Soil Carbon Sequestration Affected by Cropping Changes from Upland Maize to Flooded Rice Cultivation

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Abstract: The effects of cropping changes from upland maize to flooded rice cultivation on soil organic carbon (SOC) were studied. Three treatments of field experiments; continuous maize (M treatment), continuous paddy rice (R treatment) and maize-rice rotation (RM treatment) were made. Cropping change from maize to flooded rice resulted in an increase in soil bulk density and SOC content when compared to that of maize-rice rotation and continuous maize. The total SOC after two croppings was 16.50, 20.88 and 19.35 ton C ha^{-1} in the M, R and RM treatments, respectively. The effects of these short-term cropping changes were observed at both the aggregate level and in the humic substances of soil organic matter. The majority of SOC (ca. 65%) was present in association with the macro-aggregate (>250 μm), of which the fraction size of 250-500 μm contained the highest carbon concentration. After two croppings, the $\delta^{13}C$ values of SOC and humic substances were shifted towards the $\delta^{13}C$ values of rice straw when soil was incorporated with rice straw. The shift of $\delta^{13}C$ values towards maize straw's $\delta^{13}C$ values was also observed in cases when maize straw was incorporated into soil. The results demonstrate that a shift from upland maize to flooded rice could enhance soil carbon sequestration, and decomposition and incorporation of organic materials (maize and rice straw) into SOC and humic substances (himic acid, fulvic acid and humin) was detectable within a short time period.

Keywords: Soil organic carbon, cropping change, natural ^{13}C abundance, soil aggregate, humic substances.

1. Introduction

Increase in the atmospheric concentrations of carbon dioxide (CO_2), the most abundant anthropogenic greenhouse gas in the atmosphere, has resulted in global warming and climate change during the past centuries [1]. Reducing atmospheric concentrations of CO_2 has therefore become one of the prime goals of research in the recent times. In fact, CO_2 can be kept out of the atmosphere by capturing it in various reservoirs/systems on the earth surface.

Soil is the largest reservoir of terrestrial carbon (C), storing approximately 53% of the terrestrial carbon. Approximately 10% of the CO_2 in the atmosphere is cycled through the soil each year [2]. More than half of soil carbon is in organic form (soil organic carbon, SOC). Generally, plant biomass (above and below ground residues) is the primary source of carbon input into SOC. When biomass decomposes, it is incorporated into SOC, of which up to 70-80% is defined as humic substance [3]. Once it is becomes part of SOC, carbon is stored in soil for a long time since humic substances are recalcitrant. Smith et al. [4] estimated that 89% of global potential for agricultural greenhouse gas mitigation would be through carbon sequestration. Thus, large quantities of carbon from the atmosphere would be removed, and agricultural activity can contribute substantially to cutting greenhouse gas emissions. Net carbon sequestration occurs when a positive disequilibrium (the carbon input and incorporation into SOC is larger than the output) is sustained over some period of time, with the system eventually achieving a new, higher steady state [5]. About 15-40 years is the typical time required after the system is disturbed to reach the new steady state.

In agricultural systems, cultivation practices such as soil and crop management have been known to affect crop yields and soil carbon sequestration [4,6-7]. Crop rotations, soil tillage, fallow periods, and water management are all examples of cultivation practices that could either reduce or increase soil C sequestration [8].

Thailand is an agricultural country and the majority of land is under cultivation. Greenhouse gas emissions and carbon sequestration in agricultural soils therefore have unique roles in determining the net carbon balance within agriculture systems, and in off-setting emissions from other sectors. In 2000, for example, greenhouse gas emissions from the agriculture sector, estimated using the 1996 IPCC methodology, accounted for about 22% of the total emissions [9]. However, the contribution of soil as a carbon sink was not evaluated and thus not included in that inventory because there was no data available. The soil carbon inventory and its relationship to land use activity are the important issues that could be useful for estimating mitigation potentials and sustainable agriculture. This study investigated the effects of changing land use on SOC and its fractions. Knowing this would improve our understanding of the factors and mechanisms that affect the carbon inputs to and outputs from soil, and how these might be manipulated to enhance carbon sequestration. Our specific objective was to estimate SOC and to investigate its dynamics as affected by cropping changes from upland maize to lowland rice cultivation.

