



Impact of vegetation restoration on soil organic carbon stocks and aggregates in a karst rocky desertification area in Southwest China

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Abstract

Purpose The objectives of the study were as follows: (a) to determine the response of soil organic carbon (SOC) fractions to vegetation restoration; (b) to examine the contributions of aggregate-associated OC to total soil OC accumulation along vegetation restoration, (c) to identify the factors that affect SOC accumulation along natural vegetation restoration in a karst region in Southwest China.

Materials and methods Four vegetation restoration stages, namely, grassland, shrubland, shrub-arbor mixed forestland, and arbor forestland, were compared with cropland (CL). Soil samples were collected at depths of 0–10 cm and separated into five aggregate size fractions. SOC, light fraction OC (LFOC), easily oxidizable OC (EOC), and aggregate-associated OCs were determined for different aggregate sizes and total soil.

Results and discussion Natural vegetation restoration increased macroaggregate amount but decreased the fractions of meso- and microaggregates. Vegetation restoration significantly increased total SOC, EOC, and LFOC concentrations and stocks and soil aggregate-associated OC concentrations. The responses of EOC and LFOC in total soil and soil aggregates were more sensitive than those of SOC along with vegetation restoration. Aggregate-associated OC concentrations generally increased with a decrease in aggregate size. Macro- and microaggregate-associated OC stocks increased, but mesoaggregate-associated OC stocks decreased following the conversion of CL to a natural vegetation ecosystem. The increase in SOC stocks was primarily attributed to the macroaggregate-associated OC stocks and their changes. OC concentrations and stocks in total soil and the soil aggregates were significantly positively related to exchangeable calcium content.

Conclusions Vegetation restoration considerably affects the amount of soil aggregates, OC concentrations, and stocks in total soil and soil aggregates. Changes in OC concentrations and stocks can be more pronounced in the liable carbon fraction than in total SOC. The increase in SOC was mostly attributed to OC accumulation in macroaggregates. Exchangeable calcium also affected soil OC accumulation in total soil and soil aggregates.

Keywords Karst area · Organic carbon accumulation · Soil aggregates · Soil organic carbon stocks · Vegetation restoration

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1 Introduction

The soil organic carbon (SOC) pool is the largest reservoir pool in terrestrial ecosystems, with a global estimate of 1500 Pg to 1 m, thereby playing an important role in atmospheric CO₂ level balance (Lal 2004). Land use change has considerably affected the SOC pool (Liu et al. 2014; Qiu et al. 2015; Deng et al. 2016; Rolando et al. 2017). SOC stocks are determined by the balance of SOC input and output (Deng et al. 2016; Xiao et al. 2017). Land use change from natural ecosystems to farmland results in SOC loss (Lal 2004). For example, the conversion of forest to cropland can deplete SOC

stocks at a depth of 0–20 cm by 34% within 100 years in Central Shaanxi Province, China (Wei et al. 2013). By contrast, the reverse process, i.e., cropland conversion to natural vegetation, accumulates SOC (Wei et al. 2012; Yang et al. 2016; Hu et al. 2018a). Therefore, the latter decreases SOC loss and is recognized as a practical approach to SOC stock conservation (Li et al. 2012). However, considerable uncertainties still exist in terms of SOC accumulation along vegetation restoration. Studies have shown that the conversion of cropland to forest or grassland can either increase, decrease, or cause negligible changes (Deng et al. 2014; Li et al. 2012). These inconsistent results from individual studies indicate that soil carbon dynamics is a complex process and is affected by multiple factors along vegetation restoration, such as land use, climate, planted tree species, restoration years (Lal 2004; Laganière et al. 2010; Li et al. 2012; Deng et al. 2014), and soil variables (Yang et al. 2016). Soil accumulation changes along vegetation restoration also depend on the site or region (Deng et al. 2014; Hu et al. 2018a). In particular, the effects of soil variables, such as soil aggregates and nutrients, e.g., nitrogen and calcium, on SOC accumulation have not been well understood in karst areas.

Soil aggregates are structural units within soil those control SOC dynamics and nutrient cycling (Six et al. 2004). Land use change generally leads to soil aggregate destruction or formation (Islam and Weil 2000; Wei et al. 2012; Wei et al. 2013) and then affects SOC storage and stability. This process can increase or decrease OC mineralization because OC in aggregates can or cannot be physically protected from microbial decomposition and oxidation (Islam and Weil 2000; Wei et al. 2012; Wei et al. 2013; Six and Paustian 2014). However, the contributions of aggregate-associated OC to SOC accumulation and its influencing factors are not well understood. Inconsistent conclusions have been obtained from individual studies, because multiple factors, e.g., land use, time since vegetation restoration, and soil depth (Blanco-Canqui and Lal 2004; Six et al. 2004), affect the contributions of aggregate-associated OC to SOC accumulation. A literature survey indicated that SOC accumulation may be primarily determined by > 0.25-mm aggregates (Wei et al. 2012; Devine et al. 2014; Liu et al. 2014; Qiu et al. 2015), 0.25–2 mm aggregates (Deng et al. 2018; Zhong et al. 2019) or < 0.25-mm aggregate (McCarthy et al. 2008). SOC sequestration via soil aggregates is an important aspect of land use change/conversion that influences carbon cycling (Zhong et al. 2019). How OC accumulates and stabilizes along vegetation restoration in a karst area and whether results are similar for soil in a karst area remain unknown. The two major mechanisms are physical protection in aggregates and interaction of SOC with mineral surfaces or polyvalent cations (Kaiser et al. 2011; von Lütow et al. 2006). The karst system in Southwest China is dominated by carbonate rocks and rich in calcium. However, whether calcium will recover and play an important

role in determining SOC accumulation along vegetation restoration remains unclear.

