

Continuous Maize Cropping Accelerates Loss of Soil Organic Matter in Northern Thailand as Revealed by Natural ^{13}C Abundance

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1 **Continuous maize cropping accelerates loss of soil organic matter in northern Thailand**
2 **as revealed by natural ¹³C abundance**

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16
17 **Running title:** Cultivation effects on soil carbon stocks

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19 cultivation; soil organic matter

20 **Abstract**

21 *Aims:* The loss of soil organic matter (SOM) has widely been reported in the tropics after
22 changing land use from shifting cultivation to continuous cropping. We tested whether
23 continuous maize cultivation accelerates SOM loss compared to upland rice and forest fallow.

24 *Methods:* Because litter sources include C4 plants (maize in maize fields and *Imperata* grass in
25 upland rice fields) in Thailand, C3-derived and C4-derived SOM can be traced using the
26 differences in natural ¹³C abundance ($\delta^{13}\text{C}$) between C3 and C4 plants. We analyzed the effects
27 of land use history (cultivation or forest fallow period) on C stocks in the surface soil. Soil C
28 stocks decreased with the cultivation period in both upland rice and maize fields.

29 *Results:* The rate of soil organic carbon loss was higher in maize fields than in upland rice fields.
30 The decomposition rate constant (first order kinetics) of C3-plant-derived SOM was higher in
31 the maize fields than in the upland rice fields and the C4-plant-derived SOM in the forest fallow.
32 Soil surface exposure and low input of root-derived C in the maize fields are considered to
33 accelerate SOM loss. Soil C stocks increased with the forest fallow period, consistent with the
34 slow decomposition of C4-plant-derived SOM in the forest fallows.

35 *Conclusions:* Continuous maize cultivation accelerates SOM loss, while forest fallow and
36 upland rice cultivation could mitigate the SOM loss caused by continuous maize cultivation.

37

38

39 **Introduction**

40 In Southeast Asia, traditional shifting cultivation has been replaced by continuous cropping
41 systems (Kyuma and Pairintra, 1983). In northern Thailand, upland rice is cultivated between
42 forest fallow periods for subsistence, but continuous maize cultivation has increased along
43 paved roads with infrastructure improvement (Bruun et al., 2017). Intensive agriculture without
44 organic amendment leads to a loss of soil organic matter (SOM), which is essential for
45 increasing plant productivity and mitigating soil acidification (Kimetu et al., 2008; Fujii et al.,
46 2009; Jaiarree et al., 2011).

47 The intensification of agriculture, including tillage, generally risks increasing microbial
48 decomposition activities and soil erosion (Kimetu et al., 2008), but the impact could vary with
49 crop plants. Upland rice develops larger root systems than maize, especially under nitrogen and
50 water stress (Kondo et al., 2000). Because root litter is less decomposable than leaf litter (Fujii
51 et al., 2019), the lower shoot/root ratios of upland rice could provide greater root-derived C
52 input than maize of the same biomass (Kondo et al., 2000). In maize fields, the low root-derived
53 C input could limit the supply of SOM precursors, despite the high primary productivity
54 (Carvalho et al., 2017). Global assessment also suggests lower SOM stocks in maize fields
55 compared to other crops (West et al., 2002). We hypothesized that continuous maize cultivation
56 results in greater SOC loss compared to forest fallow or upland rice cultivation.

57 Because SOM gain or loss is dependent on the balance between C inputs (mainly litter
58 inputs) and outputs (heterotrophic respiration, leaching, and erosion), the effects of land use on
59 SOM stocks can be quantified using annual soil C budgets (Fujii et al., 2009) or long-term
60 monitoring of soil organic carbon (SOC) stocks (Fujii et al., 2019, 2020). Alternatively, SOC
61 stocks can be compared between sites that share soil attributes with different land-use histories
62 (cultivation or fallow periods). Although our study site is in a remote part of northern Thailand,
63 its land-use history has been monitored continuously (Sakai, 2005; Fig. 1), enabling us to
64 compare the effects of land-use change on SOC stocks.

65 Continuous maize cropping results in a loss of forest-derived SOM and a gain of maize-
66 derived SOM. Because maize (C4 plant) and most woody species (C3 plants) have different
67 natural ^{13}C abundances ($\delta^{13}\text{C}$) due to their different photosynthetic pathways (Yoneyama et al.,
68 2006), C3-derived and C4-derived SOM can be traced in maize fields. In addition, while upland
69 rice is a C3 plant, the dominant weed (*Imperata cylindrica*) of upland rice fields is a C4 plant.
70 This allows us to trace the dynamics of C3- and C4-derived SOM under different fallow and
71 rice cultivation periods in northern Thailand. To test the hypothesis that continuous maize
72 cultivation results in greater SOC stock loss than forest fallow or upland rice cultivation, we
73 compared the SOM dynamics of forest fallow, upland rice, and maize fields.

