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## Soil and Aboveground Carbon Stocks in a Planted Tropical Mangrove Forest (Can Gio, Vietnam)

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► **To cite this version:**

Truong van Vinh, Cyril Marchand, Khanh Tran Vu Linh, Adrien Jacotot, Thanh Nho Nguyen, et al..  
Soil and Aboveground Carbon Stocks in a Planted Tropical Mangrove Forest (Can Gio, Vietnam).  
Wetland Carbon and Environmental Management, 2021. insu-03388141

**HAL Id: insu-03388141**

**<https://insu.hal.science/insu-03388141>**

Submitted on 21 Oct 2021

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## 24 **Abstract**

25

26           Can Gio mangrove is the largest in Vietnam, developing on approximately 35000  
27 hectares. This forest was partially destroyed during the Vietnamese war. A restoration  
28 program was developed between the late 70s and the early 90s, using *Rhizophora apiculata*  
29 Blume propagules. Currently, the Can Gio mangrove forest regenerates naturally and presents  
30 a specific species zonation along the intertidal elevation gradient. *Rhizophora* dominates the  
31 inner forest at the highest elevation, while at an intermediate location, *Rhizophora* and  
32 *Avicennia* cohabit with other scattered species. The lowest position is colonized by *Avicennia*.  
33 Within this context, the main objectives of this study were to determine the soil  
34 physicochemical characteristics, as well as the quality (C/N ratios and  $\delta^{13}\text{C}$ ) and the quantity  
35 (carbon content and stocks) of the organic matter stored beneath each mangrove stand. In  
36 addition, we were interested in determining the above-ground biomass and the total carbon  
37 stocks of the ecosystem (without considering the below-ground biomass). Carbon stocks of  
38 the Can Gio mangrove forest ranged from 150 to 479 Mg C ha<sup>-1</sup>, with up to 86 % of the C  
39 stored in the upper meter of the soil. The inner forest has the highest stock, followed by the  
40 transitional forest, and the fringe forest. The depth extension of the root system of the current  
41 forest was estimated, and its contribution to the soil carbon stock was calculated, using the  
42 adjacent mudflat as a proxy for the antecedent stocks. Our results show that, for the last 40  
43 years, the current mature planted *Rhizophora* forest stored 25.26 Mg C ha<sup>-1</sup>. Consequently,  
44 mangrove plantation and restoration after the war was a success in terms of carbon storing.  
45 We suggest that the destruction of the Can Gio mangrove forests for urban development  
46 would induce the loss of an efficient CO<sub>2</sub> sink.

47

## 48 **1. Introduction**

49 Mangroves forests cover around 137760 km<sup>2</sup> between 30° N and 40° S, with the largest  
50 percentage of their surface observed between 20° N and 20° S (Giri et al., 2011). Mangroves  
51 provide significant ecosystem services such as habitat for various terrestrial and marine animals  
52 that are critical for the coastal biodiversity in the tropics (Alongi, 2008; Mumby et al., 2004;  
53 Nagelkerken et al., 2008; Saenger, 2002), reef against erosion and natural disasters (Alongi,  
54 2008; Barbier, 2006), or even as a trap for suspended and contaminant materials (Kathiresan,  
55 2004; Rivera-Monroy et al., 1999). In addition, mangroves strongly contribute to the economic  
56 growth of emerging countries (Mukherjee et al., 2014), providing charcoal, firewood, and  
57 construction materials for local communities.

58 Due to their high productivity (average of  $218 \pm 72$  Tg C yr<sup>-1</sup>) (Bouillon et al., 2008),  
59 the anoxic character of their soils that limits organic matter degradation (Kristensen et al.,  
60 2008), their high storage capacity (Breithaupt et al., 2012) and their global distribution,  
61 mangroves play a key role in carbon cycling in the coastal ocean (Kauffman et al., 2011).  
62 Mangroves can store carbon both in their biomass and soils; however, this ability depends on  
63 many parameters and may be highly variable. Their biomass varies notably according to latitude  
64 with higher biomass under the tropics (Saenger and Snedaker, 1993), climate with higher  
65 biomass under the wet than under the dry tropics (Adame et al., 2013), and even the age of the  
66 forest (Fromard et al., 1998). In addition, a large part of the carbon content may be stored in  
67 their soil, with up to 98 % and up to 90 % for estuarine and coastal mangroves, respectively  
68 (Donato et al., 2011). Their potential soil carbon storage was recently estimated at 1023 Mg C  
69 ha<sup>-1</sup> (Donato et al., 2011), much higher than other highly productive ecosystems, such as rain  
70 forests (218.6 Mg C ha<sup>-1</sup>), peat swamps (370.1 Mg C ha<sup>-1</sup>), or salt marshes (537.8 Mg C ha<sup>-1</sup>)  
71 (Alongi, 2014). However, mangrove soil carbon stocks depend also on various parameters, such  
72 as the latitudinal position of the mangrove, the tree species developing at the surface, soil

73 salinity, and even nutrient availability (Adame et al., 2013; Alongi, 2002; Jacotot et al. 2018;  
74 Kauffman et al., 2011; Rahman et al., 2015; Sanders et al., 2010; Wang et al., 2013).  
75 Particularly, their position within the tidal zone or along an estuary appears to be critical to their  
76 potential soil carbon storage capacities. For example, in an estuarine mangrove forest  
77 developing in Mexico, Adame et al. (2015) reported soil carbon stocks ranging from 744 to 912  
78 Mg C ha<sup>-1</sup> for the upper estuary and from 537 to 1115 Mg C ha<sup>-1</sup> for the lower estuary.