Labile carbon fractions, such as light fraction OC (LFOC) and easily oxidizable OC (EOC), which are small fractions of SOC, are more sensitive to land use change or management practices than total SOC (Liu et al. 2014; Chen et al. 2017). LFOC is highly labile and comprises of soil organic matter for plants and microbes (Boone 1994; Six et al. 2002) as a major source of soil nutrient (Janzen et al. 1992). Total SOC can be rapidly lost during the conversion of forest to cropland, primarily due to the significant decrease in LFOC (Wei et al. 2014). EOC is determined via chemical oxidation methods using KMnO_4 (Blair et al. 1995) and is an important indicator of soil quality (Li et al. 2018). However, the dynamics of labile carbon fraction in aggregates flowing vegetation restoration is rarely studied.

The karst region in Southwest China is characterized by limestone and dolomite and is famous for its severe rocky desertification (Liu et al. 2019b). This karst system is a highly vulnerable and sensitive ecosystem (Jiang et al. 2014; Hu et al. 2018a). In the past few decades, this fragile ecosystem has been considerably degraded by unreasonable land utilization and management, resulting in the destruction of natural vegetation, such as deforestation and cultivation and causing SOC loss and soil erosion (Hu et al. 2018a; Hu et al. 2018b; Liu et al. 2019b). Vegetation restoration is an effective strategy for SOC stocks in a karst region. Changes in soil OC stock from cropland to grassland, shrubland and forest in the karst region of Southwest China have been reported (Liu et al. 2015; Yang et al. 2016; Hu et al. 2018a). For example, Liu et al. (2015) found that SOC accumulation increased from $29.10 \text{ g}\cdot\text{kg}^{-1}$ in grassland to $73.92 \text{ g}\cdot\text{kg}^{-1}$ in primary forest. However, studies on the effects of vegetation restoration on soil OC fractions and soil aggregate-associated OC and the contributions of aggregate-associated OC to total soil OC accumulation and its influencing factors are limited. In our study, we investigated the effects of vegetation restoration on soil OC and soil OC fractions, such as LFOC, EOC, and aggregate-associated OC, in a karst region in Southwest China. The objectives of our study were as follows: (a) to determine the response of SOC, LFOC, EOC, and aggregate-associated OC; (b) to examine the contributions of aggregate-associated OC to total soil OC accumulation along vegetation restoration; and (c) to identify the factors that affect SOC accumulation.

2 Materials and methods

2.1 Description of study sites

The study sites were located in the Zhongliang Mountains in Beibei district, Chongqing, in Southwest China ($29^{\circ}39'–30^{\circ}3' \text{ N}$, $106^{\circ}18'–106^{\circ}56' \text{ E}$ (Fig. 1). The area has a typical

subtropical monsoon climate, with a mean temperature of 18 °C and an average annual precipitation of 1100 mm. The landforms are characteristics of typical karst valley landscapes, with altitudes ranging from 500 m to 700 m above sea level. The bedrock is mostly Triassic limestone. The soil is developed from Triassic limestone and is classified as calcareous lime soil in the Food and Agriculture Organization system. A rocky desertification landscape is characterized by shallow and discontinuous soil and large areas of bare bedrock (Peng et al. 2019). In the karst trough valley, carbonate rocks developed large amounts of vertical open fissures and sinkholes (Liu et al. 2019a), where surface soil and water quickly drain into the underground karst system. Therefore, severe soil erosion occurs in the karst valley, leading to rocky desertification. Rocky desertification is classified into four categories in China: no, light, moderate, and severe desertification (Jiang et al. 2014). The total area of rocky desertification in the study site is approximately 39 ha, including 26 ha of light desertification, 4.6 ha of moderate desertification, and 8.2 ha of severe desertification, accounting for 67%, 11.7%, and 21%, respectively. Vegetation type is mostly subtropical evergreen broad-leaved forest.

2.2 Experimental design and soil sampling

A space-for-time substitution approach was used to collect soils in November 2013. This approach has been widely adopted for estimating the impact of land use change on soil OC stocks (Li et al. 2012; Xiao et al. 2017). Four vegetation types with different restoration stages, namely, grassland (GL), shrubland (SL), shrub-arbor mixed forestland (SAF), and arbor forestland (AF), were selected, with cropland (CL) as the reference. The basic information of the sample sites is

provided in Table 1. The criteria for selecting land cover types were based on similar geochemical background and soil types. CL was continuously planted with maize for at least 100 years. The maize was sown in late March and harvested in late July. The four vegetation restoration stages were naturally developed from abandoned maize lands. GL was dominated by *Imperata cylindrica* (L.) Beauv. and *Lonicera japonica* Thunb. SL largely consisted of *Quercus acutissima* Carruth and *Alchornea davidii* Franch. The dominant species in SAF were *Robinia pseudoacacia* L., *Broussonetia papyrifera*, and *Trachycarpus fortunei* (Hook.) H. Wendl. AF was established on CL for > 50 years and dominated by *Cyclobalanopsis glauca* (Thunb.) Oerst., *Cinnamomum camphora* (L.) Presl., and *Cupressus funebris* Endl. GL, SL, SAF, and AF were estimated to be 8, 20, 35, and > 50 years, respectively, following agricultural abandonment based on inquiries with native elderly residents.

Two or three sites were established at each land cover type. On each site, two CL plots (5 m × 5 m), two GL plots (10 m × 10 m), two SL plots (10 m × 10 m), three SAF plots (10 m × 10 m), and two AF plots (20 m × 20 m) were used for experiment in November 2013. Three representative soil samples were randomly collected at depths of 0–10 cm from each plot for aggregate determination and SOC concentration. Meanwhile, the soil was also sampled at depths of 0–10 cm by using 100 cm³ stainless steel cylinders for measuring soil bulk density (BD) in each plot. The samples were taken to a laboratory and air-dried.