2. Experimental

2.1 Study site

The field experiments were carried out at the National Corn and Sorghum Research Center of Kasetsart University (Suwanwajokkasikit Field Corps Research Station), Nakorn Ratchasrima province, Thailand (14º37N, 101º19E). This area has been growing maize for approximately 20 years. The average annual rainfall is 1,000-1,200 mm with a mean temperature of 30ºC, a minimum of 14ºC and a maximum of 33ºC. Mean annual humidity is 85%. The characteristic of soil in this site is Pak Chong series (Pc), very fine, kaolinitic, isohyperthemic Rhodickandiustox (http://www.iicrd.ku.ac.th/ncsrc/ncsrc_001.htm). The experiments covered two cropping seasons between December 2008 and October 2009. Each cropping season took around four months with a two month fallow period in-between for land preparation. The experiments consisted of three treatments; (1) M treatment; continuous maize (*Zea mays L.)* where maize was grown in a similar manner to usual, (2) R treatment; continuous paddy rice (*Oryza sativa L.*) where the land previously planted with maize was changed to flooded rice, and 3) MR treatment; maize-rice rotation where in the first cropping was the same as that of R plots but for the second cropping, the land was regrown with maize. The experiment layout was a randomized complete block design (RCBD), with a total of 9 plots (3 treatments × 3 replications), and each plot size was 3 m × 3 m. For all the cropping systems, crop residues (maize or rice straw) at rate of 5 ton/ha were added to the soil 14 days before planting. Leaves, stems and roots of maize and rice from the previous crops were ploughed into the soil after harvest. Maize (4901 variety) was grown with a plant spacing of 25 cm and a row spacing of 75 cm. The chemical fertilizer 15-15-15 (N-P_2O_5-K_2O) was applied at planting at the rate of 312.50 kg/ha, and top-dressed with urea 28 days after planting at the rate of 156.25 kg/ha. Rice seedlings (Chai Nat 1 variety) were transplanted by hand with three plants/hill and a spacing of 25 cm × 25 cm. Fertilizer was applied four times; broadcast application with 16-16-8 at the rate of 187.50 kg/ha four days after transplanting (DAT), 62.5 kg/ha at 24 DAT during the tilling stage, 62.5 kg/ha with urea as a top dressing at 60 DAT, and 31.25 kg/ha with urea at 80 DAT. For the flooded rice treatment, continuous flooding was maintain with a water depth of 2-5 cm. Weed eradication was made at 20 and 50 DAT.

2.2 Soil sampling and analysis

Triplicate soil samples were taken randomly at 0-30 cm depth (plow layer) from the soil surface. For carbon content analysis, the soil samples were collected at weeks zero (before planting), eight and 16 after planting. After sampling, the soil was brought back to the laboratory and air-dried at room temperature. The soil was sieved through 2 mm mesh and then used for chemical and isotopic analysis. Soil pH was measured in water suspension with a 1:1 ratio of air-dried soil to water. The pH range found was between 5.31 and 6.04. As soil pH was in the acidic range, the majority of SOC was assumed to be in the organic form and we considered the total carbon measurement was equal to the total soil organic carbon. Analysis of the total carbon and nitrogen content was made by completely combusting C and N to CO_2 and N_2, respectively, and measured using the CHN Elemental Analyzer (Flash EA 1112 series, Italy). Soil bulk density was determined using the core method. Soil texture was determined by using the hydrometer method.

2.3 Soil aggregate separation

In order to investigate the effect of cultivation practices on SOC associated with different aggregate size fractions, air-dried soil was sieved and separated into 4 size classes; < 250 µm, 250-500 µm, 500-1000 µm, and > 1000 µm. The total C and N in each fraction was then measured as described above.

2.4 Extraction of soil humic substances (HS)

The isolation of HS was based on differences in the solubility in alkaline and acid solutions, closely following the protocol provided by the International Humic Substance Society (IHSS) [10-11]. Soil samples (~200 g) were preliminary rinsed with 0.1 M HCl in order to remove any carbonates, as well as light material and debris, such as pieces of undecomposed plant material and rootlets, from the soil [12-13]. After this pretreatment, the soil was shaken at 10,000 rpm by centrifugation (Refrigerator centrifuge, PK131R, Italy) in 0.1 mol/L NaOH plus 0.1 mol/L $Na_4P_2O_7$ with successive precipitation of the suspension to pH < 1.5. The insoluble fraction in both alkali and acid represented the humin fraction of the humic substance. The rest of the supernatant was acidified with 0.1 mol/L HCl and centrifuged at 10,000 rpm for 30 min. According to this standard protocol, the precipitated fraction was considered as the humic acid fraction, and the one that in the supernatant as the fulvic acid fraction. After separation, all HS fractions were dried at 105°C for 24 hrs and stored for analyses of total C and N as described above.