79         Recently, Vietnam became the first Asian country to implement a national program of  
80 payment for forest environmental services (PFES). In Vietnam, mangroves cover around  
81 270000 ha (FAO, 2015) with more than 80 % distributed in the southern part of the country,  
82 below 10° N latitude (Hawkins et al., 2010). The Can Gio mangrove, located between Ho Chi  
83 Minh City (HCMC) and the South China Sea (Bien Dong in Vietnamese), is actually the largest  
84 contiguous mangrove in Vietnam with a surface of around 35000 ha. During the Vietnamese  
85 war (1964-1971), approximately 57% of this mangrove was destroyed by the spraying of a  
86 mixture of herbicides and defoliants (Ross, 1975). After the war, in the late 70s and early 90s,  
87 a vast reforestation program was implemented by the HCMC Forest Department, and most of  
88 the Can Gio area was replanted using *Rhizophora apiculata* Blume propagules collected in the  
89 Mekong Delta. In January 2000, the mangrove forest of Can Gio was registered in the UNESCO  
90 World Network of Biosphere Reserves list. In addition, the environmental value of the Can Gio  
91 mangrove forest was taken into account in the National Strategy on Climate Change and Sea  
92 Level Rise by the Vietnamese government. Currently, the Can Gio mangrove forest is highly  
93 diversified, with a total of 77 mangrove species (35 true mangroves and 42 associates) (Tri et  
94 al., 2000), whose main species present a specific zonation along the intertidal elevation  
95 gradient. This zonation includes, from the lowest to the highest position: i) a fringe forest that  
96 is dominated by *Avicennia alba*; ii) a transitional forest, composed of a mixture of *R. apiculata*  
97 *A. alba*, *A. officinalis* and sparse *Excoecaria agallocha* and *Sonneratia alba*; and iii) an inner

98 forest, which is mainly composed of mature *R. apiculata*. It was recently demonstrated that the  
99 mineralization of mangrove-derived organic matter has a key role in sustaining coastal food  
100 webs in the Can Gio estuary (David et al. 2018a, David et al. 2019). Recent studies also showed  
101 that the total carbon stocks in the Can Gio mangrove forest could reach 1000 Mg C ha<sup>-1</sup> (Dung  
102 et al., 2016; Nam et al., 2016) and that the below-ground carbon accumulation rates may reach  
103 3.24 Mg C ha<sup>-1</sup> yr<sup>-1</sup> (MacKenzie et al., 2016). However, these studies were interested neither in  
104 soil organic carbon (SOC) quality nor in the influence of soil elevation on carbon stocks of the  
105 different stands.

106 Therefore, the main objectives of this study were: i) to evaluate the soil physicochemical  
107 parameters, the organic matter quality, and the vegetation characteristics of three different  
108 mangrove stands and of the adjacent mudflat developing along the elevation gradient, ii) to  
109 determine the carbon stocks in the biomass and in the first meter of soil, and iii) to calculate the  
110 soil carbon stocks related to forest plantation. We hypothesized that carbon stocks in the soil  
111 and in the biomass depend on the mangrove species and, therefore, to the position of the stand  
112 along the intertidal elevation gradient. To reach our goals, core samples were taken within each  
113 of the four stands. Supplementary cores were also collected in the adjacent mudflat to serve as  
114 a soil reference (i.e., before the colonization of the soil by the mangrove). Then,  
115 physicochemical parameters (pH, redox, and salinity) and the distribution of C/N ratios and  
116  $\delta^{13}\text{C}$  stable isotopes with depth in each stand were measured. Carbon stocks were determined  
117 by combining bulk density and total organic carbon content for the soil and by using biomass  
118 measurements with specific (Vinh et al., 2019) and generic allometric equations for the above-  
119 ground stocks.

## 120 **2. Methods**

### 121 **2.1. Study site**

122           The present study was conducted in the Can Gio mangrove forest in Southern Vietnam  
123 (Fig. 1a). This mangrove forest of around 35000 ha is the largest contiguous mangrove in  
124 Vietnam. Located in the district of Can Gio, one of the 24 districts of Ho Chi Minh City, the  
125 Can Gio mangrove forest is at the deltaic confluence of the rivers Sai Gon, Dong Nai, and Vam  
126 Co, flowing into the East Sea (Bien Dong in Vietnamese). The Can Gio mangrove forest is  
127 composed of several species that follow a specific zonation determined by soil elevation. From  
128 the lowest to the highest intertidal zone, the forest is composed of a mudflat that is totally  
129 denuded of vegetation; a fringe forest, dominated by *Avicennia* spp. (mainly *A. alba*); a  
130 transitional forest, mainly composed of *R. apiculata*, *A. alba*, *A. officinalis*, as well as sparse  
131 *Excoecaria agallocha* and *Sonneratia alba*; and an inner forest dominated by mature *R.*  
132 *apiculata*.

133           The climate in this region is typically monsoonal (type Am in Köppen-Geiger  
134 classification) with two main seasons: a wet season from May to November and a dry season  
135 from December to April. The annual average rainfall is 1816 mm, spread over 154 rainy days,  
136 with approximately 80 % of the rainfall occurring during the wet season. As a result, the flow  
137 of sediments transported by the river crossing the mangrove forest during the wet season is  
138 elevated (~160 million tons (Milliman and Meade, 1983)), leading to the accretion of the deltaic  
139 plain downstream. Conversely, during the dry season, the fresh water flow is strongly reduced,  
140 and the mangrove forest is severely affected by saline intrusions. The Can Gio estuary is  
141 characterized by irregular semi-diurnal tides with a tidal range of 2 to 4 m.

## 142       **2.2. Field measurements and carbon stocks**

### 143       2.2.1 Elevation

144           The elevation of each stand relative to mean sea level (MSL) was measured with a dye-  
145 type tide gauge (Clough, 2014; English et al., 1997; Schmitt and Duke, 2014). First, the cotton

146 tape was soaked in a water soluble food dye and attached to a wooden stake of about 2.5 m in  
147 length. At low tide, the wooden stakes were inserted deeply into the soil starting from the river  
148 edge. The distance between the two columns was 2 m in the mudflat, 5 m in the fringe forest,  
149 and then every 10 m until the inner forest. After high tide, we measured the height of the  
150 washout line above the ground (Albers and Schmitt, 2015; Clough, 2014). In this study, data  
151 from the Vung Tau tide station, Viet Nam (10.3333° N, 107.0667° E) were used to report our  
152 data relative to MSL in this zone. The MSL at Vung Tau tide station in 2016 was 2.16 m. To  
153 determine MSL at the study site, the following equation was used:  $MSL = \text{highest tide value} -$   
154  $MSL \text{ at Vung Tau}$ . Monthly and annual mean sea levels and the daily tide series are collected  
155 and published by the Permanent Service for Mean Sea Level (Pissierssens, 2002).

