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Soil organic carbon stabilization and associated mineral protection in typical coastal wetlands under different hydrologic conditions

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The soil carbon sequestration largely depends on soil organic carbon (SOC) stability. However, the mechanism of SOC stabilization in coastal wetlands under different hydrologic conditions remains to be clarified. In this study, the effect of mineral protection on SOC stabilization was investigated in freshwater-flooded *Phragmites australis* wetlands (FPW), tidal *P. australis* wetlands (TPW), non-flooded *P. australis* wetlands (NPW) and tidal *Suaeda salsa* wetlands (TSW). Two stabilized SOC fractions (Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC) and Al/Fe/Si oxides extracted by acid oxalate, dithionite–citrate and pyrophosphate, respectively, were determined in soil samples collected to a depth of 25 cm. The contents of soil Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC were not significantly different among the four wetlands ($p > 0.05$), with the highest mean values occurring in FPW ($1.44 \pm 0.43 \text{ g kg}^{-1}$) and TPW ($1.79 \pm 0.40 \text{ g kg}^{-1}$), respectively. The contents of Al/Fe/Si oxides (except for Si_p) and the values of mineral phase indicators were significantly lower in NPW than in TPW or FPW ($p < 0.05$). Linear regressions among Al/Fe/Si oxides, mineral phase indicators, and two stabilized SOC fractions showed that Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC were positively and significantly affected by mineral contents ($p < 0.05$). Additionally, edaphic factors such as soil organic matter, total nitrogen, soil water content, clay and silt contents were identified as important factors influencing the two stabilized SOC fractions.

KEYWORDS

SOC stabilization, Al/Fe/Si oxides, mineral phase indicators, hydrologic conditions, coastal wetlands

Introduction

As the largest terrestrial carbon pool, SOC stock in soils is three times larger than the size of atmospheric carbon pool and vegetation carbon stock (Gulde et al., 2008; Warren and Ziegler, 2017). The sequestration of carbon as SOC is a major topic of research worldwide because of its great importance in climate change mitigation and inhibition of soil degradation (Vos et al., 2017; Rügge et al., 2019). The balance between soil carbon inputs (i.e., vegetation and exogenous sources) and outputs (i.e. mineralization, erosion and leaching) determines the SOC stock, and the proportion of carbon input that becomes stabilized in SOC and the maintenance of its stability is vital to ensuring long-term SOC stocks (Rügge et al., 2019). Moreover, land-atmosphere carbon exchanges can be altered by the changes in SOC stability, leading to dynamic variations in soil carbon release (Tian et al., 2016). Furthermore, SOC stability is critical to soil quality and fertility through the regulation of soil characterization and nutrient level (Yang et al., 2020). The concept of “SOC stability” refers to the resilience or resistance to changes (Doetterl et al., 2016), with the ideal situation that SOC remains constant in its molecular composition and concentration for long periods of time (Berhe and Kleber, 2013). To sum up, the biochemical recalcitrance of molecules, the spatial inaccessibility of SOC for decomposer organisms and enzymes, the stabilization by the interactions of SOC with mineral surfaces and metal ions, and physical protection from aggregates control the spatial heterogeneity of SOC stability (Lützow et al., 2006; Lorenz et al., 2009; Sierra et al., 2016; Tian et al., 2016).

Although there are no unified standards for the assessment of SOC stability, a series of complementary techniques enable the evaluation and interpretation of the SOC stabilization mechanisms in different soils. Various techniques (i.e., thermal analysis techniques and chemical separation methods) have been considered suitable metrics in the evaluation of SOC stability (Lorenz et al., 2009; Sierra et al., 2016; Soucémariadin et al., 2018). SOC pools can be classified into labile and stable SOC pools (Gulde et al., 2008) or labile (active), intermediate and stable (persistent) SOC pools (Tian et al., 2016; Soucémariadin et al., 2018; Lyu et al., 2021). The most stable organic carbon components can be stored for hundreds of years or longer as a result of the retardation of the internal physical, chemical and microbial cycles (Gulde et al., 2008). Stable SOC contributes more to the sequestration and storage of SOC than labile SOC (Gulde et al., 2008; Soucémariadin et al., 2018). Correspondingly, the proportion of stable SOC in soil determines the stability of SOC and the capacity of soil to store SOC long term. The preservation of stable SOC enormously affects atmospheric CO₂ levels, soil fertility, plant productivity, and the soil ecosystem (Lyu et al., 2021). Both natural and anthropogenic perturbations can alter SOC stability by affecting the SOC decomposition, which depends on physicochemical properties and carbon availability for

microbial and enzyme activities (Yang et al., 2020). Therefore, it is of the utmost importance to investigate how SOC stabilization responds to the changes of environmental conditions so as to better understand the potential for enhancement of global carbon storage.

Strengthening soil carbon sequestration and CO₂ fixation has become an urgent issue in the context of global climate change. Coastal wetlands feature with great capacity for carbon accumulation, with the potential to substantially affect the global carbon budget (Baustian et al., 2021). Specifically, it has been estimated that coastal wetlands account for only 0.2% of the ocean surface, but 50% of carbon stock is sequestered in marine sediments (Macreadie et al., 2017). Furthermore, coastal wetlands are associated with high productivity and high capacity for the regulation of CO₂ levels, causing them to be major carbon sinks (Lu et al., 2017). Consequently, coastal wetlands have gained an increasing amount of research and the protection and restoration of coastal wetlands are targets for greenhouse gas emissions offsetting and carbon sequestration projects (Kroeger et al., 2017). The soil carbon sequestration rates in coastal wetlands partly depend on their unique hydrologic conditions, as flooding duration, the oxygen-deficient soil environment and high salinity lead to low decomposition rate of organic matter and tidal oscillations could increase organic matter inputs (Chambers et al., 2013; Ferronato et al., 2019; Baustian et al., 2021). Moreover, variations in wetland hydrologic conditions would induce changes in the vegetation type and soil physiochemical characterization that further cause fluctuations in the soil carbon stock (Zhao et al., 2017; Ma et al., 2017). Notably, coastal wetlands may face increased flooding under future global climate change (Ferronato et al., 2016). Thus, the impact of hydrologic variations on SOC stabilization in coastal wetlands is worthy of deeply probing into.

The fluctuations in hydrologic conditions will undoubtedly affect SOC stabilization in many ways, especially by augmenting carbon inputs and outputs. Moreover, it has been reported that waterlogging affects the chemical stability of soil organic matter by altering soil biochemical characteristics (i.e., enzyme activities and microbial activities) and the quality of organic matter (Ferronato et al., 2019). Lyu et al. (2021) showed that the sizes of intermediate and stable SOC pools were predominantly and positively controlled by the contents of nanocrystalline minerals. The initial interaction between minerals and SOM mediated controlled SOC stability by mediating the sorption of chemically diverse organic matter molecules *via* distinct surface areas and chemical functional group availabilities, and this mineralogic control has been reported by several studies conducted in volcanic soils, dry tropical forest soils, temperate forest soils and grassland soils (Lorenz et al., 2009; Soucémariadin et al., 2018; Zhao et al., 2020; Lyu et al., 2021). However, few studies have examined their effects on SOC stabilization in coastal wetlands, to our knowledge. The current study aimed to

investigate how different hydrologic conditions affect the contents of representative stabilized SOC fractions ($\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC), and stabilization mechanisms associated with minerals in typical coastal wetlands of the Yellow River Delta, China. The clarification of how SOC stabilization in coastal wetlands responds to hydrologic variations and SOC stabilization protected by minerals will inform the assessment of SOC stock in coastal wetlands, strategic decision making for wetland restoration and guidelines for climate change mitigation.

