

Article

Effects of Soil Minerals on Carbon Stability in the Mangrove Soil of the Oura River, Japan

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Abstract

Mangrove forest soil contains peat soil and mineral soil. However, the understanding of carbon stabilization in mineral soil in the mangrove forest soil is not well understood. In this study, we aimed to clarify the mechanism of carbon accumulation in mineral soil in the mangrove forest soil. The study site was located in the Oura Bay mangrove forest area on the Okinawa Island, Japan. These mangroves are spread over a small area (-10 ha) and are mainly dominated by *Bruguiera gymnorrhiza*. Given that the amount of soil in the mangrove sediments was larger than the amount of root, it was confirmed that the mangrove soil was mineral soil. In the mangrove soil, insoluble carbon, which corresponded to humin, was the most abundant than other carbons. It was found that soil minerals and metal ions affect the accumulation of NaOH-extracted carbon and Na₄P₂O₇-extracted carbon and that the soil minerals contributed significantly to the accumulation of Na₄P₂O₇-extracted carbon and insoluble carbon in the soil. In addition, it was also found that Ca contributes to the accumulation of the NaOH-extracted carbon, and Al, Ca, Fe, and Mn contributed to the accumulation of the Na₄P₂O₇-extracted carbon. These results suggest that the soil minerals or metal ions contributed to the accumulation of the three types of carbon in the Oura mangrove forest soil.

Keywords: the stability of carbon in mangrove soil, humic substances, polyvalent cations, Oura mangrove

Introduction

Mangroves play important ecological roles and are one of the most productive ecosystems that accumulate, supply, and recycle nutrients (Ewel et al., 1998; Laegdsgaard, 2001; Feller et al., 2002; Maiti and Chowdhury, 2013) as well as protect the coastlines (Marois and Mitsch, 2015). Moreover, large amounts of carbon accumulate in mangrove sediments. In fact, the amount of soil carbon in mangrove sediments has been increasing every year. According to Breithaupt et al. (2012), the estimated annual burial rate in mangroves is 163 g organic carbon m⁻²·y⁻¹, and the total carbon production is 218 Tg C·a⁻¹. This burial rate indicates that approximately 10% of the total carbon produced is stable in soil (Breithaupt et al., 2012). There are peat soil and mineral soil in the mangrove forest soil, and it has been reported that the

mechanism of carbon stabilization differs between these soils (Kida and Fujitake, 2020). However, as shown in the study of Kida and Fujitake (2020), the mechanism of carbon stabilization of the mineral soil in the mangrove forest soil is not well understood. Therefore, it is necessary to clarify the mechanism of carbon accumulation of the mineral soil in the mangrove forest soil.

One of the carbon stabilization mechanisms in soil defined by Lutzow et al. (2006) was due to the inorganic components and metal ions of the soil. In addition, organo-mineral complexes in which organic matter and inorganic matter (soil minerals and metal ions) are bonded, are formed in the soil, and the complex in the clay-, silt-, and sand-sized complexes are seen as the basic units in soil (Cristensen, 2001). Charged humic substances and clay minerals are essential for soil carbon stability (Filip and Alberts, 1994; Percival

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et al., 2000), in addition to the polyvalent cations required for their binding (Kawaguchi and Kyuma, 1959; Ahmed et al., 2002). In terrestrial ecosystems, clay-humus complexes are formed by the binding of fine soil particles and humic substances (Varadachari et al., 1994; Ahmed et al., 2002), which results in the stabilization of humic substances. There are some researches about the stabilization of carbon from humic substances in the farmland and forest soil of the subtropical Okinawa Island in Japan, wherein it has been shown that calcium (Ca) and aluminum (Al) contributed to the stability of carbon from the humic substances in the sugarcane farmland (Kinjo et al., 2009), and Ca, Al, and iron (Fe) contributed to the stability of those in the beech forest soils (Teruya and Kinjo, 2014).