#### 156 2.2.2. Cores collection and soil physicochemical parameters

157 Soil cores were collected in triplicate with a gouge auger (1 m long, 3 cm wide) attached  
158 to a cross handle. Cores were collected during the dry season of 2016 and the wet season of  
159 2017 from the surface to a depth of 100 cm in each mangrove stand as S1 in the mudflat, S2 in  
160 the fringe forest, S3 in the transitional forest, and S4 in the inner forest (Fig. 1c). The auger was  
161 carefully inserted into the soil to minimize disturbance of the core surface, twisted, and finally  
162 removed, following the method used by MacKenzie et al. (2016). Each core was then separated  
163 into different depth intervals: 0 – 2.5 cm, 2.5 – 5 cm, 5 – 7.5 cm, 7.5 – 10 cm, 10 – 15 cm, 15 –  
164 20 cm, 20 – 25 cm, 25 – 30 cm, and then every 10 cm from 30 to 100 cm. For each interval,  
165 one subsample of a known volume was collected and placed in a zippered bag that was  
166 immediately sealed in aluminum foil to minimize gas exchange and stored in a frozen box until  
167 it reached the laboratory. All samples were, then, dried by freeze-drying at -52 °C until a  
168 constant weight was achieved. Finally, the dry bulk density (DBD) of each sample was  
169 determined by dividing its dry mass by its fresh volume.

170 Additional cores were collected to measure pore-water salinity, redox potential (Eh),  
171 and pH. Pore-water salinity was measured with a hand-held refractometer. Redox potential and

172 pH were measured using, respectively, a combined Pt-Ag/Ag-Cl electrode and a glass electrode,  
173 both connected to a pH/mV/T meter (Marchand et al., 2004). The pH electrode was calibrated  
174 prior to sampling using three standard solutions of pH 4, 7, and 10 at 25 °C (National Institute  
175 of Standards and Technology, USA), and the redox electrode was checked prior to utilization  
176 with a 0.43 V standard solution and demineralized water (Marchand et al., 2004; Thanh-Nho et  
177 al., 2017).

### 178 2.2.3. Soil TOC, TN, $\delta^{13}\text{C}$ , and soil carbon stocks

179 Soil total organic carbon (TOC), total nitrogen (TN), and  $\delta^{13}\text{C}$  values were determined  
180 using an elemental analyzer coupled to an isotope ratio mass spectrometer (Integra2, Sercon,  
181 UK). The analytical precisions of the analyzer were checked using the IAEA-600 caffeine  
182 standard (IAEA Nucleus) and were less than 1 % for TOC, 0.15 % for N, and 0.3 % for  $\delta^{13}\text{C}$ .  
183 All the analyses were performed at the French Institute for the Sustainable Development (IRD)  
184 of Noumea, New Caledonia, France. The  $\delta^{13}\text{C}$  values are reported permil (‰) deviations from  
185 a Pee-Dee Belemnite (PDB) limestone carbonate as the standard using the following equation:

$$186 \quad \delta^{13}\text{C} (\text{‰}) = \left( \frac{\frac{^{13}\text{C}}{^{12}\text{C}}_{\text{sample}}}{\frac{^{13}\text{C}}{^{12}\text{C}}_{\text{standard}}} - 1 \right) \times 1000 \quad (\text{Eq. 1})$$

187 The soil carbon stocks of each mangrove stand ( $\text{Mg C ha}^{-1}$ ) were determined using the  
188 following equation:

$$189 \quad \text{Soil carbon stock (Mg C ha}^{-1}\text{)} = \text{TOC (\%)} * \text{DBD (g cm}^{-3}\text{)} * \text{depth interval (cm)} \quad (\text{Eq. 2})$$

### 190 2.2.4. Above-ground carbon stocks

191 In November 2016, three transects (150 m long and 20 m wide) were established  
192 following an elevation gradient in the mangrove forest (Fig. 1c). All transects encompassed the  
193 four different stands of the mangrove (i.e., the mudflat, the fringe, the transitional, and the inner  
194 forests). All trees within the transects were counted and their DBH at 1.3 m above the soil were  
195 measured, except for *R. apiculata*, whose diameters were measured just above the highest prop

196 root.

197 For *R. apiculata*, the above-ground biomass was calculated using the allometric  
198 equation specifically developed for this species in Southern Vietnam (Vinh et al., 2019):

$$199 \quad WR_{\text{Total}} = 0.38363 * DBH^{2.2348} \quad (\text{Eq. 3})$$

200 For the other species, the above-ground biomass was determined using the common allometric  
201 equations developed by Komiyama et al. (2005).

$$202 \quad W_{\text{Total}} = 0.251 * DBH^{2.46} \quad (\text{Eq. 4})$$

203 where, in Eq. 3,  $WR_{\text{Total}}$  is the total above-ground biomass of *R. apiculata* (kg), and D is the  
204 diameter above the highest prop root of *R. apiculata* (cm); and in Eq. 4.  $W_{\text{Total}}$  is the total above-  
205 ground biomass biomass of the other species (kg), and DBH is the diameter at breast height of  
206 the other species (cm).

207 Above-ground carbon stocks were then estimated by multiplying the above-ground  
208 biomass by a carbon conversion factor. Two conversion factors were used in this study: 0.4409  
209 for *R.apiculata*, which is the specific carbon conversion factor for this species (Vinh et al.,  
210 2019), and 0.451 for the other species (Hiraishi et al., 2014). Eventually, the total above-ground  
211 carbon stocks per area ( $\text{Mg C ha}^{-1}$ ) were calculated by summing the above-ground carbon stock  
212 of *R. apiculata* and the one for the other trees using the tree density per hectare.

### 213 **2.3. Statistical analyses**

214 A parametric two-way analysis of variance (ANOVA) was used to test the significant  
215 effects ( $p < 0.05$ ) of seasons and sites on Eh, pH, and pore-water salinity. A two-way ANOVA  
216 was also applied to assess the significant differences ( $p < 0.05$ ). A Student's t-test was used to  
217 test significant differences ( $p < 0.05$ ) between  $\delta^{13}\text{C}$  values, C/N ratios, and C concentrations of  
218 an upper layer from 0 to 40 cm and a lower layer from 40 to 100 cm, and for Eh and pH for the  
219 transitional forest and inner forest. All statistical analyses were performed using XLSTAT  
220 software version 2017.4 for Mac OS10.13.4.