74

75 **Materials and Methods**

76 **Sampling soil material**

77 Soils were collected in 2014 in Ban Rakpaendin, Chiang Rai Province (19° 50' N, 100° 20' E;
78 697 m a. s. l.), where the mean annual air temperature is 25.0°C and the precipitation is 2084
79 mm yr⁻¹. The forest and forest fallow vegetation were dominated by *Lithocarpus* and *Eugenia*
80 spp., with some planted rubber (*Hevea brasiliensis*) and orange (*Citrus reticulata*) trees. The
81 soils were clayey and classified as Typic Haplustults (Table 1; Soil Survey Staff, 2014). The
82 village land use has been affected by socio-economic development and political instability in
83 Indochina. Before the Indochina War, the *Mieng* (a hill tribe) conducted shifting cultivation of
84 upland rice (Sakai, 2005). After the war, northern Thai (*Khon Muang*) and *Hmong* migrated in
85 1982 and 1987, respectively, and started cultivating upland rice and maize in the upstream forest
86 (Fig. 1a). After the village protected the upstream forest for water security and a paved highway
87 was constructed in 1995, intensive maize cultivation expanded to the fields close to the highway
88 (Fig. 1a,b). The population increase and influx of refugees increased maize production. The
89 land use history has been recorded in the village (Fig. 1a,b). The cultivation and fallow periods
90 of the sampling locations were determined by interviews with farmers and a field survey, cross-
91 checked using satellite photos (1992, 2002, 2007, 2014) and a land use map (Sakai et al., 2005;
92 Fig. 1b).

93

94 **Calculating the contributions of C3- and C4-plant-derived carbon**

95 The physical fractionation of soil is useful for tracing the pools of labile (free) and stable
96 (mineral-associated) SOM in the light and heavy fractions, respectively (Hassink, 1995; Tan et
97 al., 2007). The SOC in the two fractions was determined as follows. Briefly, 10 g of air-dried
98 soil was dispersed in a sodium iodide (NaI) solution (1.60 g cm^{-3}), centrifuged at 2600 *g*
99 (modified from Spycher et al., 1983), and the light ($< 1.60 \text{ g cm}^{-3}$) and heavy ($> 1.60 \text{ g cm}^{-3}$)
100 fractions were recovered. The total C and N concentrations in soils were measured using a CN
101 analyzer (Vario Max CN; Elementar Analysensysteme, Langensfeld, Germany).

102 Because the surface soil horizon is most sensitive to land use change in a soil profile
103 (Tan et al., 2007), this study focused on the C stocks in the top 0–5 cm of soil, which were
104 calculated as follows:

105
$$\text{Soil C stock (0–5 cm)} = \text{Soil C} \times 0.05 \times 100 \times 100 \times \text{Bulk density}/1000 \quad (\text{Eq. 1})$$

106 where the soil C stock (0–5 cm) (Mg C ha^{-1}), soil C concentration in the light and heavy
107 fractions (g C kg^{-1}), surface 0–5-cm soil volume (m^{-3}) ($= 0.05 \text{ m} \times 100 \text{ m} \times 100 \text{ m}$), and bulk
108 density (Mg m^{-3}) were measured using three replicates of a 0.1-L core per plot.

109 The contributions of C3- and C4-plant-derived SOM to soil C were estimated using the
110 measured C-isotope signatures ($\delta^{13}\text{C}$) and a mass balance approach (Nguyen-Sy et al., 2020). A

111 sample (~50 µg C) was weighed in a tin capsule and the $\delta^{13}\text{C}$ isotope composition was measured
 112 for the litter and soil samples using an online C analyzer (NC 2500; Thermo Fisher Scientific,
 113 Waltham, MA, USA), coupled with an isotope ratio mass spectrometer (MAT252; Thermo
 114 Electron, Bremen, Germany). All $\delta^{13}\text{C}$ values are expressed relative to the international standard
 115 Vienna Pee Dee Belemnite (VPDB): $\delta^{13}\text{C} = (R_{\text{sample}}/R_{\text{VPDB}} - 1)$. The standard deviation for four
 116 replicate combustions of the same standard within a sequence was less than 0.10‰.