In a study on the factors of carbon accumulation in soil in the climatic environment of Okinawa, the soil minerals and metal ions that contribute to the stability of carbon from humic substances have been clarified from terrestrial soil (Kinjo et al., 2009; Teruya and Kinjo, 2014). However, these factors have not been clarified from the mangrove forest soil, especially from the mineral soils. Estimated from literature (Ishiga and Diallo, 2016), the mangrove forest soil on Okinawa Island may be mineral soil. Therefore, the purpose of this study was to elucidate the carbon accumulation mechanism in the mangrove forest soil,

specifically, a mineral soil from the Okinawa Island. This study was conducted to elucidate the mechanism and the effects of soil minerals and metals in humic substances.

Materials and Methods

Study site

This research was conducted in the Oura Bay mangrove forest located on the Okinawa Island, Japan (Fig. 1). According to Nakasuga et al. (1974), most of the mangrove forests scattered on Okinawa Island have a small scale of 1 ha or less, and the growing mangrove forests also have a breast height diameter of 10 cm or less and a tree height of 5 m or less. Among them, the mangrove forest area in the Oura River basin has a larger area, wherein mangroves grow more than in the other places in Okinawa. The vegetation is dominated by *Bruguiera gymnorrhiza*, *Kandelia obovata*, and *Rhizophora stylosa* (Mfilinge and Tsuchiya, 2008) and covers approximately 10 ha of the total mangrove area. *Bruguiera gymnorrhiza* is distributed throughout the mangrove forest, whereas the other mangrove species are sparse and are mainly distributed near the edges. The height of *B. gymnorrhiza* trees is approximately 4 to 7 m (Mfilinge et al. 2005). In this study, the height of *K. obovata* trees was found to be below 1 m, while no *R.*

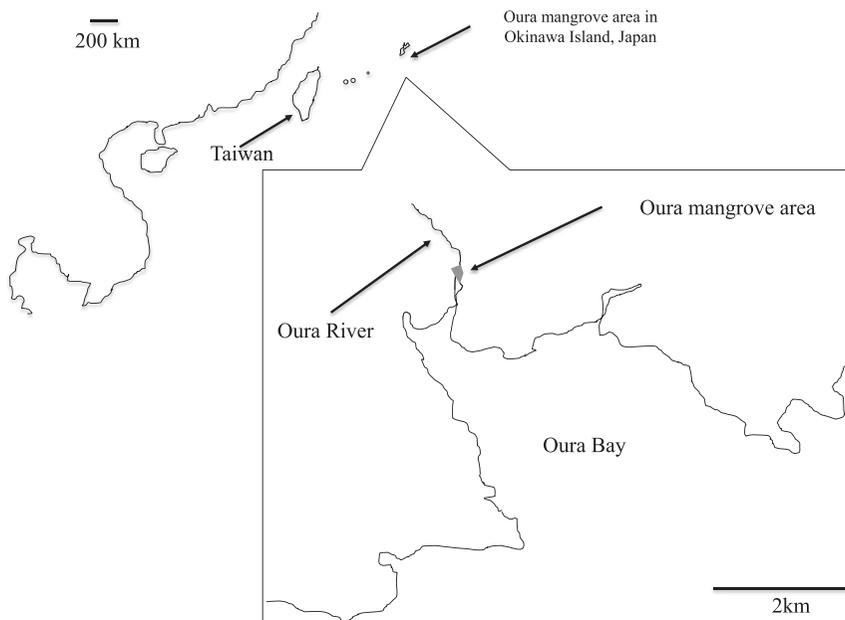


Figure 1. Map of Okinawa island, Japan and the location of the research site in Oura mangrove area.

stylosa trees were recorded. According to Mfilinge et al. (2005), the amounts of litter fall, twigs, flowers, stipules, and leaves in the autumn season and fruits in the summer were higher than those in the other seasons. In this study, little litter was noted, as the study site experiences two tides daily, causing the litter to wash away with the tidal current. These mangroves are enclosed by the Oura Bay and are located approximately 3 km from the open sea. Lugo and Snedaker (1974) described six functional types of mangrove forests, namely, overwash, fringe, riverine, basin, scrub, and hammock. While the Oura Bay mangrove forest cannot be conclusively classified, it is the most similar to the fringe type. Ten sampling points in the Oura mangrove forest area were randomly selected. From each sampling point, surface sediments at depths of 0 - 30 cm were collected using a cylinder (0.05 m in diameter, 30 cm long) at low tide and were weighed.