2.4 Stable carbon isotope analysis

Stable isotope analysis of $^{13}C/^{12}C$ of all sieved soil samples and HS fractions was performed by using a gas chromatograph combustion isotope ratio mass spectrometer (IRMS) at Georg-August-Universität Göttingen, Germany [14]. The ^{13}C natural abundance on each sample is expressed in per thousand (‰) units, by reference to the international standard PDB, according to the equation; $\delta^{13}C$ (‰) = $[R_{sample}/R_{standard}(PDB)\ -1] \times 1{,}000$ where R = $^{13}C/^{12}C$ of sample (R_{sample}) or standard ($R_{standard}$), respectively. All results are expressed relative to the international V-PDB standard [14].

2.5 Statistical analysis

One-way analysis of variance (ANOVA) was applied to determine the differences in parameter means among three treatments at the significance level of 95%.

3. Results and Discussion

3.1 Changes in general soil properties

Soil bulk density was slightly changed when land was brought under cultivation (Fig. 1). For M treatment, the bulk density decreased from 1.23 g/cm^3 at the week before planting to 1.10 g/cm^3 at the last week of the 1st crop. However, the value returned to about 1.23 g/cm^3 during the fallow period and before the 2nd crop. A similar trend was also observed during the 2nd crop. The change observed was probably due to the growth of maize root that resulted in a slightly increase in porosity. A contrasting trend was observed for flooded rice. During the 1st crop the bulk density hardly changed (R and RM). However, when rice cultivation was continued during the 2nd crop it increased from 1.23 g/cm^3 to around 1.35 g/cm^3. When maize was planted instead of rice (RM plot), the bulk density slightly decreased in a way similar to that was observed in M plot but to a lesser extent. The changes of bulk density subject to cropping may be explained by cultivation activities. Puddling and existence of water when rice was planted may have caused the sedimentation and increase in the density we observed. Puddling is common and a traditional practice for paddy rice cultivation. It is typically considered advantageous for achieving yield stability and high productivity, primarily by retaining water and nutrient resources while reducing weed pressure [15-17]. However, one of the impacts of soil puddling is dispersing surface aggregates and compressing the subsoil. A portion of the clay fraction from the surface horizon is deposited as clay-skins along pore surfaces at the top fringe of the compacted layer [18]. These processes reduce macropore volume in the upper portion of the soil profile while increasing bulk density in the subsoil layer.

Soil pH at the site was acidic with an average pH of 6.02, which decreased after planting. During the experimental periods, the values among all treatments varied from 5.31 to 6.04. At the end of the second crop, no significant difference among them was found. Similarly to soil pH, no significant change in soil texture was observed among all treatments. At the end of the second crop, the texture of soil in all treatments was clay with sand, silt and clay fractions of 12, 18, and 70%, respectively (data not shown).