Materials and methods

Sampling sites, soil collection and analysis

Four typical wetlands in the Nature Reserve of the Yellow River Delta were selected as sampling sites, including freshwater-flooded *Phragmites australis* wetlands (FPW), tidal *P. australis* wetlands (TPW), non-flooded *P. australis* wetlands (NPW) and tidal *Suaeda salsa* wetlands (TSW). FPW, NPW and TSW are located on the north bank of the Yellow River, and TPW is located on the south bank of the Yellow River (Figure 1). Different hydrologic conditions were observed in these four wetlands. Soil cores were randomly collected to a depth of

25 cm and stratified into five layers at 5 cm interval in each wetland in October 2018. Three soil samples were collected for each soil increment. Samples for the measurement of soil bulk density (BD) were collected by the cutting-ring method with 100 cm^3 cores. Methods for the pretreatment of soil samples and determination of BD, soil water content (WC), pH, electric conductivity (EC), total carbon (TC), total nitrogen (TN), soil organic matter (SOM) and soil base cation and anion (e.g., Na^+ , K^+ , Mg^{2+} , Ca^{2+} , Cl^- and SO_4^{2-}) contents can be found in Zhao et al. (2020).

The contents of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC represent the amount of stable SOC (also called chemically recalcitrant SOC and non-oxidizable SOC) and functionally passive SOC, respectively (Lorenz et al., 2009; Sierra et al., 2016). Thus, they are usually selected as representative stabilized SOC fractions. The determination of these two stabilized SOC fractions, acid oxalate-extractable Al/Fe/Si (Al_o , Fe_o and Si_o), dithionite-citrate-extractable Al/Fe/Si (Al_d , Fe_d and Si_d) and pyrophosphate-extractable Al/Fe/Si (Al_p , Fe_p and Si_p) was conducted with reference to modified chemical fractionation methods by Lorenz et al. (2009). Acid oxalate-extractable Al, Fe, and Si represent most organic complexes and irregularly arranged minerals such as allophane, ferrihydrite, and imogolite. Dithionite-citrate extracts Fe from both crystalline and poorly crystalline Fe oxides, and Al substituted into Fe oxides and produced by the partial dissolution of poorly ordered Al (oxy)

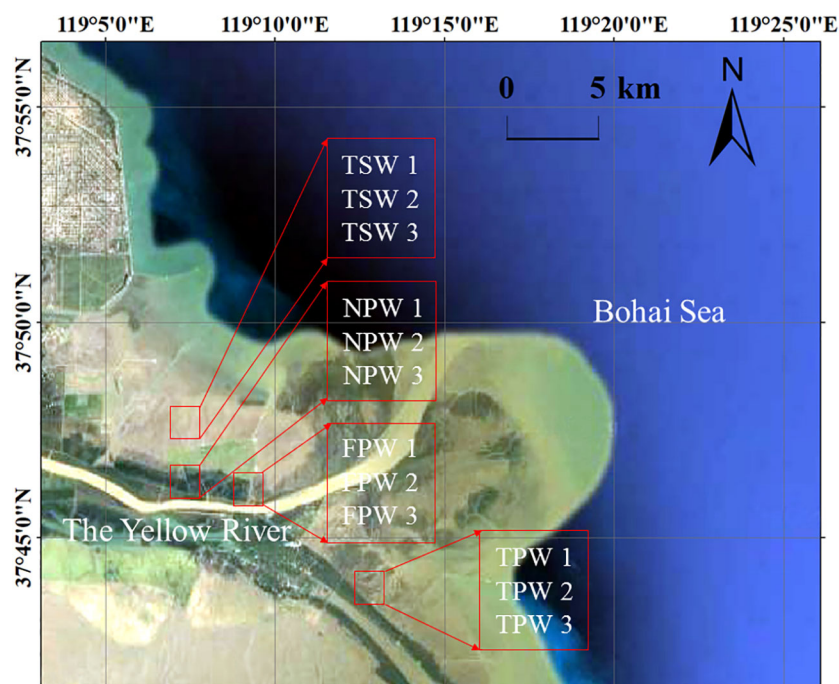


FIGURE 1
Location map of sampling sites.

hydroxides are represented by dithionite-citrate-extractable Al. All of the organically bound Al and an intermediate percentage of the organically bound Fe were extracted using sodium pyrophosphate. The fraction of total Fe oxides with poor crystallinity is represented by the Fe_o/Fe_d ratio. As a result, Fe_o minus Fe_p reflects the amount of ferrihydrite, while Fe_d minus Fe_o represents crystalline Fe oxides. The ratio of organically-bound to total amorphous aluminum, or the proportion of aluminum that is in organic complexes, is represented by the Al_p/Al_o ratio. Sesquioxide content is denoted by $Al_o+0.5Fe_o$, which is also indicative of the amount and status of nanocrystalline minerals (Lyu et al., 2021).

Statistical analysis

One-way analysis of variance (ANOVA) was conducted to identify the differences in soil properties, $Na_2S_2O_8$ resistant SOC, H_2O_2 resistant SOC, acid oxalate extractable Al/Fe/Si, dithionite-citrate extractable Al/Fe/Si, pyrophosphate-extractable Al/Fe/Si and mineral phase indicators among the four wetlands using SPSS 22.0 software package. Differences were considered to be significant if $p < 0.05$. Redundancy analysis (RDA), correlation analysis and single variable linear regression were also performed to analyze the relationships between $Na_2S_2O_8$ resistant SOC, H_2O_2 resistant SOC, Al/Fe/Si oxides, mineral phase indicators and soil properties with Canoco 4.5 software package and SPSS 22.0 software package, respectively. Figures were created using Origin 2019 software package.

Results

Soil physiochemical characteristics in the four wetlands

The SOM, TC, and TN contents were significantly higher in TPW than in NPW ($p < 0.05$), but no significant differences were observed among those in TPW, TSW, and FPW ($p > 0.05$, Table 1). The C/N ratio was not significantly different among the soils of the four wetlands ($p > 0.05$). Soil BD in TPW ($< 1.5 \text{ g cm}^{-3}$) was significantly lower than that of the other three wetlands ($> 1.5 \text{ g cm}^{-3}$, $p < 0.05$). In general, soil EC, Na^+ , K^+ , Mg^{2+} , Ca^{2+} , Cl^- , and SO_4^{2-} contents in TSW soils were significantly higher than in the other three wetlands ($p < 0.05$), with the lowest values of EC, Na^+ , and Cl^- occurring in FPW soils and K^+ , Mg^{2+} , Ca^{2+} , and SO_4^{2-} appearing in NPW soils, respectively. For pH, significantly higher values (> 9) were observed in the soils of NPW than in the soils of the other three wetlands ($p < 0.05$). The soil texture of the four wetlands was mainly composed of sand ($> 70\%$), with much lower proportions of clay and silt. The silt and sand contents in TPW were significantly higher and lower than those of NPW and FPW ($p < 0.05$). The WC was significantly higher in FPW than that in the other three wetlands ($p < 0.05$). Overall, flooding caused by tides or freshwater increased and reduced soil EC and salt ions (except for Ca^{2+}), respectively. Compared with NPW which lacks flooding from tides or freshwater, flooding conditions in TSW, TPW, and FPW increased the SOM, TN, silt, and WC contents, and lowered the sand content and pH.