2.3 Laboratory analysis

Aggregate size classes were separated using the conventional dry sieving method, following the slightly modified method

Fig. 1 Location of the study area (Zhongliang Mountains)

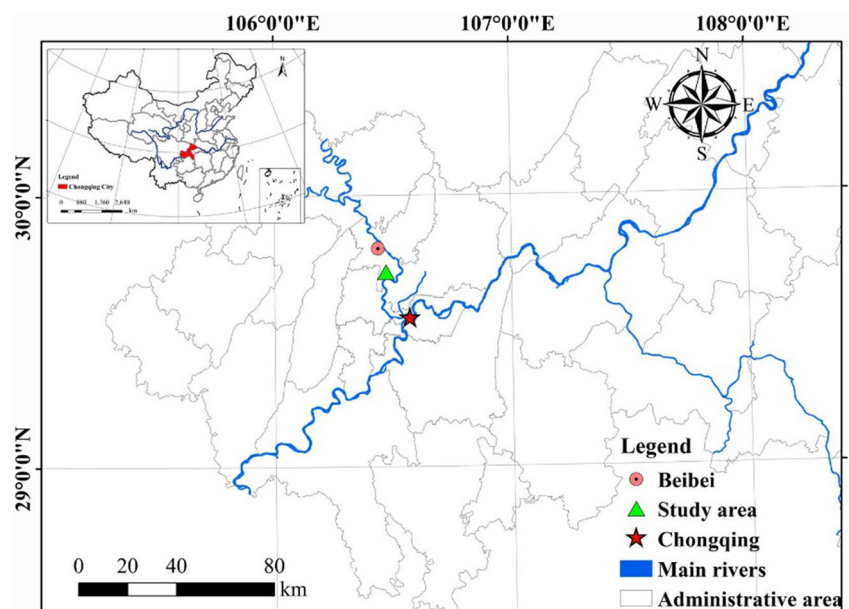


Table 1 Vegetation types and characteristics of study plots

Vegetation types	Altitude (m)	Slope (°)	Coverage (%)	Soil texture (%)			Land use history
				Sand	Silt	Clay	
CL	650–730	5	0	4.37	63.41	32.22	It has been cultivated for more than 100 years.
GL	650–660	6–11	75	2.24	62.32	35.45	Abandoned in about 8 years ago, natural recovery to a grass community
SL	650–660	10–15	60	6.62	47.78	45.60	Cultivated before 20 years ago, natural recovery to a shrub community
SAF	650–665	15–25	70	6.06	59.78	34.16	Cropland land abandoned for 35 years, and natural recovery to shrub and forest mixed community
AF	650–670	12–30	80	4.18	56.86	38.96	Cultivated before 50 years ago, then abandoned, and naturally recovered to secondary forest

Note: CL, cropland; GL, grassland; SL, shrubland; SAF, shrub-arbor mixed forestland; AF, arbor forestland; Coverage of cropland was in sampling time after maize was harvested

described by ISSAS (1978). The fresh and undisturbed soils were immediately taken to the laboratory and broken by manually along natural failure surfaces to obtain aggregate sizes of < 10 mm. These soil samples were air-dried and fractionated into 5–10-, 2–5-, 1–2-, 0.25–1-, and < 0.25-mm classes by gently shaking them through a series of sieves. The air-dried soil samples from each site and each aggregate size were sieved through 0.25-mm sieves to measure OC content. In this study, we classified aggregates into macro- (including 5–10 mm and 2–5 mm), meso- (including 1–2 mm and 0.25–1 mm), and microaggregates (< 0.25 mm).

The OC contents of the aggregate fractions and bulk soil were determined via oxidation with $KCr_2O_7 + H_2SO_4$ and titration with $FeSO_4$. LFOC and EOC concentrations from bulk soil and the aggregate fractions were also analyzed. The light (LF) and heavy fractions (HF) were separated by density fractionation method described by Lu (2000), and the LFOC content was obtained by subtracting the content of heavy fraction organic carbon (HFOC) from the total organic carbon content. EOC was measured using the method presented by Blair et al. (1995).

2.4 Data analysis

Soil OC stocks ($kg\ m^{-2}$) in total soil were calculated as follows (Wei et al. 2013; Qiu et al. 2015):

$$\text{Soil OC stocks} = \frac{OC \times D \times BD}{100} \tag{1}$$

where OC is the total soil OC, LFOC, and EOC concentrations ($g\ kg^{-1}$) of total soil when calculating OC stocks, LFOC

stocks, and EOC stocks in total soil, respectively; D is the thickness (cm) of the 0–10-cm soil depth; and BD is the bulk density ($g\ cm^{-3}$). OC stocks ($g\ m^{-2}$) in each aggregate size fraction were calculated as follows:

$$\text{Stocks of } OC_i = \frac{OC_i \times w_i \times D \times BD}{10} \tag{2}$$

where OC_i is the OC, LFOC, and EOC concentrations of the i th aggregate size fraction ($g\ kg^{-1}$ aggregate) and w_i is the total soil proportion in the i th aggregate size fraction (%).

Soil OC and aggregate size fractions were analyzed using one-way ANOVA (SPSS 17.0) with land cover types. A least significant difference test was conducted to compare between the means at $P < 0.05$. Pearson correlation was used to examine the relationships between OC accumulations in aggregates and total soils and exchangeable calcium.