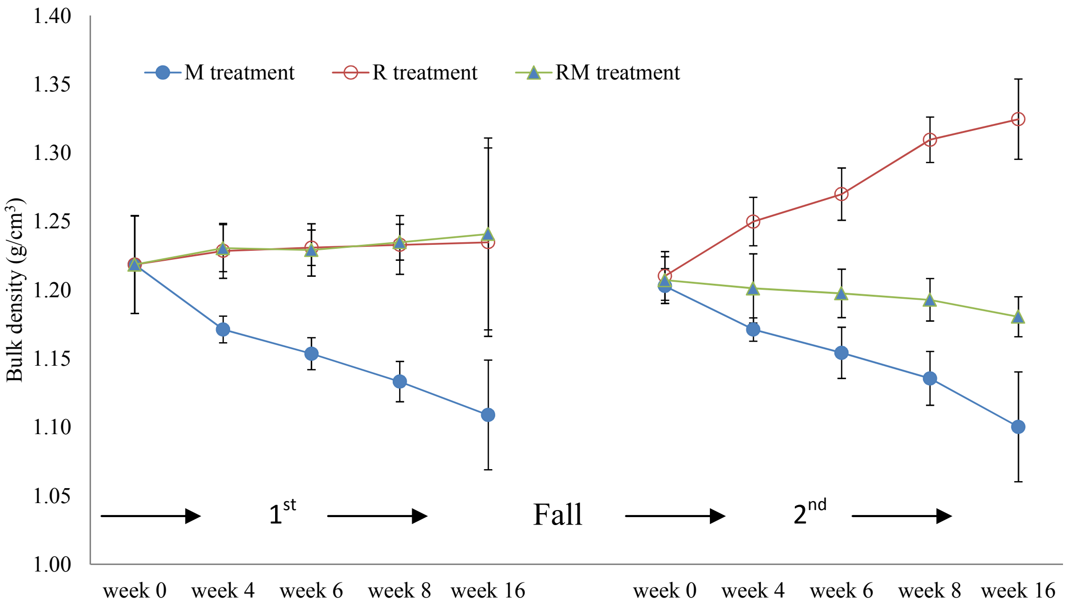

Figure 1. Changes in soil bulk density under different cropping systems. During the first crop in the R and RM treatments flooded rice was planted. During the second crop, flooded rice and maize for R and RM treatments, respectively, were cultivated. Errors bars indicate S.D. of 3 replications.

Table 1. Carbon concentration and total soil organic carbon stock in different treatments (±S.D. of 3 replications). The values for R and RM were the same during the 1st crops since they were planted with flooded rice and analysis of carbon was done on composite samples from both treatments. The values followed by the same number of asterisk or letter in the same row for carbon concentration or carbon stock are not significantly different among treatments at P <0.05, as determined by ANOVA.

	Carbon concentration			Carbon Stock		
	M treatment	R treatment	RM treatment	M treatment	R treatment	RM treatment
	(mg C/g soil)			(ton C/ha)		
1st crop						
Week 0	14.80 ± 0.32*	14.80 ± 0.32*		17.32 ± 0.34^a	18.20 ± 0.45^a	
Week 8	15.30 ± 0. 45*	14.80 ± 0.46*		17.29 ± 0.49^a	18.20 ± 0.51^a	
Week 16	16.20 ± 0.40*	16.90 ± 0.58*		17.98 ± 0.43^a	20.79 ± 0.63^b	
2nd crop						
Week 0	14.80 ± 0.52*	15.10 ± 0.46*	15.20 ± 0.44*	17.76 ± 0.56^a	18.27 ± 0.50^a	18.39 ± 0.47^a
Week 8	14.80 ± 0.50*	16.30 ± 0.58**	14.90 ± 0.42*	16.87 ± 0.53^a	21.35 ± 0.63^b	17.73 ± 0.45^c
Week 16	15.00 ± 0.45*	15.70 ± 0.47*	16.40 ± 0.40*	16.50 ± 0.48^a	20.88 ± 0.52^b	19.35 ± 0.44^c

3.2 SOC and the effects of cropping changes

Changes in agricultural practices for the purpose of increasing SOC can be made either by increasing organic matter inputs to the soil, decreasing decomposition of SOC and oxidation of SOC, or a combination of both [19]. The practices that have been known to result in increasing SOC include, for example, reducing tillage intensity in upland crops [20], decreasing or increasing the fallow period, using a winter cover crop in temperate regions, changing from monoculture to ration cropping, or altering soil inputs to increase primary production such as fertilizers, pesticides, and irrigation [21-22]. In the current study, changing from maize to flooded rice resulted in observable changes in the SOC concentrations and in the total soil carbon stocks (Table 1). The total SOC after two croppings was 16.50, 20.88 and 19.35 ton C ha^{-1} in the M, R and RM treatments, respectively. It is therefore quite obvious that changing from upland conditions under maize cultivation to lowland under flooded rice cultivation resulted in net carbon sequestration in the soil. The magnitude of C sequestration was dependent on the cropping type. When flooded rice was continued for two crops, the sequestration was 4.38 ton C/ha. However, when flooded rice was rotated with maize the sequestration was 2.85 ton C/ha. These results imply that there is a room for increasing soil carbon stock through crop managements.