TABLE 1 Soil characterization of 0–25 cm soils in the four wetlands.

	TSW	NPW	TPW	FPW
SOM (%)	0.52 ± 0.17 ^{ab}	0.38 ± 0.07 ^b	0.69 ± 0.20 ^a	0.57 ± 0.32 ^a
TC (%)	1.28 ± 0.23 ^a	1.04 ± 0.09 ^b	1.43 ± 0.23 ^a	1.37 ± 0.52 ^{ab}
TN (%)	0.033 ± 0.008 ^{ab}	0.026 ± 0.004 ^b	0.042 ± 0.009 ^a	0.036 ± 0.021 ^{ab}
C/N ratio	10.73 ± 1.91 ^a	9.96 ± 1.44 ^a	11.03 ± 1.95 ^a	10.83 ± 2.09 ^a
BD (g cm^{-3})	1.59 ± 0.09 ^{ab}	1.57 ± 0.03 ^a	1.41 ± 0.09 ^c	1.51 ± 0.04 ^b
EC _{1:5} (mS cm^{-1})	15.24 ± 7.21 ^a	1.22 ± 0.56 ^c	2.48 ± 0.45 ^b	0.49 ± 0.13 ^d
pH _{1:5}	8.58 ± 0.13 ^b	9.30 ± 0.47 ^a	8.47 ± 0.22 ^b	8.39 ± 0.42 ^b
Na^+ (mg kg^{-1})	4401.00 ± 1338.70 ^a	1121.17 ± 284.03 ^c	1885.46 ± 338.36 ^b	331.23 ± 121.53 ^d
K^+ (mg kg^{-1})	133.37 ± 56.68 ^a	14.33 ± 4.79 ^d	65.75 ± 13.39 ^b	26.11 ± 6.49 ^c
Mg^{2+} (mg kg^{-1})	400.61 ± 315.15 ^a	17.30 ± 8.13 ^d	62.52 ± 22.51 ^b	30.10 ± 16.02 ^c
Ca^{2+} (mg kg^{-1})	201.62 ± 60.71 ^a	42.28 ± 7.34 ^d	74.47 ± 17.76 ^c	93.71 ± 28.35 ^b
Cl^- (mg kg^{-1})	6691.04 ± 2062.61 ^a	1339.06 ± 407.99 ^c	2327.85 ± 430.06 ^b	386.0 ± 122.02 ^d
SO_4^{2-} (mg kg^{-1})	1045.89 ± 612.59 ^a	108.49 ± 66.49 ^d	513.36 ± 110.43 ^b	160.45 ± 149.58 ^c
Clay (%)	1.19 ± 3.11 ^a	0.00 ± 0.00 ^a	0.16 ± 0.37 ^a	0.08 ± 0.30 ^a
Silt (%)	18.89 ± 13.75 ^{ac}	5.37 ± 4.60 ^b	22.24 ± 3.22 ^a	12.14 ± 8.14 ^{bc}
Sand (%)	79.92 ± 16.25 ^{abc}	94.63 ± 4.60 ^a	77.60 ± 3.45 ^c	85.88 ± 4.33 ^{ab}
WC (%)	22.86 ± 1.96 ^{bc}	22.09 ± 0.70 ^c	22.79 ± 0.58 ^b	26.71 ± 4.33 ^a

^{abcd}Different letters represent significant differences ($p < 0.05$) among sampling sites.

The contents and profile distribution of the two stabilized SOC fractions

The $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC contents in the four wetlands are shown in Figure 2. Both $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC showed no significant differences among the four wetlands ($p > 0.05$), and the mean values of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC at 0–25 cm soil depth were lower than H_2O_2 resistant SOC in all four wetlands. The highest mean values of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC were observed in FPW ($1.44 \pm 0.43 \text{ g kg}^{-1}$) and TPW ($1.79 \pm 0.40 \text{ g kg}^{-1}$), respectively, while the lowest mean values of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC were found in NPW ($1.19 \pm 0.44 \text{ g kg}^{-1}$) and TSW ($1.50 \pm 0.52 \text{ g kg}^{-1}$), respectively.

Figure 3A shows the profile distribution of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC in the 0–25 cm soils of the four wetlands. The $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC in soils of TSW, NPW and TPW showed an “increasing before decreasing” tendency as depth increased, with higher $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC levels occurring in the 10–15 cm soils. And the highest values of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC among five soil layers in TSW, NPW and TPW were $1.70 \pm 0.37 \text{ g kg}^{-1}$, $1.53 \pm 0.72 \text{ g kg}^{-1}$ and $1.90 \pm 0.13 \text{ g kg}^{-1}$, respectively. The $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC in 0–5 cm soils was significantly higher in FPW ($1.80 \pm 0.62 \text{ g kg}^{-1}$) than in NPW ($0.82 \pm 0.31 \text{ g kg}^{-1}$, $p < 0.05$), while $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC in other soil layers showed no significant difference among the four wetlands ($p > 0.05$).

The distribution pattern of soil H_2O_2 resistant SOC was similar to that of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC in TSW, NPW and TPW, showing an initial increasing then decreasing trend (Figure 3B). The H_2O_2 resistant SOC content in 0–5 cm soils was significantly higher in soils of TPW ($1.85 \pm 0.37 \text{ g kg}^{-1}$) and FPW ($2.35 \pm 0.87 \text{ g kg}^{-1}$) than that in soils of NPW ($0.90 \pm 0.16 \text{ g kg}^{-1}$, $p < 0.05$), while a

significantly higher value of H_2O_2 resistant SOC in 10–15 cm soils of NPW ($2.19 \pm 0.30 \text{ g kg}^{-1}$) and TPW ($2.10 \pm 0.54 \text{ g kg}^{-1}$) than that of FPW ($1.42 \pm 0.26 \text{ g kg}^{-1}$) was observed ($p < 0.05$). The H_2O_2 resistant SOC in 5–10 cm, 15–20 cm and 20–25 cm soils showed no significant differences among the four wetlands ($p > 0.05$), with higher values occurring in soils of FPW (5–10 cm, $2.17 \pm 1.08 \text{ g kg}^{-1}$).