3 Results

3.1 Aggregate size distribution

Vegetation restoration remarkably affected macro- and mesoaggregate amounts. However, no significant difference in microaggregate amount was observed (Fig. 2). The dominant aggregate size fraction was 5–10 mm, which significantly increased ($P < 0.05$) after vegetation restoration. The proportion of 2–5-mm aggregate size fraction was considerably decreased compared with that in CL. The extent of increase of the 5–10 mm aggregate size fraction was greater than the decrease of the 2–5-mm aggregate size fraction, causing an

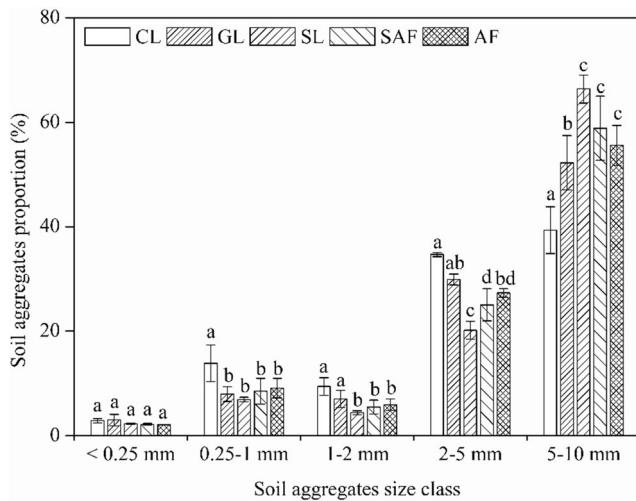


Fig. 2 Soil aggregate proportion (%) at the 0–10 cm depth among vegetation types. Error bars are mean standard error. Different letters reflect significant differences among vegetation types ($P < 0.05$). CL, cropland; GL, grassland; SL, shrubland; SAF, shrub-arbor mixed forestland; AF, arbor forestland. Macroaggregates > 2 mm; mesoaggregates 0.25–2 mm; microaggregates < 0.25 mm

increase in the macroaggregate amount following vegetation restoration. The proportion of mesoaggregate amount (0.25–2 mm) was significantly decreased ($P < 0.05$) after vegetation restoration. A slight decrease was observed in microaggregate amount after the conversion of CL to SL, SAF, and AF.

3.2 Total SOC concentrations and stocks

Total SOC concentrations and stocks significantly increased following vegetation restoration on abandoned CL (Fig. 3). The increase in GL relative to that in CL was insignificant (Fig. 3). SOC concentration decreased in the following order: AF $>$ SAF $>$ SL $>$ GL $>$ CL. The OC concentrations in total soil increased by 11%, 39%, 76%, and 92% after CL conversion into GL, SL, SAF, and AF, respectively. A similar pattern was observed in SOC stocks, with the highest value (3.68 ± 0.2 kg m⁻²) in AF, followed by in SAF (3.46 ± 0.12 kg m⁻²), SL (2.91 ± 0.33 kg m⁻²), GL (2.26 ± 0.24 kg m⁻²), and CL (1.96 ± 0.25 kg m⁻²). Compared with CL, SOC stocks that significantly increased from SL to AF presented an increase of 48%, 78%, and 86%, respectively.

3.3 Liable carbon fraction concentrations and stocks

Changes in LFOC and EOC varied with vegetation types (Fig. 3). Conversion from CL to a natural ecosystem significantly increased OC concentrations and stocks in LFOC and EOC after the SL stage ($P < 0.05$). LFOC and EOC concentrations and stocks exhibited a variation tendency similar to that of SOC following vegetation restoration (Fig. 3). The average increase in LFOC concentrations and stocks ranged from 51% to 194% and from 45% to 201% after the conversion

of CL to a natural ecosystem. EOC concentrations and stocks increased from 42% to 198% and 47% to 191%, respectively. Changes in LFOC and EOC were higher than those in SOC after vegetation restoration.

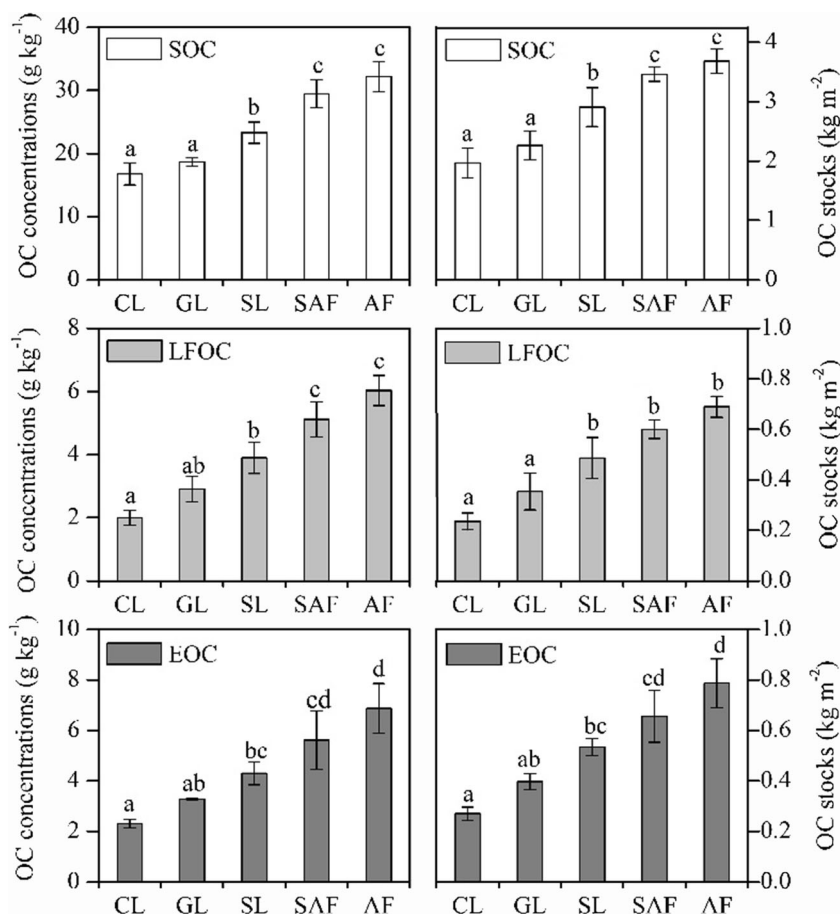
3.4 Aggregate-associated OC concentrations and stocks