117 The sample $\delta^{13}\text{C}$ values depend on the mixture of C3 plant-derived C and C4-plant-derived
 118 C, as expressed in the following equation (Nguyen-Sy et al., 2020):

$$119 \quad \delta^{13}\text{C}(\text{Sample}) = x \times \delta^{13}\text{C}(\text{C3}) + (1 - x) \times \delta^{13}\text{C}(\text{C4}) \quad (\text{Eq. 2})$$

120 where $\delta^{13}\text{C}$ (sample) is the measured $\delta^{13}\text{C}$ value of the sample, $\delta^{13}\text{C}$ (C3) is derived from the
 121 C3-plant-derived C (av. -28.6‰), $\delta^{13}\text{C}$ (C4) is derived from the C4-plant-derived C (av.
 122 -12.5‰), and x denotes the proportion of C3-plant-derived C. The proportions of C3-plant-
 123 derived C (x) and C4-plant-derived C ($1 - x$) were estimated with the following equations:

$$124 \quad \textit{Proportion of C3 derived C} = \frac{\delta^{13}\text{C}(\text{Sample}) - \delta^{13}\text{C}(\text{C4})}{\delta^{13}\text{C}(\text{C3}) - \delta^{13}\text{C}(\text{C4})} \quad (\text{Eq. 3})$$

$$125 \quad \textit{Proportion of C4 derived C} = 1 - \frac{\delta^{13}\text{C}(\text{Sample}) - \delta^{13}\text{C}(\text{C4})}{\delta^{13}\text{C}(\text{C3}) - \delta^{13}\text{C}(\text{C4})} \quad (\text{Eq. 4})$$

126 The SOM loss data were fitted to a single exponential decay function:

$$127 \quad R_r/R_i = e^{-kt} \quad (\text{Eq. 5})$$

128 where R_r and R_i are the initial and remaining C stock (Mg C ha^{-1}) respectively, k is the
129 decomposition rate constant (yr^{-1}), and t is the cultivation or fallow period (yr).

130

131 **Statistics**

132 All results are expressed on an oven-dried weight basis for soil samples (105°C , 24 h) and plant
133 samples (70°C , 48 h). Significant ($P < 0.05$) differences between the light and heavy fractions
134 or between land-use types in the soil C stocks, $\delta^{13}\text{C}$ values, and decomposition rate constants
135 (k) were tested using analysis of variance (ANOVA). The SOM loss data were fitted to Eq. 5
136 using the least-squares technique. The differences in the slopes of the linear regression
137 equations of SOM loss were compared between land-use types. All statistics were performed
138 using SigmaPlot 14.5 and tested at a significance level of 0.05, unless otherwise stated.

139

140 **Results**

141 **Characteristics of the litter and soil density fractions**

142 Soil C stocks were positively correlated with the forest fallow period (Fig. 2a). The annual gain
143 of $0.84 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ in soil corresponded to 16.2% of the annual litterfall C input in the forest
144 fallow (Table 1; Fig. 2a). By contrast, the soil C stocks were negatively correlated with the
145 cultivation period in both the upland rice and maize fields (Fig. 2b,c). Comparison of the slopes

146 of the linear regressions indicated that the SOC loss rate was significantly ($P < 0.05$) higher in
147 the maize field than in the upland rice field (Fig. 2bc).

148 The $\delta^{13}\text{C}$ values of *Imperata* grass and maize litter (C4 plants) differed significantly
149 from those of forest litter (C3 plants) (Table 1). This supports the precondition of our study that
150 C3 plant-derived and C4 plant-derived SOM can be traced in soils. The C/N ratios and C
151 concentrations were significantly ($P < 0.05$) lower in the heavy fractions than in the light
152 fractions (Table 2). The $\delta^{13}\text{C}$ values in the heavy fraction were significantly ($P < 0.05$) higher
153 than in the light fraction in the forest soils, but no significant differences in the $\delta^{13}\text{C}$ values
154 were found between the heavy and light fractions in the upland rice and maize fields (Table 2).

155

156 **Contribution of C3- or C4-plant-derived carbon to soil organic matter**

157 In both the upland rice and maize fields, the $\delta^{13}\text{C}$ values in the light and heavy fractions were
158 positively correlated with cultivation period (Fig. 3a,b). By contrast, the $\delta^{13}\text{C}$ values in the light
159 and heavy fractions of the forest sites were negatively correlated with the fallow period (Fig.
160 3a,b). C3-plant-derived C was dominant at all sites, but the respective proportions of C3-plant-
161 derived C decreased to 62% and 67% in the light and heavy fractions of the cropland soils. The
162 proportion of C4-plant-derived C in the light fractions increased from 10% to 38% in the maize
163 fields and from 8% to 24% in the upland rice field (Table 2). Similarly, the proportion of C4-

164 plant-derived C in the heavy fractions increased from 3% to 33% in the maize fields and from
165 11% to 32% in the upland rice field (Table 2). The C3-plant-derived C stock decreased with
166 cultivation period in the maize (Fig. 4a) and upland rice (Fig. 5a) fields. However, the
167 proportion of C4-plant-derived C in the heavy fraction decreased from 30% to 6% in the forest
168 fallow (Table 2). The total C stock in the soil increased with the fallow period in the forest
169 fallows, but the C4-plant-derived C stocks decreased (Fig. 5b).