Sample preparation

The collected sediments were weighed and freeze-dried (Eyela FDU-1200). The dried sediments were ground by hand (roots were visually removed as much as possible) and the soil and roots in the sediments were separated, weighed, and ground for further analysis (< 2 mm).

Roots in the sediments

The carbon content of the roots in the collected sediments was measured using an NC analyzer (SUMIGRAPH NC-220F).

Soil in the sediments

Soil pH (soil:water = 1:2.5) was measured using an electrode (HORIBA D-54). Organic matter in the sediments was decomposed with a 6 % hydrogen peroxide (H_2O_2) solution to measure the particle distribution. After decomposition, the clay and silt fractions were separated from the residual sediments using a sedimentation and decantation method with sodium tripolyphosphate, then the fractions were dried and weighed. The residual material after collecting the clay and silt fractions was the sand fraction, then the sand fraction was dried and weighed.

The total soil carbon content of the sediment soil was measured using an NC analyzer (SUMIGRAPH NC-220F). The extraction and quantification methods for the soluble and insoluble carbon in the sediment

soil have been described elsewhere (Kumada, 1981). Briefly, 0.5 g of air-dried soil was treated with 0.1 M sodium hydroxide (NaOH) for 30 min in a hot water bath (100 °C), and the soil sample was divided into NaOH-soluble and NaOH-insoluble fractions by centrifugation ($17,618 \times g$, 20 min). Next, the NaOH-insoluble fraction was treated with 0.1 M tetrasodium pyrophosphate ($Na_4P_2O_7$) for 30 min in a hot water bath (100 °C) and further divided into the $Na_4P_2O_7$ -soluble and $Na_4P_2O_7$ -insoluble fractions by centrifugation ($17,618 \times g$, 20 min). Both the NaOH- and $Na_4P_2O_7$ -soluble fractions were passed through a membrane filter (0.85 μm) to remove the soil particles, and the carbon content of both soluble fractions was determined by back titration with 0.1 M potassium permanganate ($KMnO_4$) solution. The carbon content of the insoluble fractions was calculated by subtracting the carbon content of both soluble carbons from the total soil carbon content (measured using an NC analyzer).

The methods for extracting and measuring the three types of cations are as follows. First, the cations that were not adsorbed on the soil were removed. Five grams of soil and 50 mL of ultrapure water were placed in a glass tube, and the tube was shaken for 30 min. The supernatant and soil were separated by centrifugation ($17,618 \times g$, 20 min). After removing the cations, three types of cations were sequentially extracted. Exchangeable cations were extracted with 0.1 M ammonium chloride (NH_4Cl) solution. After extracting the exchangeable cations, the NaOH- and $Na_4P_2O_7$ -soluble fractions were extracted using the method described above. The three fractions were treated with 6 % H_2O_2 solution until the solution became transparent due to the decomposition of organic matter in solution. Calcium, magnesium (Mg), sodium (Na), and potassium (K) extracted with the NH_4Cl solution and Al, Ca, Fe, K, Mg, and manganese (Mn) extracted with the NaOH and $Na_4P_2O_7$ solutions were measured by ICP-AES (Shimadzu ICPE-9000). However, Mg and Mn extracted with NaOH could not be detected because of their low amount.

Statistical analysis

All statistical analyses were performed using IBM SPSS Statistics Desktop for Japan version 19.0 (IBM Corp., Armonk, NY, USA). The associations of carbon contents extracted with the NaOH and $Na_4P_2O_7$ solutions with mineral contents were assessed using

Pearson's correlation analysis.