Vegetation restoration considerably increased aggregate-associated SOC concentrations (Fig. 4) in the following order: GL $<$ SL $<$ SAF $<$ AF. SOC concentrations varied with different aggregate size fractions. After the conversion of CL to GL, SL, ASF, and SF, the macroaggregate-associated (average of 5–10-mm and 2–5-mm aggregate size fractions), mesoaggregate-associated (average of 1–2-mm and 0.25–1-mm aggregate size fractions) and microaggregate-associated SOC concentrations increased from 15.5% to 104%, 11.5% to 104%, and 36% to 122%, respectively. The microaggregate fraction presented a higher increase than the macroaggregate and mesoaggregate fractions, suggesting that vegetation restoration considerably influenced OC content when the particle size was < 0.25 mm. The distribution trends of aggregate-associated SOC concentrations were similar, and SOC concentrations in aggregates generally increased with a decrease in aggregate size in all vegetable types (Fig. 4).

Vegetation restoration remarkably affected macroaggregate-associated OC stocks, but no considerable effect on mesoaggregate-, and microaggregate-associated OC stocks was observed (Fig. 4). After vegetation restoration, increases were observed in the macroaggregate-, mesoaggregate-, and microaggregate-associated OC stocks. The increase in OC stocks in macroaggregate was mostly observed in the 5–10-mm aggregate size fraction. When land use changed from CL to GL, SL, ASF, and SF, OC stocks in the macroaggregate fractions significantly increased by 57%, 169%, 175%, and 180%, respectively, in the 5–10-mm aggregate size fraction ($P < 0.05$) but only significantly increased by 19% in the 2–5-mm aggregate size fraction after the conversion of CL to AF ($P < 0.05$). The highest increase in aggregate-associated OC stocks was observed in the 5–10-mm aggregate sized fraction in different vegetation stages, followed by in the < 0.25 -mm aggregate size fraction. The conversion of CL to GL and SL decreased the mesoaggregate-associated OC stocks, and the decreases were 23% and 36% in the 1–2-mm class size fraction and 26% and 19% in the 0.25–1-mm aggregate size fraction. However, the mesoaggregate-associated OC stocks in SAF and AF were 14% and 29% higher than those in CL, respectively.

The distribution trend of aggregate-associated SOC stocks was presented as macroaggregate $>$ mesoaggregate $>$ microaggregate, which was the opposite to that of SOC concentrations (Fig. 4). The macroaggregate-associated OC stocks comprised a large proportion of the total soil OC stocks

Fig. 3 The effects of vegetable restoration on SOC concentration and stock at the 0–10 cm depth. Error bars are mean standard error. Different letters mean significant differences among vegetation types ($P < 0.05$). CL, cropland; GL, grassland; SL, shrubland; SAF, shrub-arbor mixed forestland; AF, arbor forestland. SOC, organic carbon in total soils; EOC, easily oxidizable organic carbon; LFOC, light fraction organic carbon



after vegetation restoration, whereas the mesoaggregate-associated OC stocks accounted for only a small proportion. Moreover, the proportion of the microaggregate-associated OC stocks was unaffected by vegetation restoration. In CL soil, the macroaggregate- and mesoaggregate-associated OC stocks accounted for 67% and 29% of the total soil OC stocks, respectively. The conversion of CL to GL, SL, SAF, and AF increased the macroaggregate-associated OC stocks but decreased the mesoaggregate-associated OC stocks (Fig. 4). Macroaggregate- and mesoaggregate-associated OC stock proportions increased and decreased from 9% to 14% and from 10% to 14%, respectively, along vegetation restoration. The variation tendency of the SOC stocks with aggregates was similar to that of aggregate distribution.