170 Fitting the SOM loss data to an exponential decay gives the decomposition rate
171 constants or k values, except for the light fraction of the forest sites (Table 3; Figs. 4a, 5a,b).
172 The k value of the heavy fraction was significantly ($P < 0.05$) lower than that of the bulk soil
173 in the forest fallow sites (Table 3). By comparison, the k value of the heavy fraction was
174 significantly ($P < 0.05$) higher than that of light fraction in both the upland rice and maize fields
175 (Table 3). The k values of SOM decomposition in the bulk soil were in the order of maize field
176 (0.044) > upland rice field (0.031) > forest (0.020) (Table 3).

177

178 **Discussion**

179 **Effects of tillage on soil organic matter decomposability in different density fractions**

180 SOM in the light fraction generally decomposes faster than SOM associated with minerals in
181 the heavy fraction (Hassink, 1995; Tan et al., 2007). Our study also showed that the C4-plant-

182 derived SOM in the light fraction of forest fallow soils also decomposes faster than in the heavy
183 fraction (Table 3). The SOM in the heavy fraction has higher $\delta^{13}\text{C}$ values and lower C/N ratios
184 due to selective respiration of ^{12}C relative to ^{13}C (Table 2) and the higher degree of humification,
185 compared to the light fraction (Table 2; Wagai et al., 2020).

186 In the maize field, C3-plant-derived C in the heavy fraction decomposes faster than in
187 the light fraction (Table 3). This contrasts with reports that recalcitrant SOM in the heavy
188 fraction decomposes slowly (Hassink, 1995; Tan et al., 2007), but faster turnover of the heavy
189 fraction has also been reported (Crow et al., 2007). SOM turnover could vary, depending on
190 soil types and tillage practices (Gregorich et al., 1995; Sollins et al., 2009). Tillage and input
191 of accessible labile litter could stimulate microbial activity to decompose the humified SOM
192 (priming effects) in the heavy fraction for nutrient mining (Dimassi et al., 2014). Note that C3-
193 plant-derived SOM is not equal to forest-derived SOM in the maize fields, as C3 plant litter
194 inputs from weeds (e.g., *Asteroidae* spp.) could be supplied to C3-derived SOM in the maize
195 fields. This induces risk of underestimating the decomposition rate constants of the light
196 fraction.

197

198 **Effects of land-use types on soil carbon stocks**

199 We hypothesized that continuous maize cultivation results in greater loss of SOC stocks
200 compared to forest fallow or upland rice cultivation. This is supported by the higher SOC loss
201 rate in the maize field than in the upland rice field (Fig. 2b,c) as well as higher rate constants
202 of C3-plant-derived SOM decomposition in the maize fields compared to C3-plant-derived
203 SOM decomposition in the upland rice fields and C3-plant-derived SOM decomposition in the
204 forest fallows (Table 3; Figs. 4, 5). This is consistent with a net C loss in the annual C balance
205 between litter inputs and heterotrophic respiration (Fujii et al., 2009) as well as a global
206 assessment reporting lower soil C stocks in maize fields compared to other crops (West et al.,
207 2002).

208 The slower decomposition of C3-plant-derived SOM in the upland rice fields compared
209 to the maize fields (Table 3) is consistent with the lower SOM loss rates in upland rice fields
210 (Fig. 2b). As in the maize field, C3-plant-derived SOM is not equal to forest-derived SOM in
211 the upland rice fields, as upland rice-derived SOM could also be provided. Based on the lower
212 shoot/root ratios of upland rice, root-derived C inputs would be greater in the upland rice fields
213 than in the maize fields (Kondo et al., 2000). This could mitigate the loss of C3-plant-derived
214 SOM in the upland rice fields (Fig. 5a). The limited erosion due to rice straw covering the soil
215 surface could also mitigate SOM loss.

