plain, as the concentration of SOC increased from 0 to 5 % the carbon became drastically more recalcitrant (e.g., from 40 to 80 %). Sediment texture may play a role and differs between the two basins. Barataria Basin marshes have a higher silt content (~49 %) and Chenier Plain marshes sites have a high clay content (Abbott et al., 2019), with the dredge sediment being ~62 % clay (Edwards and Proffitt, 2003). LC binds with clay particles, which enhance preservation while carbon associated with silt tends to be of higher recalcitrance (Plante et al., 2006).

4.4. Soil carbon in created marshes becomes more recalcitrant with age associated with a shift to plant carbon

Young created marshes had low relatively labile SOC content that became more recalcitrant with small increases in plant-derived SOC content. The dredge material in the 0-year-old marsh had a low $\delta^{13}\text{C}$ signature, similar to that of land plants (-29 to -23 ‰) and terrestrial (-33 to -29 ‰) and marine petroleum (-30 to -21 ‰; Park and Epstein, 1961). Terrestrial and more likely, marine petroleum are predicted to be the dominant SOC sources in dredge material in these marshes, which was hydraulically

pumped from a major navigational channel where a mixture of oil spill hydrocarbons and vegetation detritus deposited from upriver likely deposits. Soil $\delta^{13}\text{C}$ increased with marsh age as the vegetation contributed increasingly more carbon to the soil pool. Depletion of the $\delta^{13}\text{C}$ signature in young marshes may be due to carbon from dredge material, algal, allochthonous carbon from previously eroded marsh material, as well as mixing with the new OM that is starting to accumulate (DeLaune, 1986; Chmura et al., 1987; Craft et al., 1988). Algal $\delta^{13}\text{C}$ values are within the range of the soil in the brackish and created marshes in this study (Smith and Epstein, 1971). However, algal production tends to be low in natural marshes in Louisiana (Sullivan and Currin, 2002) and may not have had a large influence on soil $\delta^{13}\text{C}$ values here (Chmura et al., 1987; Chmura and Aharon, 1995). In the oldest created marshes (20 and 32 years), soil carbon signatures are only about 4 ‰ depleted from predicted values, which is the amount of depletion linked to the accumulation of lignin associated with decomposition (Benner et al., 1987), indicating that the primary carbon source is the local vegetation that is undergoing decay processes in the soil. The difference between bulk and recalcitrant $\delta^{13}\text{C}$ decreased with marsh age as the soils became less dominated by labile dredge material and accumulated more recalcitrant plant-derived OM.

4.5. The role of mineral sedimentation in LC, RC, and SOC accumulation

Mineral sediments from rivers, primarily the Mississippi River, are the foundation of coastal marshes in Louisiana, yet today most of the marshes are isolated from direct river sediment input (Hatton et al., 1983; DeLaune and Pezeshki, 2003; Nyman et al., 1990). Thus, plants are the primary contributors to accretion and the contribution of mineral sediment is variable (Turner et al., 2002). The findings of this study show the importance of mineral sediment deposition to the accumulation of RC and SOC. Across natural marshes, 40 % of the variability in RC and SOC accumulation rate was explained by the rate of mineral sediment accumulation. The $\delta^{13}\text{C}$ data indicated that RC in natural marshes was derived from the local vegetation. Mineral sediment deposition increases RC inputs by stimulating an increase in plant productivity through a combination of reduction in anaerobic stress, introduced nutrients (DeLaune et al., 1981; DeLaune et al., 1990), and/or introduced iron (which buffers toxicity; King et al., 1982). Additionally, marshes with dense vegetation may also trap more mineral sediment as the vegetation slows water velocity encouraging mineral sediments to settle out (Mudd et al., 2010). However, at the outfall of the Davis Pond River Diversion, which supplied over four times the amount of mineral sediment to the freshwater marsh, carbon accumulation rates were similar to the other freshwater marshes. Furthermore, carbon pools of the sediment-rich marshes of Wax Lake Delta, which had 1.5 times greater mineral density were similar to the Chenier Plain sediment-limited created marshes (McClellan et al., 2021). However, marsh carbon accumulation and mineral sedimentation are less tightly and positively correlated (40 %) than in other locations (>89 %; Unger et al., 2016; Neubauer, 2008).