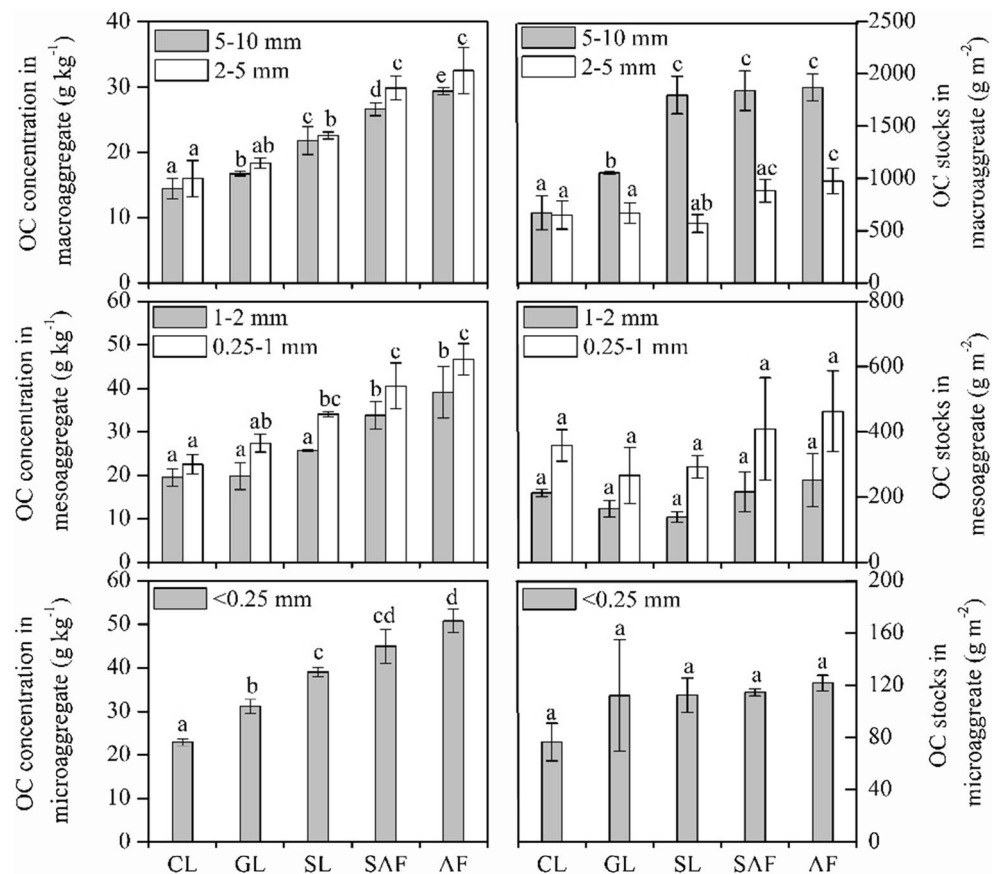
3.5 SOC fraction concentrations and stocks with aggregates

Table 2 shows that vegetation restoration considerably influenced EOC and LFOC concentrations in aggregates. EOC and LFOC concentrations in different aggregate sizes significantly increased after CL was abandoned ($P < 0.05$) and increased in the order of $GL < SL < SAF < AF$. EOC concentrations in macro-, meso-, and microaggregates increased from 58% to

228%, 28% to 214%, and 43% to 200%, respectively. LFOC concentrations in macro-, meso-, and microaggregates increased from 62% to 240%, 44% to 178%, and 52% to 228% along vegetation restoration, respectively. The distribution trend of EOC and LFOC concentrations in aggregates was similar to that of SOC concentrations in aggregates along with vegetation restoration (Fig. 4, Table 2). The highest and lowest concentrations were observed in micro- and macroaggregates, respectively. Overall, EOC and LFOC concentrations in aggregates increased with a decrease in aggregate size in all vegetation restoration stages. This trend was similar to that of SOC concentration.

Vegetation restoration significantly increased soil EOC and LFOC stocks with macro- and microaggregates, but mesoaggregate-associated EOC and LFOC stocks were unaffected by natural recovery (Table. 1). Macroaggregate- and microaggregate-associated EOC stocks increased from 32% to 253% and 52% to 129%, respectively, and significantly increased from SL to AF (Table. 1). A significant increase was observed in mesoaggregate-associated EOC stocks in AF relative to that in CL (Table. 1). The highest increase was observed in the 5–10-mm fraction. Similar to EOC stocks in aggregates, LFOC stocks with macro- and microaggregates significantly increased from SL to AF and only significantly

Fig. 4 The effects of vegetable restoration on aggregate-associated OC concentrations and stocks at the 0–10 cm depth. Different letters mean significant differences among vegetation types ($P < 0.05$). CL, cropland; GL, grassland; SL, shrubland; SAF, shrub-arbor mixed forestland; AF, arbor forestland. Macroaggregates > 2 mm; mesoaggregates 0.25–2 mm; microaggregates < 0.25 mm



increased in mesoaggregate-associated LFOC stocks in AF relative to that in CL. Macroaggregate- and microaggregate-associated LFOC stocks increased from 39% to 391% and 61% to 136%, respectively. The highest increase was also observed in the 5–10-mm fraction. Overall, the EOC and LFOC stocks in aggregates increased with an increase in aggregate size in all vegetation restoration stages. This trend was similar to that of SOC stocks. The proportion of the macroaggregate-associated EOC and LFOC stocks increased from 9% to 14% and 13% to 24%, respectively, and the proportion of the mesoaggregate-associated OC stocks decreased from 7% to 13% and 13% to 19%, respectively.

4 Discussion

4.1 Impacts of vegetation restoration on soil aggregates

Our results showed that the amount of macroaggregates (> 2 mm) increased. This finding is consistent with the result of Zhong et al. (2019), wherein the proportions of macroaggregates (> 2 mm) increased significantly after farmland was changed to three types of vegetated soil. In our study, the amounts of meso- (0.25–2 mm) and microaggregates

(<0.25 mm) decreased after the conversion of CL into GL, SL, SAF, and AF. These findings are inconsistent with the results of Zhong et al. (2019), who reported that farmland conversion into three types of vegetated soil increased the proportion of mesoaggregates (0.25–2 mm) but decreased the amount of microaggregates (0.053–0.25 mm). Tang et al. (2016) used the dry sieving method and found that the amount of > 5 -mm aggregates significantly increased but the amount of < 0.25 -mm aggregates significantly decreased from bare land to woodland and GL in a karst region. These results suggested that vegetation restoration can improve macroaggregate formation. Afforestation or vegetation restoration increases the input of plant residues, root biomass, and exudates, enhancing soil aggregation (Tisdall and Oades 1982; Wei et al. 2012; Zhong et al. 2019). The decrease in disturbances following vegetation restoration promotes soil aggregation (Tang et al. 2016; Zhong et al. 2019). By contrast, tillage or artificial disturbances can break down large aggregates into small fractions (Liu et al. 2014; Tang et al. 2016). Our results showed that CL had a lower amount of 5–10-mm aggregates than the other land use types (Fig. 2). This finding may be attributed to macroaggregate destruction by tillage. Mesoaggregate (2–0.25 mm) amount significantly increased (Qiu et al. 2015; Wei et al. 2012; Deng et al. 2018; Zhong et al. 2019), using the wet sieving method, and the 2–5-mm aggregates also