## 221 **3. Results**

### 222 **3.1. Can Gio mangrove distribution**

223 The relative elevations of the soil surface at the four sampling stations increased from the  
224 river toward the inner forest. The *A. alba* forest developed between +20 and +30 cm above  
225 MSL, the transitional forest between +30 and + 40 cm, and the *Rhizophora* stand between +40  
226 and + 75 cm above MSWL (Fig. 2).

227 The fringe forest had a mean DBH value of 10.7 cm, a tree density of 1327 ind ha<sup>-1</sup>, and  
228 a basal area of 11.9 m<sup>2</sup> ha<sup>-1</sup>. The latter parameter increased to the inner forest with a value  
229 reaching 28.6 cm; the inner forest was also characterized by the highest mean DBH, 18.6 cm.  
230 The transitional forest was characterized by the highest tree density, 3727 individuals per  
231 hectare (ind ha<sup>-1</sup>), and the lowest DBH, 8.5 cm (Table 1).

### 232 **3.2. Soil physicochemical parameters (pore-water salinity, Eh, and pH)**

233 Pore-water salinity remained relatively stable along the core profiles during both seasons  
234 in the mudflat as well as in the fringe and the transitional forests, with values ranging from 10.0  
235 to 23.3, 18.0 to 28.3, and 12.0 to 30.0, respectively (Fig. 3 a, d, g). However, in the inner forest,  
236 pore-water salinity increased from 10.0 and 27.3 at the soil surface to 18.0 and 31.7 at 20 cm  
237 depth, during the dry and the wet season, respectively. Below 20 cm, pore-water salinity  
238 remained stable until 100 cm of depth (Fig. 3 l). When integrating the entire sampled profile,  
239 the season had a significant effect ( $p < 0.001$ ) on pore-water salinity, with higher values during  
240 the dry season in all stands, with the exception of the transitional forest where no significant  
241 differences ( $p > 0.05$ ) in mean pore-water salinity were observed. In addition, pore-water  
242 salinity was significantly different between the four zones in both seasons ( $p < 0.001$ ). Mean  
243 values were  $20.07 \pm 1.78$  and  $12.47 \pm 3.48$  in the mudflat,  $24.8 \pm 1.8$  and  $19.8 \pm 1.3$  in the fringe  
244 forest,  $19.0 \pm 1.8$  and  $19.5 \pm 3.5$  in the transitional forest, and  $29.5 \pm 2.8$  and  $16.3 \pm 4.5$  in the  
245 inner forest, for the dry and the wet season, respectively. Consequently, pore-water salinity

246 increased along the intertidal elevation gradient with lower values at the lowest position and  
247 the higher values in the higher position.

248 In the four zones, Eh values decreased with depth, from 150 to -50 mV in the mudflat,  
249 from -50 to -250 mV in the fringe and the transitional forests and from -50 to -400 mV in the  
250 inner forest (Fig. 3 b, e, h, m). When integrating the whole cores, from the top to the bottom, at  
251 100 cm depth, Eh significantly ( $p < 0.001$ ) decreased from the mudflat to the inner forest. Mean  
252 values were  $-10.7 \pm 104.9$  and  $46.6 \pm 90.9$  mV in the mudflat,  $-52.2 \pm 70.0$  and  $-107.9 \pm 84.1$   
253 mV in the fringe forest,  $44.7 \pm 104.5$  and  $-118.9 \pm 93.6$  mV in the transitional forest, and  $-268.9$   
254  $\pm 185.9$  and  $-117.4 \pm 57.2$  mV in the inner forest, for the dry and the wet season, respectively.  
255 However, although Eh was higher during the dry season than during the wet season, these  
256 differences were not significant in any of the stands ( $p > 0.05$ , Table 2). Finally, Eh values in  
257 the upper layer (0 – 40 cm) were significantly different ( $p < 0.05$ ) from the lower layer (40 –  
258 100 cm) for both the transitional forest and inner forests.

259 Concerning pH, values did not vary significantly during both seasons in the mudflat,  
260 with values ranging from 6.9 to 7.5 (Fig. 3c, f, k and n). For the other stands, pH values varied  
261 with depth and with season, from 6.2 to 7.6. When integrating the whole core (i.e., from the soil  
262 surface to 100 cm depth), pH values were significantly different between stands ( $p < 0.001$ ),  
263 with higher mean values in the mudflat, followed by the fringe and the transitional forests, and  
264 with the lowest mean value in the inner forest. In addition, there were significant differences ( $p$   
265  $< 0.05$ ) between the upper layer (0 – 40 cm) and the lower layer (40 – 100 cm) for both the  
266 transitional and inner forests.

### 267 **3.3. Soil organic matter characteristics (DBD, TOC, TN, $\delta^{13}\text{C}$ , and C/N ratios)**

268 For all stands, DBD values remained stable along the soil profile, from the surface to  
269 100 cm depth. However, mean DBD values for the first meter of soil were statistically different  
270 ( $p < 0.001$ ) between each stand. The mean DBD values were  $0.52 \pm 0.06$ ,  $0.62 \pm 0.02$ ,  $0.63 \pm$

271 0.03, and  $0.64 \pm 0.08 \text{ g cm}^{-3}$  in the mudflat, the fringe, the transitional, and the inner forests,  
272 respectively.

273 In the mudflat and in the fringe forest, TOC values were stable from the soil surface to  
274 the bottom of the core, with mean values of  $2.42 \pm 0.34$  and  $3.2 \pm 0.27 \%$ , respectively (Fig. 4  
275 a). In the transitional and the inner forests, TOC values between the upper (0 – 40 cm) and the  
276 deep layers (40 – 100 cm) were significantly different ( $p < 0.05$ ). Mean TOC values were  $4.14$   
277  $\pm 1.05$  and  $7.03 \pm 3.10 \%$  for the transitional forest and  $4.36 \pm 1.31$  and  $6.75 \pm 2.80 \%$  for the  
278 inner forest for the upper and the deep layers, respectively (Fig. 4 a). In addition, the average  
279 TOC values for the first meter of soil were significantly different ( $p < 0.001$ ) between the four  
280 zones.

