

STABLE CARBON ISOTOPE FOR EVALUATING SOIL ORGANIC CARBON BETWEEN RICE-CORN ROTATION AND CORN MONOCROPPING

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Received: February 09, 2024; Revised: May 20, 2024; Accepted: June 04, 2024

Abstract

Soil organic carbon (SOC) is a significant component that plays an important role in soil fertility and crop sustainable production. This study investigated SOC in soil profiles of rice-corn rotation and corn monocropping using a stable carbon isotope of SOC ($\delta^{13}\text{C}_{\text{SOC}}$) technique. The soil samples were collected from two plots of rice-corn rotation and corn monocropping fields. Soil sampling was conducted in Tak province, involving the collection of 25 soil cores per plot down to a depth of 50 cm. Each soil profile was further segmented into ten depth layers: 0-5 cm, 5-10 cm, 10-15 cm, 15-20 cm, 20-25 cm, 25-30 cm, 30-35 cm, 35-40 cm, 40-45 cm, and 45-50 cm. The collected soil samples were then subjected to air drying, pulverization, and sieving through a 200-mesh sieve. The $\delta^{13}\text{C}_{\text{SOC}}$ in rice-corn rotation ranged from -28.41‰ to -23.72‰ while corn monocropping was -24.59‰ to -15.83‰. The results revealed that the values of $\delta^{13}\text{C}_{\text{SOC}}$ for each soil profile gradually increased with soil depth. Since $\delta^{13}\text{C}_{\text{SOC}}$ levels were negatively correlated to SOC. Therefore, SOC decreased with increasing soil depth in both the rice-corn and corn fields. However, significantly higher SOC contents were observed in rice-corn rotation rather than in corn monocropping fields ($p < 0.05$). SOC contents were higher in soil with rice straw retention than without rice residues. The rotation of rice-corn practice could potentially retain organic carbon in soil better than corn monocropping.

Keywords: Stable carbon isotope technique; Soil organic carbon; Rice-Corn rotation; Corn monocropping

Introduction

Soil fertility in agricultural ecosystem is a main component on enhancing crop quality and yield. Soil organic carbon (SOC) is widely recognized as an indicator of soil fertility, it plays a crucial role on soil structure, nutrient supply, water-holding capacity, soil microbial activity, and soil characteristics. SOC results mainly from plant residues, animal remains and soil microbial activity.

The low cost and sustainable practices to increase SOC in cultural system are crop residue retention, farm yard manure application, and tillage minimization (Turgut *et al.*, 2005; Blanco-Canqui and Lal, 2009; Fuentes *et al.*, 2010; Dou *et al.*, 2014).

The stable carbon isotope ($^{13}\text{C}/^{12}\text{C}$, or $\delta^{13}\text{C}$) is widely used to study the dynamics of SOC, the

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DOI: <https://doi.org/10.55766/sujst-2024-04-e03552>

Suranaree J. Sci. Technol. 31(4):030208(1-6)

change in carbon source due to crop change, the carbon turnover from C3 to C4 plant in soil, the fate of carbon in crop-soil ecosystems and the dynamics of residue decomposition in soil. The carbon isotopic technique is applied of studies in soil science, agriculture, ecology, and biology due to its naturally occurring isotopes (Dou *et al.*, 2014).

Gregorich *et al.* (2001) reported that the $\delta^{13}\text{C}$ values in soil suggested that crop residue input enhanced the SOC content more than inorganic fertilizer application in the same amount of nutrients. Many studies noted that SOC was related directly to the organs of plant residue retention. Müller *et al.* (2016) found that incorporation of maize carbon derived from shoots and roots increased SOC considerably. Using stable carbon isotope technique, studies revealed that the root derived carbon contributed more than that from above ground biomass to soil carbon sequestration (Kong and Six, 2010).

Khalid *et al.* (2014) observed the incorporation of poultry manure increased SOC which in turn boosted soil hydrologic and hydraulic properties of the sandy soil. Although, research indicated that farm yard manure was helpful in improving SOC for increasing soil fertility and crop yield. The spread of animal manure as a soil amendment caused negative environmental impacts such as increased soil salinity, soil pH, bacteria, water sources pollution, and greenhouse gas emission. Bacteria found in manures such as *E. coli* and *Salmonella* have often been transferred to fresh crop which implicated to human health (Black *et al.*, 2021).