Table 2 EOC and LFOC concentrations and stocks in aggregates

Vegetation types	Aggregate sizes				
	5–10 mm	2–5 mm	1–2 mm	0.25–1 mm	< 0.25 mm
EOC concentrations (g kg ⁻¹)					
CL	2.18 ± 0.41 a	1.88 ± 0.12 a	2.54 ± 0.13 a	3.22 ± 0.33 a	4.11 ± 0.86 a
GL	2.95 ± 0.01 a	3.38 ± 0.17 ab	3.11 ± 0.64 a	4.31 ± 0.30 b	5.87 ± 0.21 b
SL	3.90 ± 0.57 a	4.00 ± 0.30 ab	4.57 ± 0.11 a	7.02 ± 0.24 c	9.79 ± 0.45 c
SAF	5.05 ± 1.09 b	5.49 ± 1.51 bc	6.98 ± 1.36 b	7.88 ± 0.13 d	11.12 ± 0.26 d
AF	5.67 ± 0.72 b	7.41 ± 1.33 c	8.37 ± 0.78 b	9.57 ± 0.09 e	12.32 ± 0.29 e
LFOC concentrations (g kg ⁻¹)					
CL	1.44 ± 0.02 a	1.86 ± 0.54 a	2.50 ± 0.45 a	3.49 ± 0.60 a	3.45 ± 0.15 a
GL	2.34 ± 0.11 a	3.00 ± 0.67 b	3.86 ± 0.52 ab	4.67 ± 1.32 ab	5.22 ± 0.68 b
SL	3.46 ± 0.67 b	4.20 ± 0.02 c	4.61 ± 0.35 bc	5.90 ± 0.23 b	7.37 ± 0.30 c
SAF	4.30 ± 0.44 bc	5.42 ± 0.45 d	5.75 ± 0.92 c	8.15 ± 0.86 c	10.07 ± 0.49 d
AF	5.12 ± 0.52 c	6.00 ± 0.03 d	7.52 ± 0.60 d	8.84 ± 0.04 c	11.29 ± 1.31 d
EOC stocks (g m ⁻²)					
CL	101.71 ± 32.75 a	75.94 ± 2.20 a	27.63 ± 2.87 a	51.20 ± 6.74 ab	13.93 ± 5.01 a
GL	185.50 ± 5.10 a	122.17 ± 6.80 ab	27.23 ± 13.47 a	41.10 ± 7.47 a	21.19 ± 8.36 ab
SL	321.47 ± 47.27 b	100.00 ± 5.05 ab	24.70 ± 2.46 a	60.06 ± 4.10 ab	28.06 ± 1.28 bc
SAF	344.83 ± 43.62 b	159.89 ± 42.85 bc	44.27 ± 12.86 ab	78.02 ± 20.35 bc	28.49 ± 2.42 bc
AF	359.22 ± 14.33 b	221.78 ± 44.20 c	58.80 ± 15.07 b	114.91 ± 22.65 d	31.95 ± 0.54 c
LFOC stocks (g m ⁻²)					
CL	66.16 ± 10.10 a	75.60 ± 24.72 a	26.93 ± 0.60 a	55.04 ± 3.42 ab	11.45 ± 1.29 a
GL	146.98 ± 2.29 b	109.83 ± 35.82 a	32.15 ± 5.50 ab	46.53 ± 24.01 a	18.47 ± 5.63 ab
SL	285.16 ± 55.44 c	105.33 ± 12.59 ab	25.04 ± 4.97 a	50.66 ± 7.12 ab	21.12 ± 1.07 bc
SAF	296.29 ± 23.73 c	160.52 ± 31.34 bc	36.87 ± 12.48 ab	81.21 ± 25.40 ab	25.78 ± 2.01 c
AF	324.73 ± 4.79 c	190.56 ± 3.12 c	54.18 ± 13.12 b	93.87 ± 17.31 b	27.04 ± 3.03 c

Note: CL, cropland; GL, grass land; SL, shrubland; SAF, shrub-arbor mixed forestland; AF, arbor forestland. Macroaggregates > 2 mm; mesoaggregates 0.25–2 mm; microaggregates < 0.25 mm. Means and standard errors. With columns, by the different lowercase letters indicate significantly different ($P < 0.05$) in different vegetation types

increased (Zhong et al. 2019) after the conversion of CL to natural vegetation. These findings are inconsistent with our results due to the different separation methods for aggregates used in the present study. Thus, the amount of 2–5-mm size fractions in macro- and mesoaggregates (0.25–2 mm) decreased in this study.