281 C/N ratios were relatively stable throughout the entire sampled soil profile in the mudflat  
282 and in the fringe forest with values ranging from 11.2 to 12.1 and from 11.2 to 15.7, respectively  
283 (Fig. 4 b). However, similar to TOC, C/N ratios between the upper 40 cm of soil in the  
284 transitional and inner forests were statistically different ( $p < 0.05$ ) when compared to the lower  
285 60 cm. The mean values of C/N ratios were 12.2 and 18.2 for the transitional forest and 17.5  
286 and 25.7 for the inner forest for the upper and the lower layers, respectively (Fig. 4 b). In  
287 addition, when considering the complete sampled profile, C/N ratios were significantly  
288 different ( $p < 0.001$ ) between the four stands and increased landward. The mean values were  
289  $11.61 \pm 0.22$ ,  $12.08 \pm 1.09$ ,  $14.44 \pm 4.53$ , and  $22.78 \pm 5.49$  for the mudflat, the fringe forest, the  
290 transitional forest, and the inner forest, respectively.

291 Concerning  $\delta^{13}\text{C}$ , in the mudflat and in the inner forest, the values slightly increased  
292 from  $-30 \text{ ‰}$  and  $-32 \text{ ‰}$  at the soil surface to  $-27 \text{ ‰}$  and  $-31 \text{ ‰}$  at 20 cm depth and then remained  
293 relatively stable until the bottom of the cores (Fig. 4 c). In the fringe forest,  $\delta^{13}\text{C}$  values rapidly  
294 dropped from  $-27 \text{ ‰}$  at the soil surface to  $-32 \text{ ‰}$  at 10 cm depth, and then gradually increased  
295 to a mean value of  $-29 \text{ ‰}$  from 20 to 80 cm depth. However, a second rapid drop to  $-34 \text{ ‰}$  was

296 observed in the profile at 90 cm depth (Fig. 4 c). Finally, in the transitional forest,  $\delta^{13}\text{C}$  values  
297 decreased from -29 ‰ at the soil surface to -30 ‰ at 25 cm depth and then remained stable  
298 until 100 cm depth (Fig. 4 c). When integrating the entire core, the mean  $\delta^{13}\text{C}$  were significantly  
299 different between the four zones ( $p < 0.001$ ), with mean values of  $-28 \pm 0.8$  ‰,  $-30 \pm 2.3$  ‰,  $-$   
300  $29 \pm 1.4$  ‰, and  $-31 \pm 0.3$  ‰ in the mudflat, the fringe, the transitional forest, and the inner  
301 forests, respectively.

### 302 **3.4. Carbon stocks in the above-ground biomass and in the soils of the different stands**

303 Above-ground carbon stocks were estimated at  $24.3 \pm 5.1$  Mg C ha<sup>-1</sup> for the fringe  
304 forest,  $91.7 \pm 29.4$  Mg C ha<sup>-1</sup> for the transitional forest, and  $118.8 \pm 9.5$  Mg C ha<sup>-1</sup> for the inner  
305 forest. In addition, above-ground carbon stocks were significantly different between the  
306 different stands ( $p < 0.001$ ). No above-ground biomass was measured in the mudflat due to the  
307 absence of vegetation in this zone. Soil carbon stocks increased with the elevation gradient,  
308 with higher values in the inner forest ( $360.4$  Mg C ha<sup>-1</sup>), followed by the transitional forest  
309 ( $219.8$  Mg C ha<sup>-1</sup>), the fringe forest ( $157.6$  Mg C ha<sup>-1</sup>), and the mudflat ( $150.2$  Mg C ha<sup>-1</sup>) (Fig.  
310 5). In addition, soil carbon stocks were significantly different between the different stands ( $p <$   
311  $0.001$ ). Eventually, the total carbon stocks (without the below-ground biomass) were  $150.2 \pm$   
312  $19.6$ ,  $181.9 \pm 24.9$ ,  $311.5 \pm 28.1$ , and  $479.2 \pm 32.6$  Mg C ha<sup>-1</sup>, for the mudflat, the fringe, the  
313 transitional forest, and the inner forest, respectively (Fig. 5).

## 314 **4. Discussion**

### 315 **4.1. Mangrove zonation in Can Gio**

316 Between 1978 and 1994, a vast mangrove reforestation program was undertaken with  
317 *R. apiculata* as a primary species (Hong and San, 1993). When *Rhizophora* stands were  
318 established, the mangrove area extended through natural regeneration and rapid colonization of  
319 mudflats along riverbanks notably by *A. alba*, which can be considered as a pioneer species

320 (see for example Balke et al. (2011); Brunt and Davies (2012); Naidoo and Naidoo (2017);  
321 Proisy et al. (2009)) (Fig. 2 a, b). This fringe forest developed at lower elevations as compared  
322 to the *R. apiculata* forest and was separated from the latter by a transitional forest composed of  
323 several species. Mangrove zonation often manifests itself as a mosaic that varies according to  
324 physical, biological, and chemical interactions established between plant and substrate in a  
325 given area. Pore-water salinity was often considered as the main driver of the zonation  
326 (Banerjee et al., 2013; Marchand et al., 2012; 2011), because mangrove plants have different  
327 abilities to cope with this factor (Ellison, 1998; Mckee, 1993; Walsh, 1974). For instance, under  
328 semi-arid climate, pore-water salinity increased landward, where evaporation processes were  
329 intense due to the rare periods of immersions and low precipitation and could reach to a value  
330 greater than 50. As a consequence, *Avicennia* trees, which can cope with high salinity (Kendall  
331 and Skipwith, 1969; Khan and Aziz, 2001; Marchand et al., 2004; Ukpong, 1997), developed  
332 at higher tidal position than the *Rhizophora* trees, which colonized the seaward zone of the  
333 mangrove forest. In Can Gio, the zonation was the opposite because pore-water salinity never  
334 reached such high values. In fact, pore-water salinity increased from the fringe forest to the  
335 interior forest due to its higher elevation that induced more evaporation, but it was never higher  
336 than 30. The Can Gio mangrove forest is an estuarine mangrove with freshwater inputs from  
337 the Sai Gon and the Dong Nai Rivers. Along the estuary, salinity values ranged from 2 to 26  
338 during the year; and even at the mouth of the estuary, salinity never reached the value of  
339 seawater, possibly due to the high freshwater inputs from these two rivers and also from the  
340 Mekong delta, which is further south of the study site (David et al., 2018b; Thanh-Nho et al.,  
341 2018). Furthermore, during the rainy season, the intense rainfall brought additional freshwaters  
342 that induce a dilution of pore-water salinity, with mean values for the mature *Rhizophora* forest  
343 decreasing from 29 to 16. In the latter stand, salinity also increased with depth, notably because  
344 dilution with rainwater occurred in the upper sediment during the rainy season, but also possibly

345 because dissolved salts could migrate at depth through convection processes and accumulate  
346 there (Marchand et al., 2004).