Results

Carbon content of the sediments

The proportion of soil ($98.4 \pm 1.6\%$) in the sediments was significantly higher than that in the roots ($1.6 \pm 1.6\%$), indicating that the soil in the sediments is mineral soil. Soil carbon in the sediment content was below $40 \text{ g}\cdot\text{kg}^{-1}$, as shown in Fig. 2 (a). Mfilinge and Tsuchiya (2008) have reported soil carbon contents ($26.9 - 20.8 \text{ mg}\cdot\text{g}^{-1}$) in the Oura mangrove forest sediments, which were similar to the values in this study (Fig. 2 (a)). The root carbon content was approximately $260 \text{ g}\cdot\text{kg}^{-1}$ (Fig. 2 (a)). These roots were likely *B. gymnorhiza* because it is the dominant tree in this region. The root carbon content recorded in this study (Fig. 2 (a)) was lower than that reported in another study in Sri Lanka for the same mangrove tree (Perera and Amarasinghe, 2013). Although the exact reason could not be clarified, it might be related to differences in the height and diameter at the breast height of *B. gymnorhiza*. Figure 2 (b) presents the contents of soluble (extracted with NaOH and $\text{Na}_4\text{P}_2\text{O}_7$) and insoluble carbon in the mangrove forest soil. The content of soluble carbon extracted with the NaOH and $\text{Na}_4\text{P}_2\text{O}_7$ solutions was below $10 \text{ g}\cdot\text{kg}^{-1}$ and that of insoluble carbon was approximately $10-30 \text{ g}\cdot\text{kg}^{-1}$. Insoluble carbon accounted for most of the soil carbon. Figure 3 presents different types of carbon contents in the sediments at a depth of 30 cm. The results presented in Fig. 3 were calculated

by multiplying the mean contents of total soil carbon, the carbon extracted with NaOH, the carbon extracted with $\text{Na}_4\text{P}_2\text{O}_7$, and the insoluble carbon (in Fig. 2 (b)) by the soil weight at a depth of 30 cm. Root carbon content was calculated by multiplying the root weight at a depth of 30 cm. Soil and root weight are the amount of soil and root contained in the cylinder (0.05 m in diameter, 30 cm long) used for the sediment sampling. In the sediments, the content of insoluble carbon was the highest, followed by that of root carbon. Since both soluble carbon contents were low, it is possible that the soil carbon in the sediments may be difficult to elute from this soil.

Minerals in the sediment soil

Figure 4 shows that the particle distribution in the mangrove soil. The content of sand (approximately $0.7-0.8 \text{ kg}\cdot\text{kg}^{-1}$) was higher than that of silt (approximately $0.05-0.1 \text{ kg}\cdot\text{kg}^{-1}$) and clay (approximately $0.1-0.21 \text{ kg}\cdot\text{kg}^{-1}$). The soil texture is sandy clay loam (SCL) based on the particle distribution (Fig. 4). Sand in the soil may be sea or river sand from the Oura Bay or river. Figure 5 shows the different types of inorganic elements in the soil. As mentioned above, excess elements were removed with water before extracting the inorganic elements. Therefore, the inorganic elements in Fig. 5 are only those adsorbed on the soil particles. Among the exchangeable cations, the amount of Na, which is derived from sea water, was higher than that of the other cations. In the NaOH-extracted fractions, four elements were detected, Al was the most abundant,

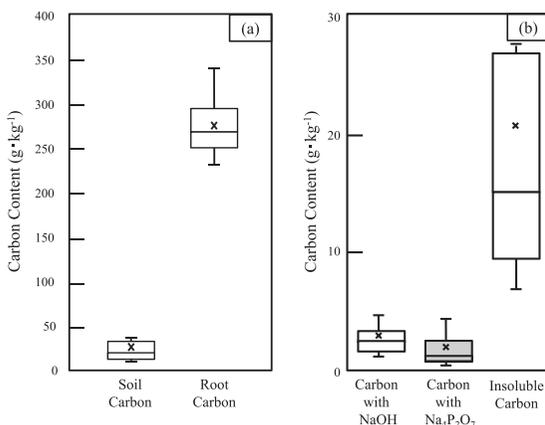


Figure 2. Carbon contents of root and soil in mangrove sediments (a) and soluble and insoluble carbons in mangrove soil (b). Boxplot lines represent median, 25, and 75 percentiles. Cross and black mark represent average and outlier.

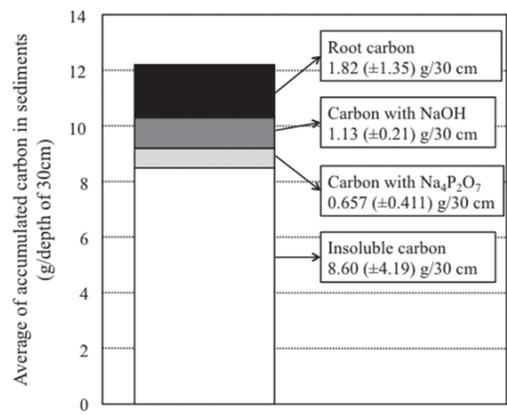


Figure 3. Average of accumulated different types of carbon in sediment at a depth of 30 cm.

followed by K or Fe. In the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted fractions, Fe was the most abundant, followed by Ca, Mg, and K. The detected Al, Fe, and Mn may be derived from clay minerals, and the other elements are from sea or river waters.