216 The main source of C4-plant-derived SOM in the forest fallow is *Imperata* grass, as
217 upland rice was traditionally cultivated before forest fallow started (Sakai, 2005). SOM of C4
218 plant origin can decompose faster than SOM of C3 plant origin (Wynn and Bird, 2007).
219 However, the decomposition of C4-plant-derived SOM in the forest fallow is much slower than
220 the decomposition of C3-plant-derived SOM in the croplands in our study (Table 3). Potential
221 reasons for the preservation of C4-plant-derived SOM in forest fallow are aggregate formation
222 under no tillage, less erosion, and soil acidity (Gregorich et al., 1995; Fujii et al., 2019). As
223 seen in the lower decomposition rate constants of the heavy fraction (Table 3), no tillage
224 practice under forest fallow vegetation favors the development of aggregates and physical
225 protection of SOM in aggregates (Tan et al., 2007; Wagai et al., 2020). In addition, the forest
226 canopy can limit erosion by rainfall and temperature rise by sunlight (Pimentel and Kounang,
227 1998). Acidic soil limits cellulose decomposition in forest litters (Hayakawa et al., 2014; Fujii
228 et al., 2019).

229

230 **Implications for sustainable soil management**

231 SOM gain or loss is directly related to the C sink/source function of soil and indirectly affects
232 sustainable maize productivity via effects on soil acidity, because maize growth is sensitive to
233 acidity (Calba et al., 2006; Minasny et al., 2017). Soil pH increased with the C concentration in

234 the heavy fraction of cropland soil (Fig. S1). SOM has two functions in pH control—pH
235 buffering due to the weak acid nature of functional groups and consumption of protons via the
236 net mineralization of organic anions (Poss et al., 1995; Fujii et al., 2012). Soil acidification
237 under continuous cropping can be mitigated by the mineralization of SOM that has accumulated
238 under the forest fallow (Fujii et al., 2009, 2021). Therefore, a gain or loss of SOM leads to a
239 respective gain or loss of soil potential to neutralize acidity. The annual gain of $0.12 \text{ Mg C ha}^{-1}$
240 yr^{-1} in cropland soils corresponded to only 2.8% of the annual maize residue C input (Table 1;
241 Figs. 4c, Fig. S2c) compared to 16.2% of the annual litterfall C input in the forest fallow soils
242 (Table 1; Fig. 2a). Judging from the finding that soil C accumulation rates in the forest fallow
243 exceed SOM loss rates in the cropland soils (Fig. 2), forest fallow has high potential to mitigate
244 soil degradation. When we compare upland rice and maize, upland rice has slower rates of SOM
245 loss despite a similar gain rate of C4-plant-derived C in soil (Figs. 2b,c, Fig. S2c). Upland rice
246 cultivation and forest fallow both involve low soil disturbance, which favors the development
247 of soil aggregates and mitigation of erosion (Pimentel and Kounang, 1998). These SOM
248 preservation mechanisms should be applied to develop a land-use strategy to mitigate soil
249 degradation, such as agroforestry involving crop and rubber or orange trees.

250

251 **Conclusions**

252 We showed quantitatively that continuous cropping of maize or upland rice leads to a loss of
253 SOC. Especially, continuous cropping causes a greater loss of SOC compared to upland rice
254 cultivation. The decomposition of C4-plant-derived SOM was slower in forest fallow than the
255 decomposition of C3-plant-derived SOM in maize or upland rice fields. Since the soil C stocks
256 increased with the forest fallow period and upland rice cultivation has a slower loss of SOC,
257 the inclusion of forest fallow and upland rice cultivation in a land-use strategy is optimal for
258 maintaining SOC stocks.

259

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264

265 **Conflict of Interest:** The authors declare that they have no conflict of interest

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345

346 **Table captions**

347 **Table 1** Site information of litter and soil

348

349 **Table 2** Soil C stocks and ^{13}C natural abundance in light and heavy fractions

350

351 **Table 3** Decomposition rate constants of C3 or C4 plant-derived organic matter in the light and
352 heavy fractions

353 **Figure captions**

354 **Fig. 1** (a) Land use change from shifting cultivation in the upstream area to continuous cropping
355 in the downstream area close to highway, (b) land use map in 1992 and 2004, and (c) sampling
356 locations on topographic map (contours with 50m interval). Data source was Sakai (2005).

357

358 **Fig. 2** Changes in soil carbon stocks (0-5 cm) of (a) forest fallow, (b) upland rice field, and (c)
359 maize field.

360

361 **Fig. 3** Changes in soil $\delta^{13}\text{C}$ values of (a) light fraction and (b) heavy fraction of the surface soil
362 (b).

363

364 **Fig. 4** Changes in stocks (0-5 cm) of (a) C3-plant-derived C (bulk soil), (b) C3-plant-derived C
365 in the light and heavy fractions, (c) C4-plant-derived C (bulk soil), and (d) C4-plant-derived C
366 in the light and heavy fractions in the maize fields. The curves represent fitting with single
367 exponential decay function, while lines represent fitting with single linear regression.