Comparison of Al, Fe and Si oxides in soils of the four wetlands

The contents of Al extracted by acid-oxalate, dithionite-citrate and pyrophosphate in the soils of the four wetlands are shown in Figures 4A–C, respectively. The content of soil Al_o in NPW was significantly lower than that in the other three wetlands ($p < 0.05$), with a mean value of $0.22 \pm 0.04 \text{ g kg}^{-1}$. However, the content of soil Al_o showed no significant difference among TSW, TPW and FPW ($p > 0.05$), with the highest mean soil Al_o content observed in TPW ($0.39 \pm 0.10 \text{ g kg}^{-1}$). TSW ($0.38 \pm 0.09 \text{ g kg}^{-1}$) and TPW ($0.40 \pm 0.06 \text{ g kg}^{-1}$) had significantly higher soil Al_d contents than NPW ($0.28 \pm 0.03 \text{ g kg}^{-1}$, $p < 0.05$). However, Al_d in FPW ($0.40 \pm 0.12 \text{ g kg}^{-1}$) did not differ significantly from NPW and TSW ($p > 0.05$), but was significantly lower than TPW ($p < 0.05$). As for Al_p , a significantly lower value in NPW ($0.033 \pm 0.01 \text{ g kg}^{-1}$) was found than in FPW ($0.054 \pm 0.04 \text{ g kg}^{-1}$), but no significant differences were observed among TSW, NPW and TPW, or among TSW, NPW and FPW ($p > 0.05$).

The contents of acid-oxalate, dithionite-citrate and pyrophosphate extractable Fe are shown in Figures 4D–F, respectively. The content of soil Fe_o in NPW ($0.65 \pm 0.17 \text{ g kg}^{-1}$) was significantly lower than that in TPW ($1.36 \pm 0.30 \text{ g kg}^{-1}$) and

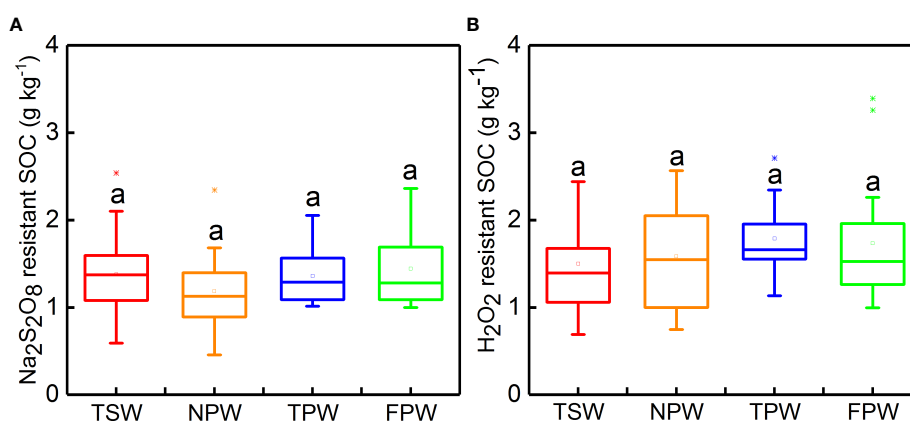


FIGURE 2

Box chart of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC (A) and H_2O_2 resistant SOC (B) in 0–25 cm soils in the four wetlands. (Different lowercase letters represent significant differences at $p < 0.05$ level).

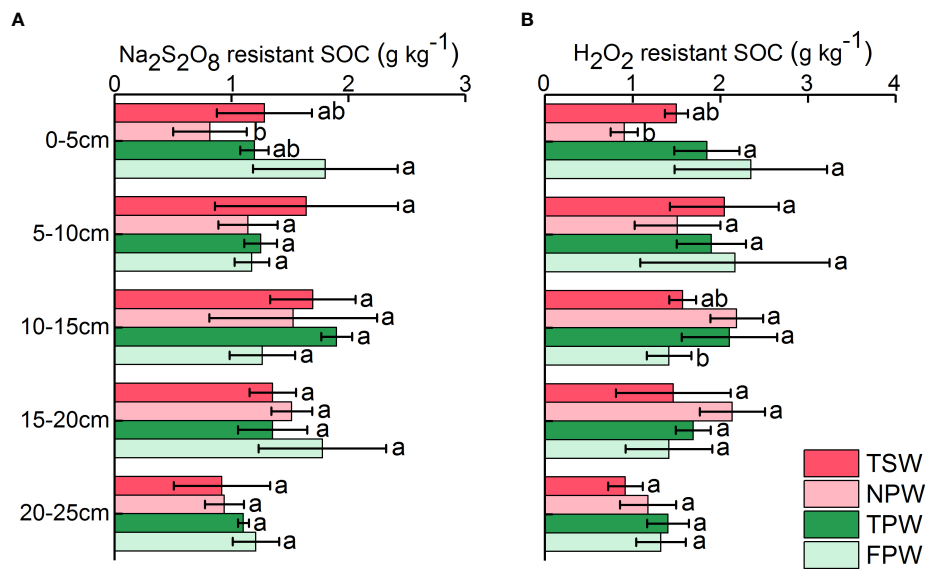


FIGURE 3
Profile distribution of soil Na₂S₂O₈ resistant SOC (A) and H₂O₂ resistant SOC (B) in the four wetlands. (Different lowercase letters represent significant differences between the same layers of the four wetlands at *p* < 0.05 level).

FPW ($1.20 \pm 0.59 \text{ g kg}^{-1}$, $p < 0.05$), but showed no significant difference with TSW ($0.80 \pm 0.28 \text{ g kg}^{-1}$, $p > 0.05$). TSW had a significantly lower soil Fe_o content than TPW ($p < 0.05$), but did not differ from FPW ($p > 0.05$). Although the content of soil Fe_d in NPW ($3.70 \pm 0.36 \text{ g kg}^{-1}$) was significantly lower than that in TSW

($4.53 \pm 0.82 \text{ g kg}^{-1}$) and TPW ($5.07 \pm 0.57 \text{ g kg}^{-1}$, $p < 0.05$), no significant difference was observed between NPW and FPW ($p > 0.05$). Moreover, no significant difference was observed between TSW and TPW or between TSW and FPW ($p > 0.05$). Both TSW and NPW had significantly lower soil Fe_p contents than TPW

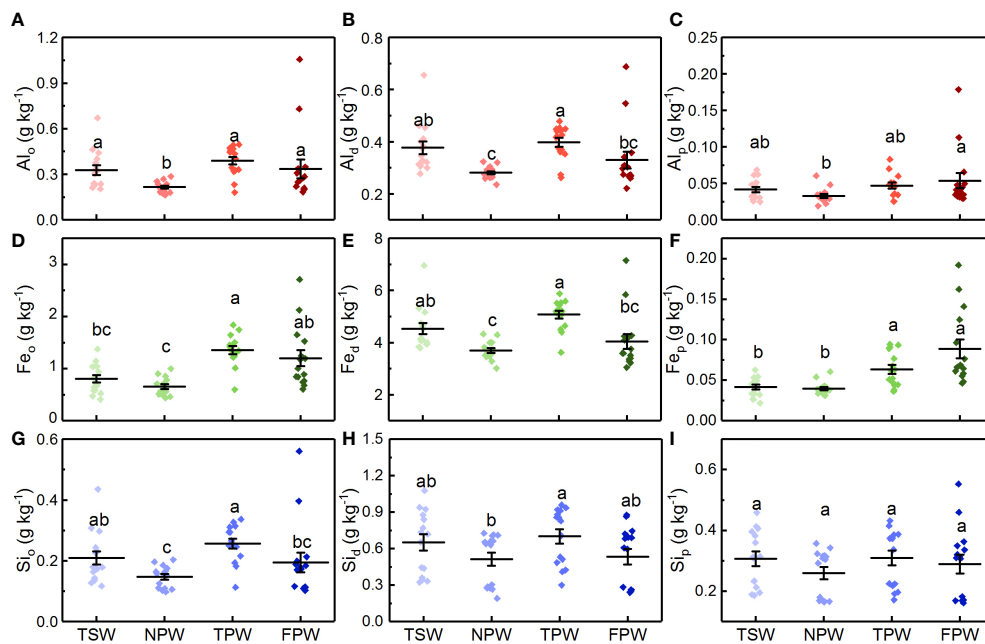


FIGURE 4
Comparison of Al_o (A), Al_L (B), Al_P (C), Fe_o (D), Fe_d (E), Fe_p (F), Si_o (G), Si_d (H) and Si_p (I) contents in the four wetlands. (Different lowercase letters represent significant differences between the same layers of the four wetlands at *p* < 0.05 level).