Negative impacts on SOC accumulations were observed with the higher rate of crop residue removal and greater application rates of nitrogen fertilizer (Smith *et al.*, 2012). For example, Blanco-Canqui and Lal (2007) found that SOC was reduced after removal of corn residue in the 2 years of corn cropping with no till area and the SOC contents varied according to soil type and removal rate. Moebius-Clune *et al.* (2008) reported that physical, chemical, and biological of soil quality effected by removing corn residue in a zero-till system. Also, SOC storage can be influenced by different crop species. Kuzyakov and Domanski (2000) revealed that plant species of cereal (wheat and barley) and pasture (ryegrass, bunchgrass, and red brome) varied in the below-ground carbon translocation. Cereals transported about 20% - 30% (ca. 1500 kg C/ha) and pasture plants about 30 - 50% (ca. 2200 kg C/ha) of total assimilated carbon production into soil. Similar results of SOC stocks in soil profiles under different crop species were also reported by West and Post (2002). The positive impacts of crop residue retention on the ability of soil to store carbon are related to the amount and quality of crop residue

input, tillage intensity, fertilization rate, the variable climate conditions, and soil characteristics (Blanco-Canqui and Lal, 2009; Fu *et al.*, 2021).

Stable isotope analysis has appeared as one of the most powerful tools for studying organic carbon in soil. The isotope analysis technique has become widespread due to the relative ease and low cost of sample preparation and analysis including lacking of chemical uses (Dou *et al.*, 2014). However, the variable crop residues to influence carbon isotopic ratios ($\delta^{13}\text{C}$) or in other word SOC, has not widely investigated in Thailand yet.

The objective of the present study was to evaluate the effect of different plant residues on total SOC in rice corn rotation and corn monocropping. The $\delta^{13}\text{C}$ contents of SOC ($\delta^{13}\text{C}_{\text{SOC}}$) and bulk density in soil profiles collected from two cultivated areas in 2018 and 2019 were determined.

Materials and Methods

Study Area

The study was conducted at Mae Ramah (16° 59' N, 98° 31' E and 195 masl) situated in the tropical highland of Tak province. (Figure 1).



Figure 1. Location of the study area

Total area of the Mae Ramah district is 1,476 km². Agriculture is a major part of the Tak economy. Tak province produces rice, corn, vegetables, fruit, beef, tilapia, and other foods. Industries in Tak include granite quarrying and jewelry.

In Mae Ramah district, the wet season is oppressive and overcast, the dry season is humid and partly cloudy, and it is hot year-round. The annual temperature typically varies from 16°C to 36°C and is seldom below 12°C or above 38°C. Mae Ramah has an extreme seasonal variation in monthly rainfall with average annual rainfall of 1540 mm, falling between May and October (Wikipedia 2020 Mae Ramat District).

Sample Collection and Preparation

Soil samplings were carried out from areas of rice-corn rotation and corn monocropping each of 3.2 hectares after harvest and thus before rice and corn planting in 2018 and 2019. The rotation and monocropping systems in this research were grown continuously. The samples were taken by a randomized block design replicated three times. Soil corer (Eijkelkamp, Netherland, dia. 8 cm) was used to collect soil at depth level of 0-50 cm. and were sliced 5 cm interval (0-5, 5-10, 10-15, 15-20, 20-25, 25-30, 30-35, 35-40, 40-45, and 45-50 cm). Roots, plants, detritus, litter, and stones were removed by hand from the samples before grinding. Samples were air dried and finely ground using a mixer mill (Model MM200, Retsch, Haan, Germany). The ground soil samples were sieved through a 125micron screen and stored in sealed plastic bag for further $\delta^{13}\text{C}$ analysis.

Carbon Isotope Analysis

Soil sample of 2 mg was encapsulated in tin capsules for stable carbon isotope ratio ($^{13}\text{C}/^{12}\text{C}$; $\delta^{13}\text{C}$) analysis. The $\delta^{13}\text{C}$ were determined at the Thailand Institute of Nuclear Technology (Public Organization) using Cavity ring-down spectrometry (Picarro Isotopic carbon analyzer G2121-i). The ratio of $^{13}\text{C}/^{12}\text{C}$ is denoted as $\delta^{13}\text{C}$ and expressed as parts per thousand or per mill (‰), relative to the international Pee Dee Belemnite standard. The $\delta^{13}\text{C}$ value was calculated from the measured carbon isotope ratios of the sample and standard gases as:

$$\delta^{13}\text{C} = [({}^{13}\text{C}/{}^{12}\text{C})]_{\text{sample}}/({}^{13}\text{C}/{}^{12}\text{C})_{\text{PDB}} - 1]1000$$

Periodic replicates were run and reference materials/standards were measured alongside samples in every analysis.

A positive δ value means that the ratio of the heavy to light isotope of the sample is higher than that of the standard while sample with a negative δ value contains isotopically lighter than the standard. The δ value always prefaces with a sign, either positive or negative.