368

369 **Fig. 5** Changes in stocks (0-5 cm) of (a) C4-plant-derived C (bulk soil) in the forest fallows and
370 (b) C3-plant-derived C (bulk soil) in the upland rice fields. The curves represent fitting with
371 single exponential decay function.

372

373 **Fig. S1** Relationship between soil pH and soil C concentration in the heavy fraction ($> 1.60 \text{ g}$
374 cm^{-3}).

375

376 **Fig. S2** Changes in stocks (0-5 cm) of (a) forest-derived C (bulk soil), (b) forest-derived C in
377 the light and heavy fractions, (c) C4-plant-derived C (bulk soil), and (d) C4-plant-derived C in
378 the light and heavy fractions in the upland rice fields. The curves represent fitting with single
379 exponential decay function, while lines represent fitting with single linear regression.

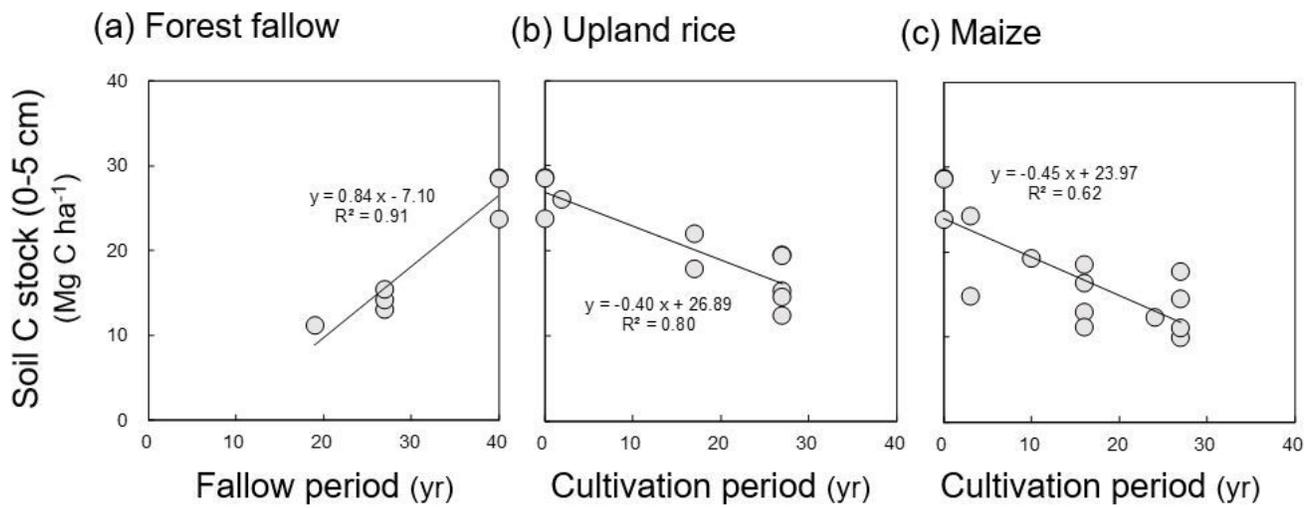


Figure 2

Changes in soil carbon stocks (0-5 cm) of (a) forest fallow, (b) upland rice field, and (c) maize field.