($0.063 \pm 0.02 \text{ g kg}^{-1}$) and FPW ($0.088 \pm 0.05 \text{ g kg}^{-1}$, $p < 0.05$), but no significant differences were found between TSW ($0.042 \pm 0.01 \text{ g kg}^{-1}$) and NPW ($0.039 \pm 0.01 \text{ g kg}^{-1}$), or between TPW and FPW ($p > 0.05$).

As shown in Figures 4G-I, the contents of soil Si_o , Si_d and Si_p in the four wetlands decreased in the order $\text{TPW} > \text{TSW} > \text{FPW} > \text{NPW}$. The content of Si_o in NPW ($0.15 \pm 0.04 \text{ g kg}^{-1}$) was significantly lower than that in TSW ($0.21 \pm 0.09 \text{ g kg}^{-1}$) and TPW ($0.26 \pm 0.06 \text{ g kg}^{-1}$, $p < 0.05$), but did not differ from that of FPW ($0.21 \pm 0.13 \text{ g kg}^{-1}$, $p > 0.05$). TPW had a significantly higher content of Si_d ($0.70 \pm 0.23 \text{ g kg}^{-1}$) than NPW ($0.51 \pm 0.21 \text{ g kg}^{-1}$, $p < 0.05$). However, no significant differences were found among TSW, NPW and FPW, or among TSW, TPW and FPW ($p > 0.05$). Unlike the Si_o and Si_d , the contents of soil Si_p in the four wetlands were not statistically different ($p > 0.05$).

Mineral phase indicators of Al, Fe and Si in the four wetlands

Table 2 shows the comparison of mineral phase indicators (Fe_o/Fe_d ratio, $\text{Fe}_d\text{-Fe}_o$, $\text{Fe}_o\text{-Fe}_p$, Al_p/Al_o ratio and $\text{Al}_o+0.5\text{Fe}_o$) among the four wetlands. There were significantly higher poorly crystalline Fe oxide contents (based on Fe_o/Fe_d ratio) in TPW and FPW than in TSW and NPW ($p < 0.05$), with mean values > 0.2 . As shown in Table 2, TSW and TPW had significantly higher crystalline Fe oxides contents ($\text{Fe}_d\text{-Fe}_o$, $3.73 \pm 0.62 \text{ g kg}^{-1}$ and $3.72 \pm 0.36 \text{ g kg}^{-1}$, respectively) than NPW and FPW ($p < 0.05$), but no significant difference was found between TSW and TPW or between NPW and FPW ($p > 0.05$). FPW has the lowest $\text{Fe}_d\text{-Fe}_o$ content of the four wetlands ($< 3 \text{ g kg}^{-1}$). The ferrihydrite content represented by $\text{Fe}_o\text{-Fe}_p$ was significantly higher in TPW and FPW than those in TSW and NPW ($p < 0.05$), which were higher than 1 g kg^{-1} in TPW and FPW. As the indicator for the relative amount of Al in organic complexes, the Al_p/Al_o ratio in TPW (0.12) was significantly lower than that in NPW and FPW ($p < 0.05$), but showed no significant difference with TSW ($p > 0.05$). The Al_p/Al_o ratio in FPW was significantly higher than that in TSW ($p < 0.05$), but did not significantly differ from NPW ($p > 0.05$). The content of sesquioxides represented as $\text{Al}_o+0.5\text{Fe}_o$ in TPW ($> 1 \text{ g kg}^{-1}$) was significantly higher than that in TSW and NPW ($p < 0.05$), but showed no significant difference with that in FPW ($p > 0.05$).

Relationships between the two stabilized SOC fractions, Al/Fe/Si oxides, mineral phase indicators and soil properties

The RDA between the two stabilized SOC fractions, Al/Fe/Si oxides, mineral phase indicators, and soil properties is shown in Figure 5. The cumulative percentage of variance of two stabilized SOC fractions was 79.1% of Axis 1 (62.1%) and Axis 2 (17.0%). The two stabilized SOC fractions, SOM, TC, TN, C/N ratio, WC, clay, silt, Al/Fe/Si oxides (except for Si_d and Si_p), and mineral phase indicators were mainly distributed on the right part of Axis 1, and the two stabilized SOC fractions were highly and positively affected by SOM, TC, TN, WC, clay, silt, Fe_o , Fe_p and Al_p . The sand content, BD, and EC were located on the left part of Axis 1, and these soil edaphic factors negatively influenced the two stabilized SOC fractions. The correlation analysis also showed that $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC was significantly and positively correlated with SOM, TC, TN, clay, WC, Al/Fe/Si oxides (except for Fe_p , Si_d and Si_p), $\text{Fe}_d\text{-Fe}_o$, $\text{Fe}_o\text{-Fe}_p$ and $\text{Al}_o+0.5\text{Fe}_o$ (Figure 6, $p < 0.05$), and significantly and negatively correlated with sand ($p < 0.05$, Figure 6). A significantly positive relationship was observed between H_2O_2 resistant SOC and SOM, TC, TN, WC, Fe/Al/Si oxides (except for Si_d and Si_p), as well as $\text{Fe}_d\text{-Fe}_o$, $\text{Fe}_o\text{-Fe}_p$ and $\text{Al}_o+0.5\text{Fe}_o$ ($p < 0.05$, Figure 6).

As shown in Table 3, Fe_o , Si_o , Al_d , Fe_d , Al_p , Fe_p , Si_p , Fe_o/Fe_d ratio, $\text{Fe}_o\text{-Fe}_p$ and $\text{Al}_o+0.5\text{Fe}_o$ all significantly contributed to the variability of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC ($p < 0.01$) in 0-5 cm soils. Moreover, Si_d and $\text{Fe}_d\text{-Fe}_o$ explained 41% and 46% of the variation of H_2O_2 resistant SOC ($p < 0.05$). In particular, the amounts of Fe_o , Si_o , Al_d , Al_p , Fe_p , Si_p , $\text{Fe}_o\text{-Fe}_p$ and the indicator for amorphous mineral ($\text{Al}_o+0.5\text{Fe}_o$) were related to more than 60% of the variability of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC ($p < 0.01$), with Fe_o , Si_o , Al_d , $\text{Fe}_o\text{-Fe}_p$ and $\text{Al}_o+0.5\text{Fe}_o$ even accounting for more than 80% of the variability of H_2O_2 resistant SOC ($p < 0.001$). However, both the variability of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC were independent of the content of Al_o and the Al_p/Al_o ratio ($p > 0.05$). Additionally, the amount of Si_d and $\text{Fe}_d\text{-Fe}_o$ were not related to $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC ($p > 0.05$). In 10-15 cm soils, Al_o , Si_o , and dithionite extractable Al/Fe/Si oxides each attributed more than 30% of the variability of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC ($p < 0.05$), whereas the content of H_2O_2

TABLE 2 Comparison of mineral phase indicators in the four wetlands.