4.6. Contributions to accretion differ between natural marshes and created marshes

Carbon accumulation in the soil contributes to marsh accretion and therefore the ability to maintain an elevation in a dynamic equilibrium with sea-level rise (Morris et al., 2002). Accretion rates in natural marshes were positively related to both labile and recalcitrant carbon stocks, but in created marshes, there was no relationship between accretion rate and carbon stocks. Likewise, mineral sediment stock was positively related to accretion rate in natural marshes but not in created marshes. These findings indicate that even after 32 years of development, dynamic feedbacks between plant productivity, and sedimentation and marsh accretion were decoupled. Although there may be inherent differences to created marshes compared to natural marshes due to the age difference, it is rare to find natural marshes of the same age as created marshes. This study also focused on the material that has accumulated in the surface to make a better comparison between the natural and created marsh surface. To contribute to accretion and therefore the upward volumetric expansion of the marsh, belowground biomass must expand throughout the soil column. With additional accommodation space afforded by dredge sediment, plant roots grow into the sediment before eventually contributing to accretion of the marsh surface. Sediment deposition on the surface allows for additional accommodation space for root growth and surface accretion. Much of the mineral stock in the created marshes was made of deposited dredged material with generally low sediment availability. With a low deposition rate and an abundance of previously deposited dredge material forming the substrate for root growth, there was no relationship between mineral sediment stock and accretion rates.

5. Conclusions

Carbon sequestration in transgressive deltaic marshes and created marshes in Louisiana over the last ~60 years is largely a result of input of recalcitrant carbon from the local vegetation. Although other studies have shown a variable relationship between mineral sediment and accretion in marshes along the Gulf coast (Turner et al., 2002), here we show that mineral sediment plays a key role in increasing the accumulation of recalcitrant

carbon, which increases the total SOC accumulation and thereby marsh accretion. Overall, RC comprised 72.2 % of the SOC across marsh types and ages indicating that most of the carbon pool is chemically stable. In created marshes, SOC pool is initially composed of labile dredge material-associated carbon and increases slowly and becomes more recalcitrant with inputs from the local vegetation. Marsh loss rates along the Louisiana coast average approximately $30 \text{ km}^2 \text{ y}^{-1}$ per year (Couvillion et al., 2017). This massive loss of recalcitrant soil carbon was derived from marsh vegetation. However, marshes created from dredge sediment have low carbon stocks that do not compare to the carbon stocks of natural marshes. Therefore, the protection and augmentation of natural marshes with their large carbon stocks may be an important strategy to mitigate climate change.

CRedit authorship contribution statement

Megan Kelsall: Investigation, Formal analysis, Writing – original draft, Visualization, Data curation. **Tracy Quirk:** Conceptualization, Methodology, Resources, Writing – review & editing, Visualization, Funding acquisition, Data curation, Supervision. **Carol Wilson:** Writing – review & editing, Resources. **Gregg A. Snedden:** Writing – review & editing, Resources.

Data availability

Data will be made available on request.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This project was supported by NOAA-Louisiana Sea Grant #: NA140AR4170099. This work was possible thanks to collaboration with Sabine National Wildlife Refuge staff. We would like to thank the numerous people and organizations that provided samples and data including Kate Abbott, Sam Shrull, CPRA, and to Tommy Blanchard, Sara Gay, and Sam Bentley with the Wetland Biogeochemistry Analytical Services and Coastal Studies Institute labs at LSU who assisted in laboratory support for this project, and the University of California, Davis Laboratory for analyzing samples.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2023.161415>.

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