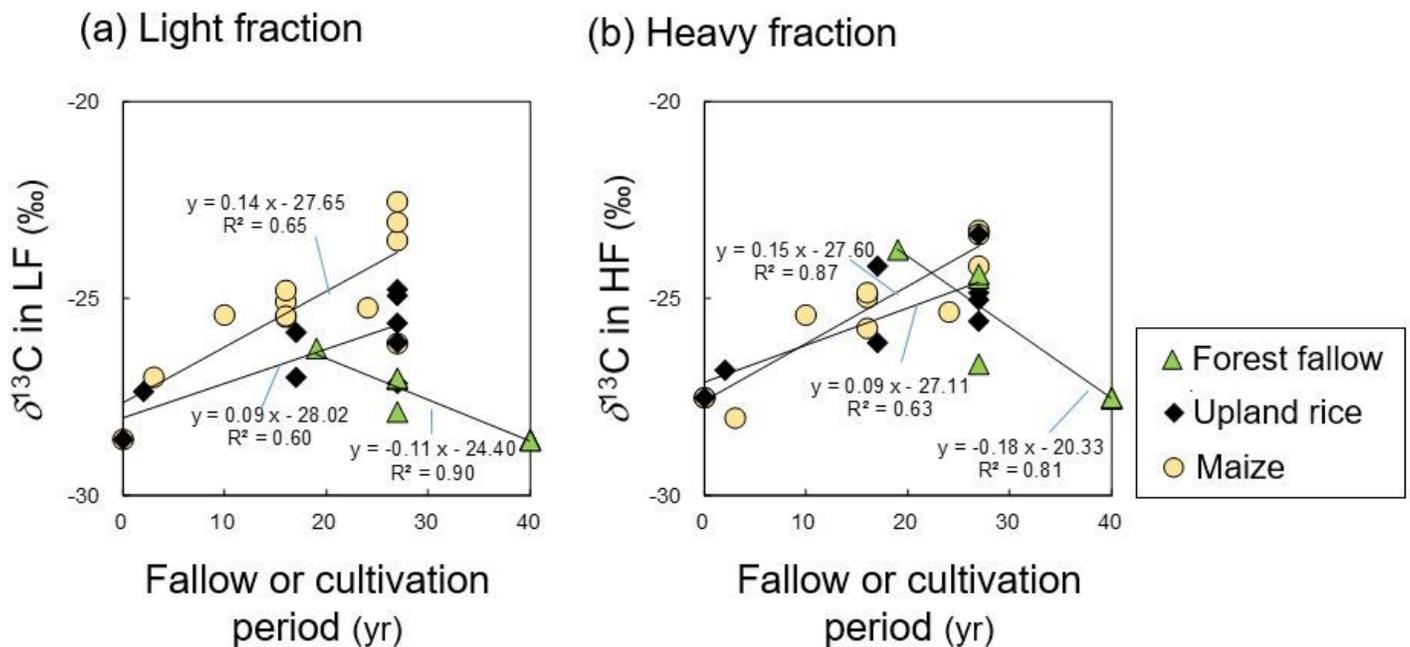


Figure 3

Changes in soil $\delta^{13}\text{C}$ values of (a) light fraction and (b) heavy fraction of the surface soil (b).