	TSW	NPW	TPW	FPW
Fe_o/Fe_d ratio	0.17 ± 0.05^b	0.18 ± 0.03^b	0.26 ± 0.04^a	0.29 ± 0.09^a
$\text{Fe}_d\text{-Fe}_o$ (g kg^{-1})	3.73 ± 0.62^a	3.04 ± 0.24^b	3.72 ± 0.36^a	2.84 ± 0.67^b
$\text{Fe}_o\text{-Fe}_p$ (g kg^{-1})	0.76 ± 0.27^b	0.62 ± 0.19^b	1.29 ± 0.28^a	1.11 ± 0.55^a
Al_p/Al_o ratio	0.13 ± 0.03^{bc}	0.15 ± 0.04^{ac}	0.12 ± 0.02^b	0.16 ± 0.04^a
$\text{Al}_o+0.5\text{Fe}_o$ (g kg^{-1})	0.73 ± 0.26^b	0.54 ± 0.12^b	1.07 ± 0.23^a	0.94 ± 0.52^{ab}

^{abc}Different letters represent significant differences ($p < 0.05$) between sampling site.

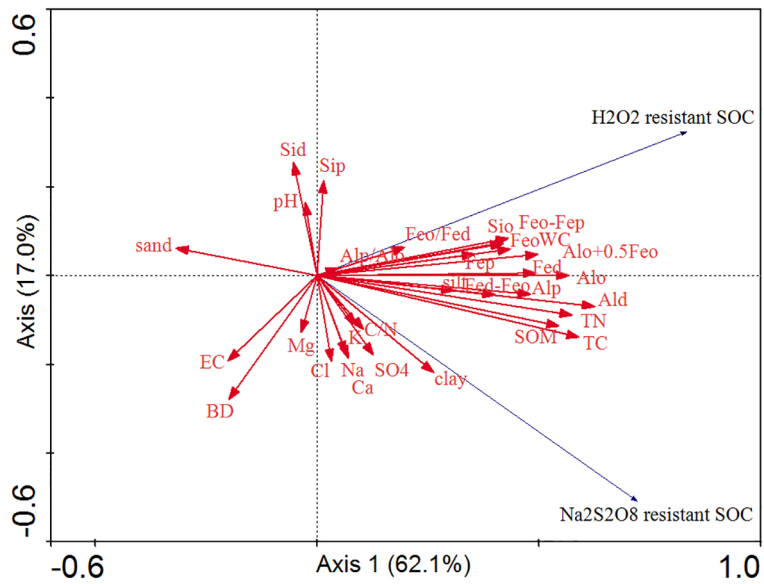


FIGURE 5 Redundancy analysis between the two stabilized SOC fractions, Fe/Al/Si oxides, mineral phase indicators, and soil properties.

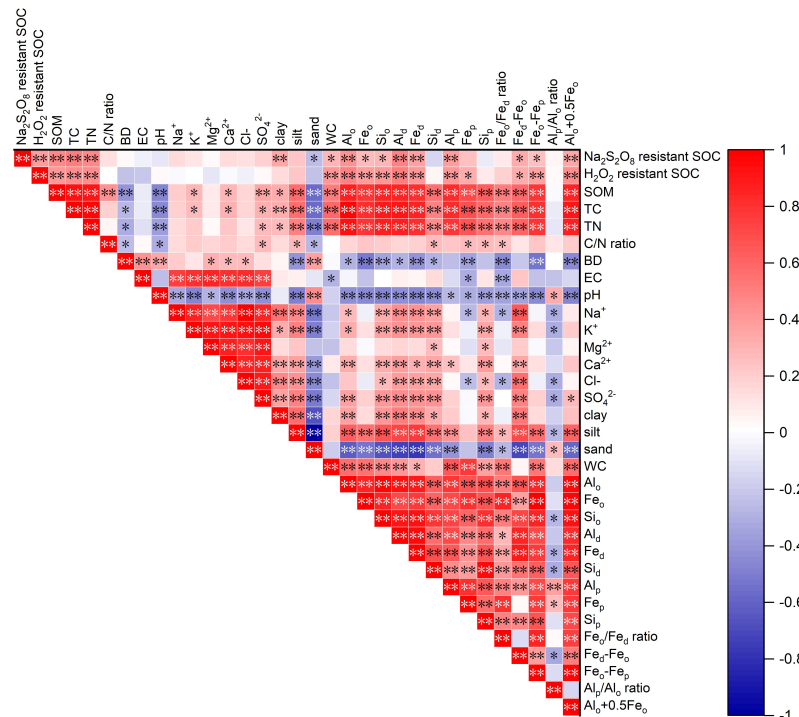


FIGURE 6 Correlation matrix of the two stabilized SOC fractions with Al/Fe/Si oxides, mineral phase indicators and soil properties. [*Correlation is significant at $p < 0.05$ level (2-tailed); **Correlation is significant at $p < 0.01$ level (2-tailed)].

resistant SOC was independent of Al/Fe/Si oxides and mineral phase indicators.

Discussion

Mineral contents and mineral phase indicators in the four wetlands

The contents of Al/Fe/Si oxides extracted by acid oxalate, dithionite–citrate and pyrophosphate were much lower than previously measured in both dry tropical forest and tropical rain forest, which is mainly caused by the difference in soil parent material and weathering process (Lorenz et al., 2009). The significantly lower contents of Al_o, Fe_o, Si_o, Al_d, Fe_d and Si_d in NPW than in TPW indicate that non-crystalline clay minerals and total “free” oxides were more enriched in TPW than in NPW. The significantly higher contents of Al_p and Fe_p in FPW than in NPW indicate that organically-bound Al and the intermediate fraction of organically-bound Fe oxides might play a more important role in SOC stabilization in FPW than in NPW.

The contents of poorly crystalline Fe oxides as indicated by the Fe_o/Fe_d ratio in 0–25 cm soils were significantly lower in NPW than in TPW and FPW, indicating that these two wetlands affected by tidal/freshwater flooding had more active Fe oxides. Higher Fe_d-Fe_o contents generally represent frequent wet and dry cycles and high weathering intensity (Lorenz et al., 2009), which was confirmed by the current study. In our study, TSW and TPW were affected by tidal oscillations and contained

significantly higher poorly crystalline Fe oxides represented as Fe_d-Fe_o than NPW and FPW. Additionally, the complexation of Al and organic acids existed in soils of the four wetlands as the content of soil Si_o was lower than 0.6 g kg⁻¹ and the Al_p/Al_o ratio was no less than 0.5 (Lorenz et al., 2009).