Soil bulk density (g/cm^3) was determined using core method and calculated as the following equation (Arunrat *et al.*, 2021).

$$\text{Bulk density} = \frac{\text{Mass of dry soil}}{\text{Vol of the soil sample}}$$

Statistical Analysis

Statistical analysis was performed using GraphPad Prism v.8 (GraphPad Software, Inc., La Jolla, California, USA). One-way ANOVA was conducted to compare differences between two sampling locations at $p < 0.05$.

Results and Discussion

In the soil profiles of rice-corn rotation and corn monocropping, the stable carbon average values of SOC ($\delta^{13}\text{C}_{\text{SOC}}$) ranged from -28.41 and -24.64 ‰ in the upper horizons or top soil (0-20 cm) and increase to -22.62 and -15.83‰. in the lower horizons or sub soil (20-50 cm), respectively. (Figure 2). There is a clear inverse correlation between the $\delta^{13}\text{C}_{\text{SOC}}$ values and the amount of SOC sequestration (Dou *et al.*, 2014; Ghafoor *et al.*, 2017). Therefore, SOC stock in rice-corn rotation was suggested to be higher than in corn monocropping and was significantly difference ($p < 0.05$).

The results revealed that $\delta^{13}\text{C}_{\text{SOC}}$ of rice-corn rotation soils has a slightly depleted isotopic composition of carbon in comparison with the $\delta^{13}\text{C}_{\text{SOC}}$ in the corn monocropping, which are characterized by types of plant residues retention. Therefore, it is reasonable to assume that the $\delta^{13}\text{C}_{\text{SOC}}$ values of rice-corn rotation and corn monocropping under the identical conditions of climate, cultivation practices, and soil types in the study area depend on the quality of the retaining crop residues during the post-harvest period. Previous studies have indicated that carbon content varied markedly among plant types due to their compositions of lignocellulosic compounds in biomass. The contents of organic compounds i.e., lignin, cellulose, hemicellulose proteins, amino acids, carbohydrates, waxes, humic and fulvic acids in various plant species effected SOC stock significantly (Martin *et al.*, 2015; Wan *et al.*, 2019; Jiao *et al.*, 2020; Miao *et al.*, 2022). Table 1 shows the contents of lignin, cellulose, and hemicellulose in straw, sugar cane (shoot and leave), bagasse, rice husk, and corn (stem, shoot and leaves) and expressed these as % content based on dry weight.

Table 1. Composition of lignocellulosic biomass in various plant species (Sawangpol *et al.*, 2013)

Biomass	Cellulose*	Hemi cellulose*	Lignin*
Rice straw	8,962.02	6,109.93	4,343.78
Sugar cane	2,964.24	2,067.56	1,693.23
Rice husk	897.12	718.98	625.90
Bagasse	894.43	525.44	468.25
Corn (stalk and stover)	258.37	146.44	122.82

* Multiply to 10^3 dry mass ($\times 10^3$ DM)

Cellulose in lignocellulosic biomass is usually have broken down into biologically usable forms. Cellulose is the most abundant compound in plant biomass of terrestrial origin, it denoted

approximately 30% of the carbon sequestered in plant materials annually. Lignin turnover is distinctly different from that of cellulose and hemicellulose, however; it plays a crucial role in carbon cycles of both terrestrial and oceanic (Austin 2010). The results of the present study were consistent with previous findings that cellulose is the dominant form of carbon input into arable soils and with the input of different types of crop residues, the differences in the composition of lignocellulosic compounds in plant residues were ultimately converted into differences in the contents of SOC. (Austin 2010; Martin *et al.*, 2015; Wan *et al.*, 2019; Jiao *et al.*, 2020; Miao *et al.*, 2022)

Apart from above ground biomass, it was documented that organic matter inputs from roots tend to contribute to SOC significantly more than aboveground plant inputs (Rasse *et al.*, 2005; Zhang *et al.*, 2015; Jackson *et al.*, 2017; Ghafoor *et al.*, 2017). The explanation is that SOC sequestration results from rhizodeposition of living roots and dead root decomposition. Root exudates of living plant are organic carbon compounds such as simple sugars, organic acids, and amino acids which release into the soil and effect the increase in SOC sequestration (Yang *et al.*, 2023). Ghafoor *et al.* (2017) and Zhang *et al.* (2015) revealed that root-derived carbon was mainly retained in soil and increased 2.3-fold more to the soil carbon sequestration than aboveground residues. Studied of Yang *et al.*, 2023 using $\delta^{13}\text{C}$ signatures revealed that signatures of $\delta^{13}\text{C}_{\text{SOC}}$ were reliable with roots $\delta^{13}\text{C}$, indicating the roots were the key source of SOC stock. It was observed that SOC mainly achieved from fine roots (Yang *et al.*, 2023). Research on four Thai upland rice cultivars grown in clay loam shown that the root diameter ranged from 1471.83 to 961.16 microns (Dechkrong *et al.*, 2022) while the highest diameter of maize root was 16.61 cm (Pakoktom *et al.*, 2021). Breidenbach *et al.* (2017) revealed the variation of microbial communities between rice roots and maize roots, The abundance of these soil microbial were found on rice root more than on maize roots. The plenty of microbial community resulted in the enhancement of soil carbon formation. The increment of soil microbial and the fluctuations in community structure were positively correlated with SOC sequestration. Previous study documented that the increased input of plant residue has driving microbial community shifts and soil carbon accumulation (Li *et al.*, 2021).