Associations of different types of carbon contents with soil minerals

The effects of soil mineral and metal ions on

carbon accumulation in the mangrove forest soil were investigated (Table 1). Sand content was strongly negatively correlated with NaOH-extracted carbon ($r = 0.92$), $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon ($r = 0.85$), and insoluble carbon ($r = -0.92$) content. In contrast, silt and clay contents were positively correlated with the three types of carbon content. Therefore, silt and clay, but not sand, likely contribute to the stability of the carbon in the mangrove forest soil. In addition, there

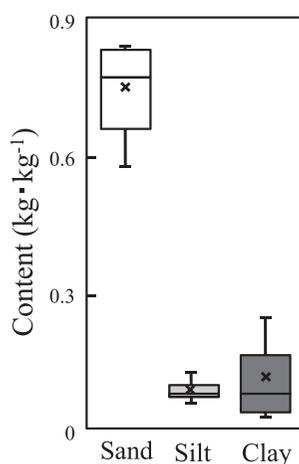


Figure 4. Sand, silt, and clay content of soil in Oura mangrove soils. Boxplot lines represent median, 25, and 75 percentiles. Cross and black mark represent average and outlier.

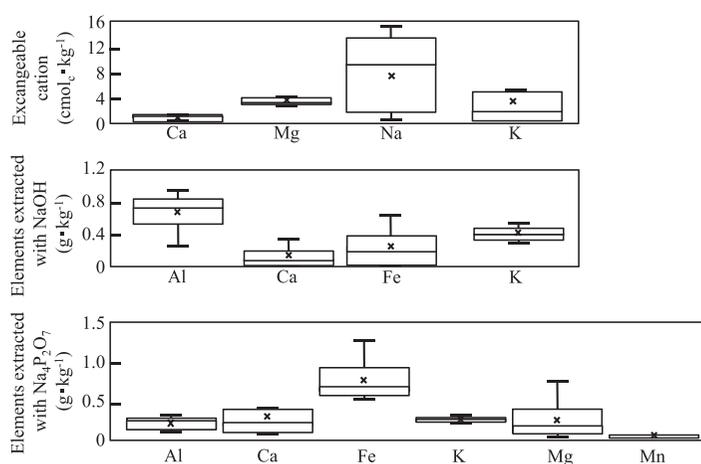


Figure 5. Different types of inorganic elements in Oura mangrove soils. Boxplot lines represent median, 25, and 75 percentiles. Cross and black mark represent average and outlier.

Table 1. The relationships between three types of carbon content and minerals content in mangrove soil by using simple regression analysis. (**: $p < 0.01$, *: $p < 0.05$)

	Sand	Silt	Clay	Extracted with NaOH					
				Al	Ca	Fe	K	Mg	Mn
Carbon extracted with NaOH	-0.92**	0.65	0.64	-0.18	0.86**	0.11	0.39	0.00	0.00
	Sand	Silt	Clay	Extracted with $\text{Na}_4\text{P}_2\text{O}_7$					
				Al	Ca	Fe	K	Mg	Mn
Carbon extracted with $\text{Na}_4\text{P}_2\text{O}_7$	-0.85**	0.66**	0.92**	0.66**	0.87**	0.66**	0.54	0.60	0.77*
	Sand	Silt	Clay	Insoluble carbon					
				Al	Ca	Fe	K	Mg	Mn
Insoluble carbon	-0.92**	0.84**	0.89**						

was a significant positive correlation between the Ca and NaOH-extracted carbon content ($r = 0.86$). Similarly, there were positive correlations between Al ($r = 0.66$), Ca ($r = 0.87$), Fe ($r = 0.66$), and Mn ($r = 0.77$) with the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon content. From this result, it is possible that these metal ions influence the NaOH- and $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon accumulation.