SOC stabilization in the four wetlands

The SOC stabilization in the four wetlands affected by different hydrologic conditions was assessed by determining the resistance of SOC against Na₂S₂O₈ and H₂O₂, which represent stable SOC and functionally passive SOC, respectively. A significant difference in the content of Na₂S₂O₈ resistant SOC was only observed between NPW and FPW or TPW in 0–5 cm soils, and the mean contents of Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC in NPW was lower than that in other three wetlands. Therefore, the three wetlands with the intermittent or long term flooding play a more important role in long-term carbon sequestration than NPW.

Long-term accumulation of soil carbon depends on the “net balance” between inputs and outputs in wetland soils (Kirwan et al., 2013). In general, both the aboveground and belowground biomass of *P. australis* wetlands are higher than those of *S. salsa* wetlands (Zhang et al., 2020), indicating that the carbon inputs tend to be higher in *P. australis* wetlands. However, SOM, Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC in the soils of TSW were not significantly different from the other three *P. australis* wetlands in 0–25 cm soils (Table 1 and Figure 2). Moreover, the contents of Na₂S₂O₈ resistant SOC and H₂O₂

TABLE 3 Coefficients of determination (R² values) for single linear regressions of Na₂S₂O₈ resistant SOC and H₂O₂ resistant SOC as a function of variables (g kg⁻¹) in 0–5 and 10–15cm soils in four wetlands (Regressions for 5–10, 15–20 and 20–25 cm soils were not shown as the two stabilized SOC fractions were not statistically different among four wetlands).

	0–5cm		10–15cm	
	Na ₂ S ₂ O ₈ resistant SOC	H ₂ O ₂ resistant SOC	Na ₂ S ₂ O ₈ resistant SOC	H ₂ O ₂ resistant SOC
Al _o	0.04	0.19	0.35*	0.05
Fe _o	0.65**	0.88***	0.22	0.05
Si _o	0.64**	0.88***	0.35*	0.06
Al _d	0.66**	0.83***	0.37*	0.03
Fe _d	0.56*	0.80***	0.34*	0.03
Si _d	0.29	0.41*	0.34*	0.03
Al _p	0.65**	0.53**	0.15	0.01
Fe _p	0.60**	0.65**	0.01	0.06
Si _p	0.69***	0.54**	0.20	0.01
Fe _o /Fe _d ratio	0.40*	0.53**	0.06	0.01
Fe _d -Fe _o	0.30	0.46*	0.32	0.01
Fe _o -Fe _p	0.64**	0.88***	0.22	0.06
Al _p /Al _o ratio	0.18	0.11	0.27	0.03
Al _o +0.5Fe _o	0.68***	0.89***	0.28	0.06

(N=12; *, **, *** represent significance at $p < 0.05$, $p < 0.01$, $p < 0.001$ level, respectively).

resistant SOC in soils of NPW were significantly lower than those in soils of TPW (H_2O_2 resistant SOC) and FPW in 0–5 cm soil layer ($\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC). This shows that decomposition rates and other factors besides litter input caused by different vegetation type importantly controlled the carbon-storage potential more in these four wetlands. For example, the priming effect could lead to a decrease in SOC stock, which accelerates decomposition rates through the stimulation of fresh organic matter inputs (Rüegg et al., 2019). The effects of oxygen availability on SOC stock should also be taken into account as oxygen availability affects microbial activity and ultimately impacts SOC decomposition. Li et al. (2021) found that oxygen availability regulated key determinants of SOC mineralization rate in paddy soils. Undoubtedly, the flooded soil status and the anaerobic soil environment caused by tidal oscillations (in TSW and TPW) and freshwater flooding (in FPW), flooding duration and soil salinity all contribute to the slow decomposition of SOM, and finally result in the accumulation of SOM over time (Chanlabut et al., 2020). This was also shown in our previous study, which confirmed that tidal oscillations benefit soil carbon sequestration (Zhao et al., 2020).

Mineral protection on SOC stabilization and its controlling factors

The protection of nanocrystalline minerals (active Al and Fe) on SOC stabilization has been reported in various soil types such as volcanic soils, dry tropical forest soils and grassland soils (Lorenz et al., 2009; Zhao et al., 2020; Lyu et al., 2021). Soil Al/Fe cations can combine with SOC through electrostatic adsorption, ligand exchange, surface complexation, hydrophobic interaction, hydrogen bonding, and cation bridging, thus forming stable organic carbon components (Catoni et al., 2016). Furthermore, high specific surface area and low crystallinity of Al/Fe oxides affect the adsorption process of organic matter, and the solubility change of redox active metals drives the coprecipitation process of organic matter and metals (Eusterhues et al., 2005; Wiesmeier et al., 2019). Organic particles in soil can be electrostatically adsorbed with Al_d and Fe_d , which reduces the turnover rate of organic matter and promotes the preservation of organic compounds in sediments (Kögel-Knabner et al., 2008; Xue et al., 2020). The significantly positive relationship between $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC, H_2O_2 resistant SOC and Al/Fe/Si oxides (except for Si_d and Si_p), Fe_d - Fe_o , Fe_o - Fe_p and $\text{Al}_o+0.5\text{Fe}_o$ (Figure 5 and Figure 6) indicates that minerals played an important role in the stabilization of SOC. The significantly lower values of Al_o , Fe_o , Si_o , Al_d , Fe_d , Fe_d - Fe_o , Fe_o - Fe_p and $\text{Al}_o+0.5\text{Fe}_o$ in NPW than in TPW or FPW may explain why lower stabilized SOC fractions were observed in NPW. Amorphous and low-crystalline minerals have a stronger ability to stabilize organic carbon than crystalline minerals by providing a broad surface area and complexing ability, organic biomolecules form metastable and intermediate complexes (Wiseman and

Püttmann, 2006; Basile-Doelsch et al., 2007; Paul et al., 2008; Berhe et al., 2012). Thus, the amount of amorphous minerals and crystalline Fe oxides as indicated by the amount of $\text{Al}_o+0.5\text{Fe}_o$ and the Fe_d/Fe_o ratio, respectively, are more important to the protection of two stabilized SOC fractions in 0–5 cm soils (Table 3).

The significantly lower mean contents of Al/Fe/Si oxides (except for Si_p) in NPW than in TPW or FPW (Figure 4), and the significantly lower values of mineral phase indicators in NPW than those in TPW indicate that the hydrologic conditions might affect the formation and amount of Al/Fe/Si oxides. Saleh et al. (2013) reported that waterlogging significantly increased the concentrations of acetic acid-extractable Si and Fe in a laboratory experiment. Compared with dry land, flooded paddy soils contained higher contents of Al/Fe oxides, and higher soil moisture in flooded soils can accelerate the association of Fe and SOC (Zhou et al., 2009; Balint et al., 2013). In addition, pH determines the binding site of Al/Fe/Si oxides with SOM, consequently controlling the transformations in the type of soil Al/Fe/Si oxides (Kögel-Knabner et al., 2008; Merino et al., 2017; Gartzia-Bengoetxea et al., 2020). Lyu et al. (2021) found that high soil moisture accompanying low soil pH promotes the formation of nanocrystalline minerals. In the current study, there was a significantly positive correlation between soil WC and Al/Fe/Si oxides and mineral phases (except for Si_d , Fe_d - Fe_o and Al_p/Al_o ratio), while pH negatively affected the contents of Al/Fe/Si oxides (Figure 6). Higher soil WC and lower pH in TSW, TPW and FPW than NPW may have promoted the precipitation of Al/Fe/Si oxides and mineral phases to some extent, partly explain the lower Al/Fe/Si oxides contents in NPW than in the other three wetlands.