#### 347 ***4.2. Influence of mangrove development on soil properties***

348 Organic carbon (OC) content in mangrove sediments usually ranges from 0.5 % to 15 %,  
349 with a median value of 2.2 % (Kristensen et al., 2008). In the Can Gio forest, soil OC increased  
350 with increasing elevation landward, ranging from 2.42 % in the low intertidal zone to 5.82 %  
351 in the high intertidal zone, indicating that the interior forest had accumulated a larger organic  
352 carbon stock. The interior forest studied here is composed of mature *Rhizophora* trees, which  
353 were planted in 1978. Therefore, mangrove-derived organic matter had accumulated in its soil  
354 for almost 40 years at the date of the coring. *Rhizophora* trees under tropical climates as in  
355 southern Vietnam are highly productive (Alongi, 2014), which can positively increase soil  
356 carbon stocks. In addition, this stand was the furthest from the tidal creek; and as a consequence,  
357 tidal flushing of leaf litter was reduced. Leaf litter can accumulate and increase the soil carbon  
358 stocks. Conversely, due to their more recent development, lower productivity, and proximity to  
359 tidal creeks, the soils of the fringe stand composed of *A. alba* contained less organic carbon  
360 than the mature inland *Rhizophora* forest.

361 These gradients of elevation, length of tidal immersion, and carbon content strongly  
362 influence redox conditions and pH of the soil. Redox potential (Eh) decreased with increasing  
363 elevation from the mudflat to the mature *Rhizophora* stand. Additionally, within the *Rhizophora*  
364 stand, the redox condition became rapidly anoxic with depth (Fig. 3). We suggest that the higher  
365 organic content beneath this stand induced a higher electron acceptor demand, which were less  
366 renewed by the tides due to its high position in the tidal zone. The higher redox values measured  
367 beneath the *Avicennia* stands may also be related to the ability of this mangrove species to aerate  
368 the sediment through its root system as described by Scholander et al. (1955). Hesse (1961)  
369 also observed that *Rhizophora* soils were anoxic most of the time and sulfidic. This difference

370 between the two species was later confirmed in different countries and was suggested to be  
371 related to different organic enrichment of the soil, specific abilities of the root system, and  
372 different positions in the tidal zone (Marchand et al., 2004; Marchand et al., 2011; Mckee,  
373 1993). pH values also varied along the elevation gradient of the studied tidal zone, decreasing  
374 from 6.8 in the mudflat to 6.2 in the mature *Rhizophora* stand. We suggest that the increased  
375 organic content and its decay processes were responsible for this soil acidification. In addition,  
376 in anoxic mangrove soils, sulfides minerals could precipitate (Balk et al., 2016), and slight  
377 modifications of the redox conditions could induce their oxidation, which could result in soil  
378 acidification (Noel et al., 2017; 2014).

#### 379 ***4.3. Characterization of soil organic matter with depth and along the intertidal elevation*** 380 ***gradient***

381 In mangrove soils, organic matter is usually a mixture between autochthonous organic  
382 matter (leaf litter, roots debris, and microphytobenthos) and allochthonous organic matter  
383 derived from marine and/or terrestrial origins (Kristensen et al., 2008). However, in a distinct  
384 area, the respective contribution of each source in the carbon pool depends on several  
385 parameters, including trees productivity, mangrove position, tidal range, freshwater inputs,  
386 position of in the intertidal zone, etc. In the Can Gio mangrove soils,  $\delta^{13}\text{C}$  and C/N ratios ranged  
387 between -28 ‰ to -31 ‰ and between 12 to 22, respectively, and were consistent with those  
388 previously reported (Bouillon et al., 2003; Prasad et al., 2017, Jacotot et al., 2018). However,  
389 OM quality differed along the intertidal elevation gradient, with higher C/N ratios and depleted  
390  $\delta^{13}\text{C}$  values as the elevation increased. These gradients suggested an increased contribution in  
391 the upper intertidal zone of mangrove-derived organic matter, characterized by elevated C/N  
392 ratios and depleted  $\delta^{13}\text{C}$  values (Bosire et al., 2005; Jennerjahn and Ittekkot, 1997; Kristensen  
393 et al., 2008; Marchand et al., 2005). These results were consistent with our previous hypothesis  
394 of a higher enrichment of the soil resulting from mangrove development and a greater

395 contribution of *Rhizophora* leaf litter in the inner forest due to the higher productivity of the  
396 stand, the limited tidal export, and the anoxic character of the soils that limits OM decay process  
397 and favors its accumulation. Conversely, towards the tidal creek side of the mangrove, the  
398 enriched  $\delta^{13}\text{C}$  and lower C/N ratios, close to 12, suggested a greater contribution of  
399 phytoplankton or phytobenthos and/or more degraded higher plant debris. In a recent study  
400 (Vinh et al., 2020), we showed that leaf litter in the *Avicennia* stand was more rapidly  
401 decomposed, and that decay rates were even enhanced during the monsoon. Consequently, we  
402 suggest that the position of the stand along the intertidal elevation gradient and its species  
403 composition influenced the organic matter characteristics of the soil.