Discussion

The mangrove forest soil in the present study was mineral soil, and it was found that most of the carbon in the soil was insoluble carbon. The insoluble carbon in this study corresponds to humin, which is a part of humic substances. So far, there have been few studies on the quantification of humin, and there are few methods used in this study. Basically, the quantification of humin is evaluated based on the amount of residue after extraction of soluble carbon. Sodium hydroxide, a mixed solution of NaOH and $\text{Na}_4\text{P}_2\text{O}_7$, and methyl isobutyl ketone (MIBK) are used as solvents for extracting soluble carbon (Hayes et al., 2017). Even with these methods (Bogan and Trbovic, 2003; Kohl and Rice, 1998; Almendros and Sanz, 1991), it has been reported that humin accounts for more than 50 % of soil carbon. Humin is considered to account for more than half of the soil carbon and has an important function in improving soil structure (Hayes et al., 2017). The general characteristics of humin include insolubility in any pH solution and a greater resistance to decomposition than other humic substances (humic and fulvic acids) (Rice and MacCarthy, 1990; Rice, 2001). Furthermore, humin is adsorbed strongly on soil minerals (aluminosilicates or clay minerals) (Malekani et al., 1997) and is composed of macro-organic substances (Hayes et al., 2017). From this literature (Hayes et al., 2017), the insoluble carbon in this study may also be macro-organic matter. When separating soil and roots, the roots were visually removed as much as possible, but it could not be denied that macro-organic matter such as these roots is not contained in the insoluble carbon. However, as mentioned above, because the soluble components were extracted in four steps during the preparation process of insoluble carbon (humin) in this study, it is considered that most of the macro-organic matter could be removed in the process. In addition, it was found that clay and silt contributed

significantly to the accumulation of insoluble carbon (Table 1). Therefore, it is possible that the insoluble carbon in this study may be a fraction of what was strongly adsorbed on the soil minerals in the mangrove forest soils.

It was also found that clay and silt contributed to the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon accumulation, but not to the NaOH-extracted carbon accumulation (Table 1). In Table 1, it also shows that metal ions contributed to the NaOH- and $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon accumulation in the mangrove forest soil. Among the extracted metals (Table 1), it was found that Ca contributes to the accumulation of the NaOH extracted carbon, and Al, Ca, Fe, and Mn contributed to the accumulation of the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon. In terrestrial soil, polyvalent cations are required for binding soil minerals and carbon (Kawaguchi and Kyuma, 1959; Ahmed et al., 2002). As shown in the literature, it is possible that the polyvalent cations extracted with NaOH and $\text{Na}_4\text{P}_2\text{O}_7$ from the mangrove forest soil contributes to the accumulation of carbon in the soil. In particular, for the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon, because soil-derived Al and Fe had a significant positive correlation, it could be inferred that these metals contribute significantly to the accumulation in the mangrove forest soil. From these results (Table 1), it was possible to identify metal ions that may affect the accumulation of the carbons extracted with NaOH and $\text{Na}_4\text{P}_2\text{O}_7$. However, in this study, we could not investigate the metal ions that affect the accumulation of insoluble carbon (humin); thus, this must be the focus of future studies.

These results suggest that the soil minerals contributed to the accumulation of the $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon, and insoluble carbon and metal ions contributed to the accumulation of the NaOH- and $\text{Na}_4\text{P}_2\text{O}_7$ -extracted carbon in the mangrove forest soil (mineral soil).

Conclusion

In this study, we investigated the mechanism of carbon from humic substance stability in mineral soil in the mangrove forest soil by studying the soil on the Okinawa Island. Correlations were obtained between $\text{Na}_4\text{P}_2\text{O}_7$ -extracted and insoluble carbon and soil minerals (silt and clay), and it was found that the soil minerals contributed significantly to the accumulation of both types of carbon in the soil. In addition,

among the extracted metals, it was also found that Ca contributes to the accumulation of the NaOH-extracted carbon, and Al, Ca, Fe, and Mn contributed to the accumulation of the Na₄P₂O₇-extracted carbon. These results suggest that the soil minerals or metal ions contributed to the accumulation of the three types of carbon in the mangrove forest soil (mineral soil).

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