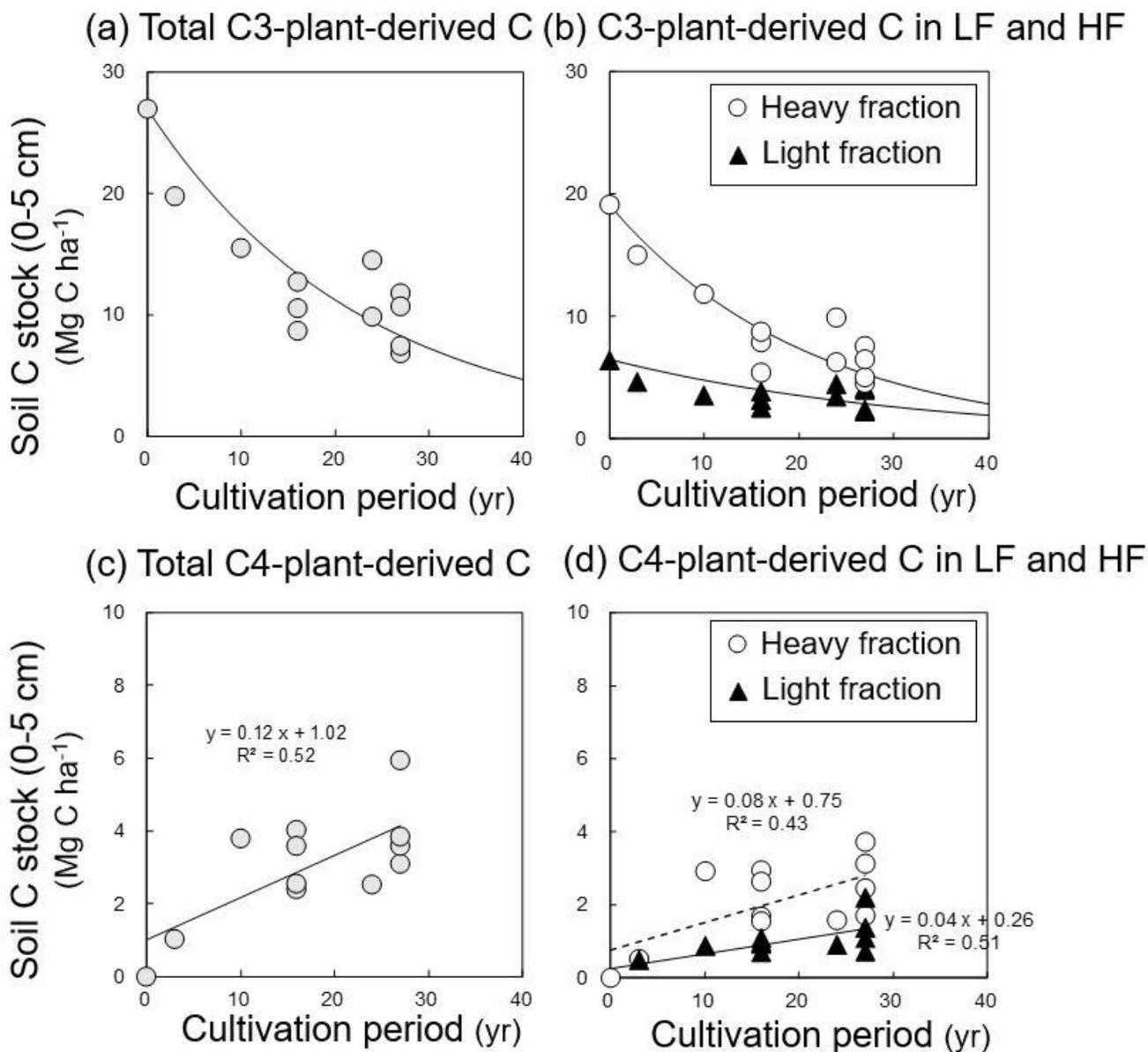


Figure 4

Changes in stocks (0-5 cm) of (a) C3-plant-derived C (bulk soil), (b) C3-plant-derived C in the light and heavy fractions, (c) C4-plant-derived C (bulk soil), and (d) C4-plant-derived C in the light and heavy fractions in the maize fields. The curves represent fitting with single exponential decay function, while lines represent fitting with single linear regression.

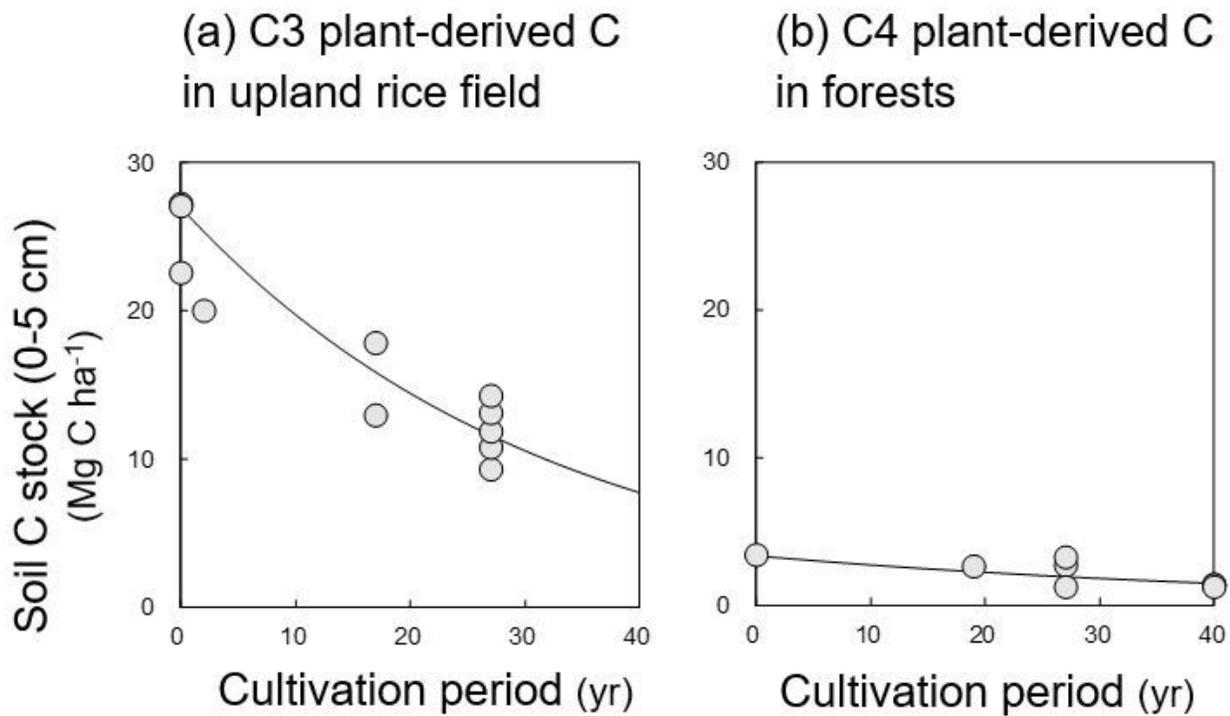


Figure 5

Changes in stocks (0-5 cm) of (a) C4-plant-derived C (bulk soil) in the forest fallows and (b) C3-plant-derived C (bulk soil) in the upland rice fields. The curves represent fitting with single exponential decay function.

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