404 Surprisingly, an organic-rich layer was observed at depth beneath the transitional and  
405 the inner forests from 40 to 100 cm depth. In the mangroves of southern Vietnam, MacKenzie  
406 et al. (2016) determined an average vertical accretion rate of around  $0.99 \pm 0.09 \text{ cm yr}^{-1}$ . This  
407 vertical accretion rate was relatively elevated and reflected notably the high sedimentation rate  
408 characterizing most Asian estuaries. During the wet season, strong rainfalls induce high erosion  
409 rates in the upper watersheds. As a result, high quantity of sediments are transported by the  
410 rivers and deposited along the coastlines, notably in mangrove forests. Following the accretion  
411 rate determined by MacKenzie et al. (2016), the upper layer observed in this study (from 0 to  
412 40 cm depth) would have started to be deposited 40 years ago, which almost corresponds to the  
413 beginning of the reforestation program started in 1978 (Hong and San, 1993). Consequently,  
414 we suggest that the upper layer corresponds to the development of the current forest (0 – 40  
415 cm), and the lower layer (40 – 100 cm) accumulated before mangrove destruction during the  
416 Vietnamese war. Interestingly, this buried layer was at least 50 % enriched in carbon compared  
417 to the upper layer for the same sediment thickness and was also characterized by higher C/N  
418 ratios (9.7 vs. 22.3 and 14.0 vs. 37.1, for the upper and the lower layers, respectively) and

419 depleted  $\delta^{13}\text{C}$  values (-30.7‰), which suggested that the former forest was more productive  
420 and/or accumulated organic carbon during a long period.

#### 421 ***4.4. Influence of mangrove development on carbon stocks***

422 Carbon stocks in the above-ground biomass were  $118.8 \pm 9.5 \text{ Mg C ha}^{-1}$  for the inner  
423 forest,  $91.7 \pm 29.4 \text{ Mg C ha}^{-1}$  for the transitional forest, and  $23.3 \pm 5.1 \text{ Mg C ha}^{-1}$  in the fringe  
424 forest. Differences between stands may be related to the age of the forests, as the inner forest  
425 was planted 40 years ago, while the other stands naturally regenerated recently. In addition,  
426 *Rhizophora* trees that dominates the inner forest and extends into the transitional forest are  
427 generally more productive than *Avicennia* ones (Komiyama et al., 2008) that colonize the fringe  
428 forest. Nevertheless, these results were in the range of those previously observed from other  
429 studies in Southern Vietnam, with values ranging from 13.4 to 210.7  $\text{Mg C ha}^{-1}$  (Dung et al.,  
430 2016; Tue et al., 2014). However, the above-ground carbon stocks in Can Gio were much lower  
431 than those in other mangrove forests. For example, in Malaysia, the above-ground carbon stocks  
432 reached 202.9  $\text{Mg C ha}^{-1}$  (Putz and Chan, 1986). This difference may be explained by the age  
433 of the forest, as the forest in Malaysia has developed for 80 years—twice the age of the one in  
434 Can Gio, and by silvicultural activities (Vinh et al., 2019).

435 In Can Gio, soil carbon stocks ranged from 150 to 360  $\text{Mg C ha}^{-1}$ , which was consistent  
436 with and even higher than the reported values for other mangrove forests in Vietnam, ranging  
437 from 144 to 233  $\text{Mg C ha}^{-1}$  (Dung et al., 2016; Tue et al., 2014). However, these values were  
438 lower than the ones of tropical mangroves that ranged from 337 to 640  $\text{Mg C ha}^{-1}$  (Adame et  
439 al., 2013; Castillo et al., 2017; DeVecchia et al., 2014; Hossain, 2014; Kauffman et al., 2011).  
440 We suggest that partial destruction of the mangrove during the Vietnamese War may have  
441 prevented organic matter to accumulate for a while and probably allowed the existing material  
442 to be eroded by tides and freshwater circulation, explaining these low values for soil carbon  
443 stocks. Soil carbon stocks in the mangrove of Can Gio represented between 70 to 86 % of its

444 total carbon stocks (without the below-ground biomass) and was consistent with other Indo-  
445 Pacific mangrove forests (Donato et al., 2011; Kauffman et al., 2011; Liu et al., 2014).

446         Recently, some authors suggested that integration depth is of major concern when  
447 determining carbon stocks in mangrove forests (Lunstrum and Chen, 2014; Marchand, 2017;  
448 Jacotot et al., 2018). In Can Gio, integrating the soil carbon stocks down to one meter takes into  
449 account the stocks linked to the development of the current forest, replanted after the  
450 Vietnamese war as discussed above, and a part of the stocks that accumulated before the  
451 destruction of the forest. When considering only the development of the current forest (i.e., 0 –  
452 40 cm, assuming a sedimentation rate of  $0.99 \pm 0.09 \text{ cm yr}^{-1}$  as discussed above), soil carbon  
453 stocks were much at  $85.7 \pm 33.92 \text{ Mg C ha}^{-1}$  in the inner forest. We did not calculate this stock  
454 for the other stands considering that it was a natural regeneration that occurred later than the  
455 planting and had an OM quality depth profile different from the mature *Rhizophora* forest. In  
456 addition, knowing the age of the forest and the amount of carbon stored in its soil since its  
457 development allowed the precise contribution of this forest to the enrichment in soil organic  
458 matter to be determined. Doing this, the antecedent carbon stocks (i.e., the stocks that were  
459 present before the apparition of the forest) must be determined (Lal, 2005). In our study, the  
460 stocks in the mudflat were chosen as a proxy for the antecedent carbon stocks. Therefore, for  
461 the last 40 years, the actual forest contributed to an enrichment of  $25.26 \text{ Mg C ha}^{-1}$  in the inner  
462 forest. As a result, the carbon burial rate in the mature *Rhizophora* was  $\sim 0.6 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ ,  
463 which is lower than the global value of  $1.35 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$  reported by Bouillon et al. (2008).  
464 However, our result may be underestimated, considering that the mudflat was probably enriched  
465 by mangrove-derived organic matter, as suggested by the depleted  $\delta^{13}\text{C}$  values and the high  
466 TOC content. Nevertheless, this study demonstrated that increasing mangrove areas by either  
467 restoration or expansion is an important way of increasing carbon storage in the coastal ocean,  
468 and mangroves must be considered in future climate change mitigation programs.