The interaction between Al/Fe oxides characterized by high activity and clay particles featured with low surface area adsorption can form crystalline or poor crystalline minerals and physically stabilize SOC (Wagai and Mayer et al., 2007; Sarkar et al., 2018). The correlation analysis showed that, except for Al_p/Al_o ratio, soil Al/Fe/Si oxides and mineral phase indicators had positive correlations with soil clay and silt contents, and had a very significant negative correlation with soil sand (except for Fe_p , $p < 0.05$). Additionally, through functioning as binders, Al/Fe oxides can form stable aggregates with clay and silt particles so as to protect SOC (Six et al., 2004). Conversely, a high sand content inhibits the enrichment of Al/Fe oxides, and reduces their protection effects on SOC (Cai et al., 2016). Thus, the lower silt content and higher sand content in NPW are responsible for lower Al/Fe oxides than the other three wetlands to some degree.

Furthermore, the quality of the carbon (specifically, the C/N ratio) and the availability of nutrients influence the reduction and dissolution of Al/Fe/Si oxides in soils (Pallud et al., 2020). Petruzzelli et al. (2005) found that the relatively low C/N ratio (e.g., 9.7) and organic matter content promotes the reduction degree of ferrihydrite and goethite. Thus, it can be concluded that the lower C/N ratio and SOM content might be another

reason for the lower contents of Fe_o , Fe_d and Fe_p in NPW than in the other three wetlands.

Martí-Roura et al. (2019) found that some base ions in soil can aggregate with Al and Fe cations and form minerals, a process that is conducive to the enrichment of Al ions and their mineral phases. Our correlation analysis showed that the soil base cations were significantly positively correlated with Al/Fe/Si oxides and mineral phases (except for Fe_o , Al_p , Fe_o/Fe_d ratio, $\text{Fe}_o\text{-Fe}_p$, Al_p/Al_o ratio and $\text{Al}_o + 0.5\text{Fe}_o$). This corresponds to the higher Al/Fe/Si oxides contents in TSW, TPW and FPW (Table 1). Moreover, the correlation analysis also showed that Na^+ and Cl^- were significantly negatively correlated with Fe_p , Fe_o/Fe_d ratio and Al_p/Al_o ratio, which may further reduce Al/Fe/Si oxides contents and mineral phases in the NPW. Overall, the deficiency of flooding in NPW might be the reason for lower acid oxalate, dithionite-citrate and pyrophosphate extractable Al/Fe/Si oxides and lower contents of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC.

In brief, soil edaphic factors can indirectly alter SOC stabilization through affecting the formation of Al/Fe/Si oxides and minerals phase. Thereby, it can be concluded that hydrologic conditions posed both direct and indirect effects on the mineral protection aspect of SOC stabilization.

Direct influence of some edaphic factors on SOC stabilization

Numerous studies have shown that environmental variables (i.e., soil depth, moisture, vegetation type, and climate) affected SOC stability (Sierra et al., 2016; Soucémariadin et al., 2018; Lyu et al., 2021). The quantity and quality of SOM affect soil carbon content by influencing the functional groups and quantity of soil organisms (Chen et al., 2020), while nitrogen input can affect the wetland carbon transformation by improving the productivity and organic matter content of the wetland ecosystem (Bragazza et al., 2006). Neff et al. (2002) reported that long-term nitrogen input through nitrogen fertilization can stabilize the heavier and mineral-associated SOC fractions. This means that high nitrogen content can increase the input of C, and store carbon for a longer period. In this study, $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC were significantly and positively correlated with SOM, TC and TN (Figure 6). The contents of TC and TN in NPW were lower than in TSW, TPW and FPW, which may partly explain the generally lower $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC contents in NPW than in the other three wetlands.

Among all the edaphic factors, soil texture is particularly important in carbon sequestration. Clay and silt particles combine with soil carbon to form stable clay-organic carbon complexes in soils, which play an important role in preventing microbial oxidation (Six et al., 2002). Cai et al. (2016) reported that fine soil particles (i.e., silt and clay) are capable of stabilizing mineral-

related organic carbon and sequestering persistent SOC. Moreover, Wiesmeier et al. (2014) showed that clay play an important role in maintaining the stability of soil organic carbon. These results support the conclusion that finer soil particles stabilize more SOC than coarser sand particles. The significant relationships between $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and clay (positive), silt (positive), and sand (negative) (Figure 6, $p < 0.05$) may explain the higher $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC accumulation in TSW, TPW, and FPW compared with NPW (Figure 2).

Soil WC could affect soil carbon stability by altering the mineralization rate of SOC (Yin et al., 2019). Ferronato et al. (2019) showed that the decreased oxygen content under long-term flooding and saturated soils reduced the decomposition of SOM. Moreover, soil WC controls the transport of nutrients, the diffusion of enzymes, the movement of microorganisms and the efficiency of microbial utilization of nutrients, further modulating soil carbon stock (Qu et al., 2021). In the current study, $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC were significantly correlated with soil WC in a positive way (Figure 6, $p < 0.05$). The significantly higher soil WC in FPW than in TSW, TPW and NPW may be another reason for higher $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC in FPW.

Conclusions

This study explored the changes in the content of chemically separated stable SOC ($\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC) and functionally passive SOC (H_2O_2 resistant SOC) and associated mineral protection provide by Al/Fe/Si oxides extracted by acid oxalate, dithionite-citrate and pyrophosphate, respectively, in four typical coastal wetlands under different hydrologic conditions. Compared with NPW, flooding caused by tides in TPW or freshwater in FPW significantly enhanced the contents of both $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC in 0-5 cm soil layer. Multiple statistical analyses showed that Fe_o , Si_o , Al_d , Fe_d , Si_d , Al_p , Fe_p , Si_p , Fe_o/Fe_d ratio, $\text{Fe}_d\text{-Fe}_o$, $\text{Fe}_o\text{-Fe}_p$ and $\text{Al}_o+0.5\text{Fe}_o$ provided mineral protection on the two stabilized SOC fractions, and some edaphic factors also attributed to the variability of $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC. Thereby, the effects of hydrologic conditions on the amount of Al/Fe/Si oxides, the values of mineral phase indicators and edaphic factors (i.e., SOM, TN, pH, WC and soil texture) caused the variations in $\text{Na}_2\text{S}_2\text{O}_8$ resistant SOC and H_2O_2 resistant SOC among the four wetlands. Overall, more attention should be paid to the effects of hydrologic conditions on SOC stabilization and their possible impacts on carbon sequestration in coastal wetlands. Furthermore, from the perspective of carbon sequestration capacity, the strategic decision on wetland type selection when conducting a degraded wetland restoration program may refer to the results of this study.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

Author contributions

HZ: investigation, methodology, visualization, writing-original draft. JJ: formal analysis, writing - original draft. QZ: conceptualization, investigation, writing-review and editing, funding acquisition. JW: funding acquisition, supervision. YG: writing-review and editing, funding acquisition, supervision. YH: investigation, methodology. GC: supervision. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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