In addition, it is observed from figure 2 that distribution of $\delta^{13}\text{C}_{\text{SOC}}$ for these soils of the studied areas demonstrates similar patterns. The $\delta^{13}\text{C}_{\text{SOC}}$ contents in both plots increased below the humus horizons (top soil 0-20 cm) and gradually increased further down along the profiles (sub soil 20-50 cm).

The relative maximums of $\delta^{13}\text{C}_{\text{SOC}}$ were seen in the humus horizons of buried newly plant residues in soil. Formation of SOC in top soil (0-20 cm) occurred when the newly input of crop residues were amendment and prompted decomposition which accelerated by irrigation (water input). In the present study, the soil preparation stage started 2-3 weeks after harvest, a disc harrow was used to incorporating crop residues left in the field into the top soil at 20 cm depth. The soil organic carbon at upper 0-20 cm depth was significantly influenced by crop retention. Whilst the soil of lower depth, 20-50 cm, SOC contents were relatively lower and similar pattern between the rice-corn rotation and corn monocropping. The finding results were consistent with previous studies that the vertical distribution of SOC decreased with soil depth, with an opposite trend of $\delta^{13}\text{C}_{\text{SOC}}$ (Kuzayakov and Domanski 2000; Dou *et al.*, 2014; Zhang *et al.*, 2015; Wang 2020; Hu *et al.*, 2024).

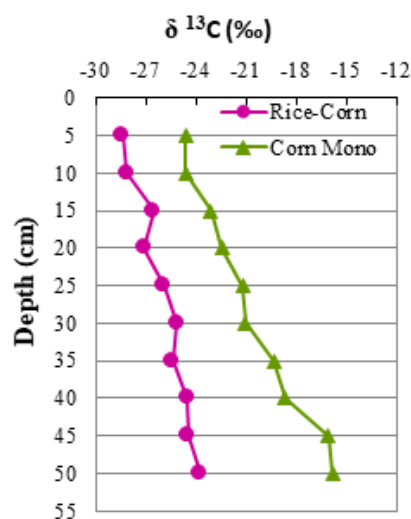


Figure 2. $\delta^{13}\text{C}_{\text{SOC}}$ depth profiles for rice-corn rotation and corn monocropping

Results shown that corn monocropping has higher bulk density than rice-corn rotation in both 2018 and 2019 crop year. Bulk density ranged from 1.62 - 1.74 for corn monocropping and 1.28 to 1.46 for rice-corn rotation which were significantly difference in both soil types ($p < 0.05$) and at all depth intervals. It revealed that the higher SOC in crop rotation field caused the lower bulk density. The lower bulk density is an indicative of more soil porosity, less soil compaction risks, improvement of water infiltration and promoting root penetrability to access water and nutrient at depth. Therefore, rice-corn rotation practice could enhance agricultural

productivity and sustainability as well as reduce the negative environmental impacts such as crop wastes.

Conclusions

The frequent removal of aboveground biomass reduced the development and accumulation of SOC. Crop residues (above ground biomass) and living roots exudates (below ground biomass) provided significantly soil carbon inputs which reflect by increasing net plant productivity. This study indicated that plant carbon contents, amounts of residues, and crop types were crucial for estimating the potential magnitude of carbon sequestration in different crop biomass.

The important factors of SOC storage and its spatial variation across land uses are the retention of plant residues of different plant types and quantity of residues input including soil microbial community composition within the soil profile especially in deeper soils (0.6-2.0 m) (Tao *et al.*, 2023). This study could conclude that the quality and quantity of rice straw retention to soil could promote the sequestration of SOC in the higher contents than that of corn biomass. The rotation of rice-corn practice could potentially retain more organic carbon in soil compared to corn monocropping. In addition, the higher SOC contents were increased soil porosity, water infiltration, root penetration and decreased soil compaction risks.

Acknowledgement

Funding for this research was provided by the National Research Council of Thailand. Special appreciation is given to Mr. Saengthit Pingwang and Mr. Prasit Khamal, the landowners for their hospitality and dedicated help with providing information and allowing access to their paddy and corn field for the whole research period.

The authors thank to the anonymous reviewers of the critical review and the editorial staffs of the Suranaree J. Sci. Technol. for assistance in the revision and production of this manuscript.

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