One of the important changes associated with changing from upland maize to paddy rice cultivation is flooding. Under flooded conditions, the rate of decomposition of added organic materials as well as SOM are considered to be slower due to being under anaerobic conditions than under aerobic conditions found in the uplands, leading to an accumulation of SOC. In a study that surveyed storage and sequestration potential of SOC in China, Pan et al. [23] found a higher topsoil carbon content of paddy soil compared to soils in dry croplands. Several studies in China have also identified paddy soils as one of the most important SOC accumulators [24-27]. Our results further indicate that when paddy soil is converted back to upland conditions (e.g. maize), a loss of SOC occurs (Table 1). Rotating between upland maize and flooded rice may help compensate for the loss of SOC that otherwise is accelerated under upland conditions. However, the effects of such rotation on maize and rice yields need to be further studied in order to find the conditions that support both SOC sequestration and a sustainable yield. In addition, it should be noted that the results described in the current study were derived from a short-term experiment (two crops). A longer study period, sufficient to allow soil to adjust to the new equilibrium, is needed for more accurate sequestration potential estimates. Nevertheless, our results do indicate that under tropical cropping systems, diversifying cropping systems such as rotation between upland and lowland crops is one potential options for enhancing SOC.

3.3 Distribution of SOC among aggregate sites

To get insights into the changes of SOC due to cropping changes and to understand the factors that influence soil sequestration potential, the amount of SOC and its stable isotopic composition associated with different aggregate sizes was investigated. Soil aggregate is a part of particulate SOC that is separated in to macro-aggregate (>250 μm) and micro-aggregate (53-250 μm). SOC associated with micro-aggregate is known to be more physically protected and is therefore more biochemically recalcitrant. On the other hand, SOC associated with macro-aggregate is more mineralizable and therefore more dynamic [28]. Monitoring the change of SOC associated with macro-aggregate was therefore the purpose in this study, since this would be the SOC fraction sensitive to cultivation changes.

The results showed that in all treatments the majority of SOC (ca. 65%) were associated with macro-aggregates (>250 μm, Table 2). Among them, the fraction size of 250-500 μm contained the highest carbon concentration. Only about 32-34% of SOC was found in the micro-aggregate fraction. The effects of cropping change either to flooded rice or flooded rice-upland maize rotation on SOC distribution among aggregate sizes was not observed during the first crop cycle. However, after the second crop for the RM treatment with macro-aggregate size of 500-1000 and >1000 μm a significant increase in SOC (8-10%) was found when compared to M treatment. Others were either slightly increased or decreased but were not statistically significant.

Table 2. Soil organic carbon distribution in different aggregate size fractions at the end of the 2nd crop (± S.D. of 3 replications). The values in parenthesis after the 2nd crop indicate the percent changes of SOC distribution when compared to that of M treatment. The values for the R and RM were the same during the 1st crops since they were planted with flooded rice and analysis of carbon was done on composite samples from both treatments. The values followed by the same letter(s) in the same row are not significantly different among treatments at P <0.05, as determined by ANOVA.

Time	C-distribution percentage (%)		
	Continuous maize	**Continuous paddy rice**	**Maize-rice rotation**
Beginning of 1st crop			
<250 μm	33.63±0.72^a	32.54±0.74 ^{a}	
250-500 μm	35.98±0.81 ^{a}	36.76±0.82 ^{a}	
500-1000 μm	28.50±0.71 ^{a}	28.83±0.70 ^{a}	
>1000 μm	1.88±0.03 ^{a}	1.88±0.04 ^{a}	
After 2nd Crop			
<250 μm	32.17±0.69 ^{a}	31.37±0.73 ^{a} (-2.48%)	30.74±0.71 ^{a} (-9.56%)
250-500 μm	36.30±0.80 ^{a}	36.82 ^{a} ± 0.82 (1.43%)	35.65±0.81 ^{a} (-1.79%)
500-1000 μm	29.72±0.77 ^{a}	29.94±0.73 ^{a} (0.74%)	31.69±0.80^b (6.63%)
>1000 μm	1.77±0.04 ^{a}	1.87±0.03 ^{a} (5.65%)	1.92±0.04^b (8.47%)