469 **5. Conclusions**

470 Degraded by the spraying of defoliants during the Vietnam War, mangrove forests in  
471 Can Gio Estuary successfully recovered through replantation and natural regeneration. They  
472 now store a high amount of carbon both in their biomass and in their soils. Their destruction for  
473 infrastructure development along the coastline would result in the loss of an efficient CO<sub>2</sub> sink.

474 The main conclusions of this study can be summarized as follow:

475 1. C stocks in the above-ground biomass were significantly different between stands,  
476 increasing landward, reaching up to  $118.8 \pm 9.5$  Mg C ha<sup>-1</sup> for the mature *Rhizophora* stand.  
477 Differences in carbon stocks in the above-ground biomass between stands were related to  
478 different forest ages, mangrove species, and tree densities, the latter being managed by thinning.

479 2. The specific zonation, with planted *Rhizophora* trees at the highest elevation in the  
480 tidal zone (with a limited pore-water salinity value due to a monsoon-dominated climate), and  
481 natural colonization of the river banks by *Avicennia* trees resulted in gradients in the soil  
482 physicochemical properties from the mudflat to the inner forest. Due to their more recent  
483 development, lower productivity, and proximity to tidal creeks, the soils of the fringe stand  
484 composed of *A. alba* contained less organic carbon than the mature inland *Rhizophora* forest.  
485 These gradients of elevation and of carbon content strongly influenced the redox conditions and  
486 pH in the soil; with both decreasing from the mudflat to the mature *Rhizophora* stand.

487 3. Regarding soil organic matter quality, the  $\delta^{13}\text{C}$  and C/N ratio values suggested a  
488 higher contribution of mangrove-derived organic matter in the inner forest, most probably  
489 because of its age, the high productivity of the stand, and the distance from the tidal creek that  
490 limits leaf litter flushing. At depth, beneath the mature *Rhizophora* stand and the transitional  
491 forest, an increased organic content combined with depleted  $\delta^{13}\text{C}$  values and C/N ratio increase  
492 suggested an elevated contribution of vascular plant debris to the soil organic matter pool. We

493 suggest that this enrichment reflected the past mangrove forest before its destruction during the  
494 war.

495 4. Soil carbon stocks in the mature *Rhizophora* forest, down to one meter, represented  
496 almost three times the stock in the above-ground biomass. However, when considering only the  
497 upper soil, which was related to current forest development as evidenced by  $\delta^{13}\text{C}$  values and  
498 C/N ratios, stocks in the soil and in the above-ground biomass were similar, and the carbon  
499 burial rate was lower than  $1 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ .

500 In a future research effort, the net ecosystem productivity of the Can Gio mangrove  
501 forest, Southern Vietnam's largest, should be studied, possibly using the eddy-covariance  
502 technique. The influence of the monsoon on its productivity should also be considered.

503

## 504 Acknowledgements

505 This study was supported by an ARTS grant from IRD, and by the Air Liquide  
506 Foundation. The authors would like to thank Mr. Tran Nhat Toan, Mr. Le Anh Tuan, and Mr.  
507 Truong Van Hoang for their valuable assistance during fieldwork. We also thank Leocadie  
508 Jamet and Jean-Louis Duprey from the LAMA laboratory for their assistance in the analyses.  
509 We also would like to express our gratitude to the authorities of Can Gio District People  
510 Committee, CGMBR, Vam Sat Ecological Tourist Centre – Phu Tho Tourist Company who  
511 facilitated the collection of field data.

512

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803 Table 1. Vegetation structure in the different zones. MF: Mudflat; FF: Fringe forest; TF:  
 804 Transition forest; IF: Interior forest; Aa: *Avicennia alba*; Ao: *Avicenia officinalis*; Ct: *Ceriops*  
 805 *tagal*; Aa: *Sonneratia alba*; Ra: *Rhizophora apiculata*, AGC above-ground C stock, SC soil C  
 806 stock.

Sites	Species	DBH (cm)	Tree density ha <sup>-1</sup>	Basal area (m <sup>2</sup> ha <sup>-1</sup> )
MF	-	-	-	-
FF	<i>Aa, Ao</i>	10.7	1327	11.9
TF	<i>Aa, Ao, Ct, Sa, Ra</i>	8.5	3727	21.2
IF	<i>Ra</i>	18.6	1127	28.4

807

808 Table 2: Two-way ANOVA tests for pH, Eh and salinity values showing the effect of sampling  
 809 site and seasons on soil parameters. \* and \*\*\* indicate statistically significant effects for p-  
 810 Values < 0.05 and < 0.001, respectively .ns means no statistically significant difference (p >  
 811 0.05).

Parameters	n	Source of variation		
		Sites	Seasons	Interaction
pH	150	48.9***	5.9*	22.8***
Eh	150	19.1***	1.4 ns	11.9***
Pore-water salinity	150	1137.7***	203.4***	23.7***

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813

814 **Figures captions**

815

816 Figure 1: Study sites location. (A) Vietnam map, (B) Can Gio Estuary and (C) Site study with  
817 three transects from the mudflat to the inner forest.

818

819 Figure 2: Mangrove distribution along the transect at Can Gio mangrove, (a) and (b) distribution  
820 of mangrove species from river towards the land, (c) elevation of the intertidal zone and  
821 mangrove zonation.

822

823 Figure 3: Mean salinity, Eh and pH values beneath the different mangrove zones studied:  
824 Mudflat (a, b, c), fringe forest (d, e, f), transitional forest (g, h, k), and inner forest (l, m, n).  
825 Orange lines present the values measured during the dry season and blue lines present the values  
826 measured during the rainy season.

827

828 Figure 4: C contents (a), C/N ratios (b) and  $\delta^{13}\text{C}$  values (c) profiles. Black, light brown, orange,  
829 blue, green lines represent mudflat, fringe forest, transition forest, and mature *Rhizophora*, (d)  
830 picture of buried dead trunk of *Rhizophora* below the actual root system.

831

832 Figure 5: Ecosystem C stocks in the different sites along the elevation gradient. Green bars  
833 represent above-ground C stock, grey bars represent soil C stocks from 0 to 40 cm depth, orange  
834 bars represent below-ground C stocks from 40 to 100 cm depth, blue bars represent below-  
835 ground C stock from 0 to 100 cm depth. Solid vertical black lines present the standard deviation  
836 (SD).

837

838