Changes of SOC associated with aggregate were further studied by using naturally abundant stable carbon isotopes. The basis of this approach is that due to different photosynthetic pathways, the δ^{13}C values of C3 plants (e.g. rice) generally range from −33‰ to −22‰, whereas the values for C4 vegetation (e.g. maize) range from −17‰ to −9‰ [29]. These values slightly change in response to local and agronomic conditions. The analysis of rice and maize samples found that their δ^{13}C values were within the typical ranges for C4 and C3 plants; -29.48±0.42‰ and -13.03±0.54‰ (S.D.±3 replications), respectively. It is thus expected that when rice is planted and rice straw is incorporated into the soil that had been grown with maize, the δ^{13}C values of SOC will shift towards the δ^{13}C values of rice plant. The rate of change mainly depends on the amount of rice straw incorporated and the rate of rice straw decomposition and incorporation into SOC. Data shown in Table 3 indicates that after two crops of rice cultivation, the δ^{13}C values of the SOC associated with micro-, macro-aggregate and bulk soil had shifted towards the isotopic signature of rice. This means that rice plant biomass was decomposed and subsequently incorporated into all aggregate fractions. In the RM treatment when maize was planted alternatively with rice, the δ^{13}C values of SOC were also distinct from those planted with only maize. The δ^{13}C values of RM treatment were between those of M and R treatments. Thus, we were able to demonstrate that in a short time span (two croppings), the measurement of only C concentrations as shown in Table 2 may not be able to tell us much about the changes in soil carbon, but using stable isotope techniques with high measurement precision (±0.10‰) can help us gain insights into SOC dynamics as affected by cropping changes. It tells us that SOC is quite sensitive to cropping changes, and replacement of the SOC derived from previous crops with that of SOC derived from the new crop could occur within a short time period. This, however, is not necessarily detectable when only the changes of SOC mass are measured.

Table 3. The δ^{13}C values of SOC in different aggregate size fractions at the end of the 2nd crop (±S.D. of 3 replications). The values followed by the same letter(s) in the same row are not significantly different among treatments at P <0.05, as determined by ANOVA.

Aggregate	**δ^{13}C values ((‰)**		
	M treatment	R treatment	RM treatment
< 250 μm	-18.94±0.08^a	-20.72±0.48^b	-20.20±0.19^b
250-500 μm	-18.62±0.17^a	-20.79±0.57^b	-20.08±0.31^b
500-1000 μm	-18.94±0.29^a	-20.29±0.93^b	-20.10±0.44b
Bulk soil	-18.70±0.09^a	-20.98±0.38^b	-20.31±0.16^c

3.4 Effects of cropping change on humic substances

Humic substances are important components of soil organic matter (SOM) which determine the physical, chemical and biological properties of soil. They also represent the principal surface reservoir of carbon and plant nutrients [30]. The contents and quality of humic substances and SOM have been well studied and are affected by, for example, land use, cultivation practices, climate conditions and the amount and quality of residue incorporated into the soil [30].

For short-term changes in the cropping system, as in the current study, significant changes in humic substance concentrations were not observed. However, during the second crop, humic acid and fulvic acid fractions were slightly increased in the R and RM treatments when compared to those of the M treatment (Fig. 2). It was also found that the majority of humic substances are present in the form of humic and fulvic acids (50-60 mg C/g soil). For humin, its content was less than 15 mg C/g soil in all treatments and no change was found during both the first and second crop.

Possibly due to the short-time span, the observed changes in the mass of humic substances was negligible or within the error range associated with sampling and analysis. The use of stable carbon isotope, as mentioned earlier, can be very helpful in studying the dynamics of SOC/SOM change over a short time period [31]. We found that cropping changes from maize to rice significantly altered the δ^{13}C values of humic fractions (Table 4). As expected, the most negative δ^{13}C values of humic fraction were observed under R treatment (<-20‰), and most positive under M treatment (>-19‰). The δ^{13}C values of RM were in between those of the R and M treatment. Among the three fractions of humic substances, it seems that humic acid fraction was most sensitive to change from maize to flooded rice, as the scale of change of its δ^{13}C values was highest, and among them it showed the most negative value. These results indicate that SOM turnover was quite quick, as there was an incorporation of newly added organic material into the humic substances within two cropping periods. Measurement of changes in mass only was not able capture such changes (e.g. Fig. 2).

Table 4. The $\delta^{13}C$ values of SOC in humic substance fractions and bulk soil (± of SD of 3 replicates). The values followed by the same letter(s) in the same row are not significantly different among treatments at P <0.05, as determined by ANOVA.

HS fraction	$\delta^{13}C$ values **(‰)**		
	M treatment	R treatment	RM treatment
Before cultivation			
Humic acid fraction	-19.62 ± 0.07^{a}	-19.60 ± 0.07^{a}	
Fulvic acid fraction	-19.70 ± 0.07^{a}	-19.70 ± 0.08^{a}	
Humin fraction	-19.20 ± 0.06^{a}	-19.19 ± 0.09^{a}	
End of 2nd crop			
Humic acid fraction	-19.24 ± 0.15^{a}	-21.35 ± 0.12^{b}	-20.43 ± 0.17^{c}
Fulvic acid fraction	-17.90 ± 0.30^{a}	-20.83 ± 0.28^{b}	-19.09 ± 0.27^{c}
Humin fraction	-18.73 ± 0.16^{a}	-20.68 ± 0.15^{b}	-19.64 ± 0.18^{c}

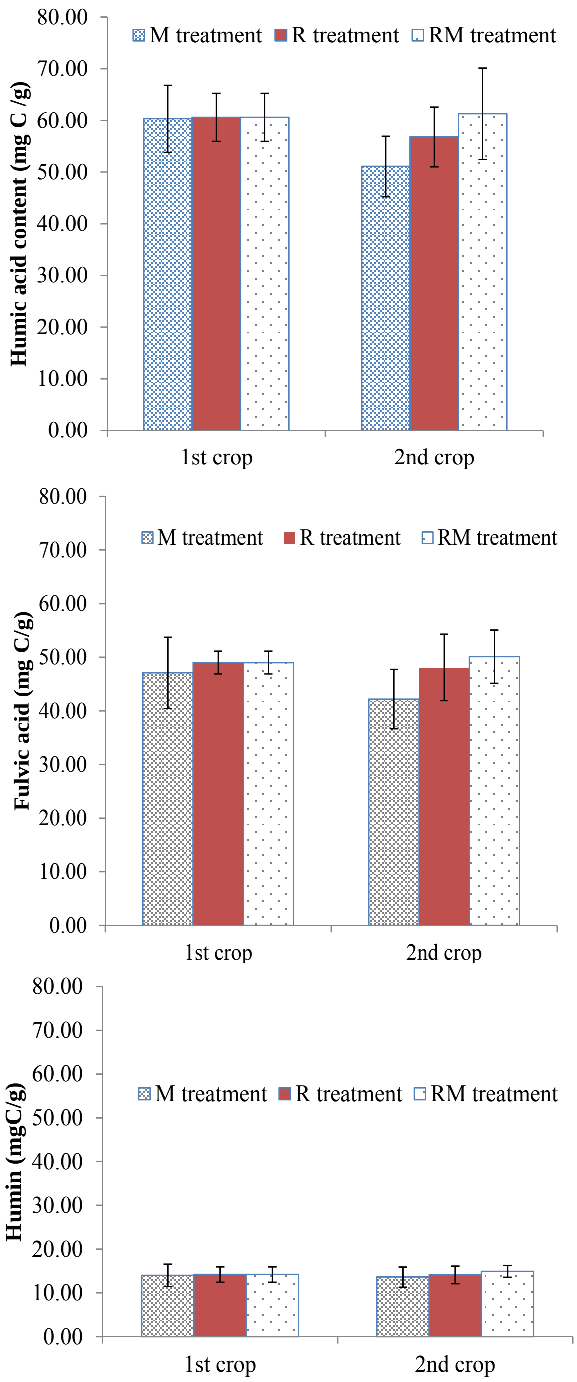

Figure 2. Changes in SOC associated with (a) Humic acid fraction (b) Fulvic acid fractions and (c) Humin fraction. Error bars indicate SD of 3 replications.

4. Conclusions

Our results demonstrate that the dynamics of SOM and SOC can be at least semi-quantitatively measured by a combination of measuring carbon mass changes and its stable carbon isotope composition. This is possible when there is a shift in cropping from C3 to C4 plants or *vice versa*. In the current study, cropping changes from upland maize to flooded rice cultivation could enhance soil carbon sequestration. Crop rotation between maize and flooded rice also resulted in enhancing soil carbon sequestration, but to a lesser extend when compared to that of continuous planting of flooded rice. The inputs of new organic material through decomposition and humification can be measured in aggregate and humic substance fractions. Our results indicate that the decomposition of organic materials (maize and rice residues) is very rapid and the incorporaion of the decomposition products into humic substances is obvious within one or two croppings.

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