4.2 Accumulation of OC concentrations and stocks following vegetation restoration

Land use change considerably affects soil carbon dynamics by altering carbon input, decomposition, and turnover (Zhang et al. 2015; Deng et al. 2016). The conversion of CL to natural vegetation can enhance SOC by increasing carbon input from new vegetation (Laganière et al. 2010; Han et al. 2017; Xiao et al. 2017; Deng et al. 2018; Hu et al. 2018a). Similar results were found in our study, wherein SOC concentrations and stocks at soil depth of 0–10 cm significantly increased along with vegetation restoration (Fig. 3). Several explanations were

proposed. First, vegetation restoration increases carbon input from aboveground and belowground biomass, facilitating SOC accumulation. This explanation is also supported by previous studies (Laganière et al. 2010; Deng et al. 2016). Second, an increase in OC input can promote soil aggregate formation and stability (Tang et al. 2016; Zhong et al. 2019), thereby facilitating the physical protection of SOC in aggregates (Blanco-Canqui and Lal 2004). In the present study, vegetation restoration significantly increased the amount of macroaggregates (5–10 mm, Fig. 2), promoting soil aggregation. The lowest SOC concentrations and stocks in CL can be attributed to OC loss resulting from high decomposition, soil erosion, and OC input reduction with tillage (Deng et al. 2016). Long-term tillage destroyed soil aggregates, leading to an increase in the amount of 0.25–5-mm aggregates in CL compared with that in vegetation restoration stages. These findings suggested that vegetation restoration tends to concentrate on large aggregates, protecting SOC in aggregates. However, tillage broke down large aggregates into small aggregates.

The EOC and LFOC fractions exhibited increasing trends with vegetation restoration, similar to that of SOC (Fig. 3). This observation is consistent with a previous study (Zhao et al. 2015). The higher concentrations and stocks of EOC and LFOC in vegetation restoration stages are possibly due to the supply of plant litter and roots in soil compared with that in CL. This finding is also supported by other studies (Elliott 1986; Zhao et al. 2015; Yu et al. 2017). By contrast, a decrease in aboveground biomass and root biomass and an increase in OC mineralization by tillage and fertilization can result in the lowest LFOC concentrations in CL (Wei et al. 2014). For example, the harvest and removal of maize in CL decrease litter input to soil annually. EOC is higher in forest and GL than in farmland due to the complete decomposition of organic matter and the decrease in biomass amount in farmland (Liu et al. 2014). By contrast, the amount of vegetative coverage and the quantity and decomposition of litter result in a considerable amount of carbon input in forest. The liable carbon fraction is more sensitive to land use change or management practices than total SOC (Liu et al. 2014; Saha et al. 2011; Chen et al. 2017). The results of the present study demonstrated that the conversion of CL to GL, SL, SAF, and AF significantly increases total SOC, EOC, and LFOC concentrations and stocks (Fig. 3). Our results exhibited the average increases of 54%, 117%, and 124% in total SOC, EOC, and LFOC concentrations, respectively, after CL conversion to natural vegetation. Similar results were found in OC stocks, wherein vegetation restoration increased by an average of 57%, 119%, and 127% in total SOC, EOC, and LFOC stocks, respectively. These findings confirmed that changes in OC concentrations and stocks can be more pronounced in the liable carbon fraction than in the total SOC.

SOC can accumulate after vegetation restoration, but SOC accumulation with aggregates has received minimal attention in karst regions. Vegetation restoration considerably affects the concentrations and stocks of SOC, EOC, and LFOC associated with different aggregate sizes (Table 3, Fig. 4). Total SOC, EOC, and LFOC concentrations with aggregates considerably increased along with vegetation restoration. This result is consistent with the finding that OC in aggregates significantly increases following vegetation restoration (Wei et al. 2012; Deng et al. 2018; Zhong et al. 2019). OC concentrations in aggregates increase with an increase in aggregate size in all land uses (Fig. 3, Table 2). Microaggregate fraction exhibited a higher increase than macro- and mesoaggregate fractions, suggesting that vegetation restoration considerably influences OC concentration when particle size is <0.25 mm. Similar findings were reported by Li et al. (2013) and Tang et al. (2016), who found that SOC in microaggregates (<0.25 mm) were higher than in other aggregate sizes. Tisdall and Oades (1982) reported that OC concentrations increase as aggregate size decreases. These results are consistent with aggregate

hierarchy theory, which states that OC preferentially accumulates in small aggregate size fractions (Zhang et al. 2015). This result may be due to the high CaCO₃ and clay contents of lime soil in karst areas (Laganière et al. 2010). Exchangeable calcium is the key factor that is responsible for soil carbon dynamics (Xiao et al. 2017). This finding confirmed that calcium exhibits significant positive correlations with OC in total soil and aggregates (Table 3). However, an increase in SOC concentrations in aggregates with an increase in size class can be due to the integration of small aggregates into large aggregates and organic binding agents (Yu et al. 2017). This aggregate hierarchy concept relies on organic matter as the primary binding agent (Liu et al. 2018). Changes in aggregate-associated OC stocks are generally related to changes in aggregate amount and aggregate-associated OC concentrations (Wei et al. 2012; Qiu et al. 2015). Thus, variations in aggregate-associated OC stocks were inconsistent with the changes in aggregate-associated OC concentrations (Fig. 4). SOC stocks were determined more by macroaggregate-associated OC stocks than mesoaggregate- and microaggregate-associated OC stocks. The lowest aggregate-associated OC concentrations were observed in macroaggregates, but the highest aggregate amount was the 5–10-mm aggregates, suggesting that macroaggregate-associated OC stocks contributed the most to total SOC stocks (Fig. 4). However, the highest aggregate-associated OC concentrations was found in microaggregates, and the lowest aggregate amount was observed in microaggregates (<3%, Fig. 2), resulting in <5% total SOC stocks (Fig. 4). The significant increase in macroaggregate-associated OC stocks was primarily due to an increase in macroaggregate amount and macroaggregate-associated OC concentrations. These results are similar to those of Wei et al. (2012). An increase in microaggregate-associated OC stocks was mostly attributed to an increase in microaggregate-associated OC concentrations because microaggregate amount slightly decreased after the conversion of CL to other vegetation types. This observation is consistent with the results of Wei et al. (2012), wherein a net increase in microaggregate-associated OC stocks was found because the increase in OC concentration (165%) in microaggregates was higher than the decrease in microaggregate amount (48%). The changes in mesoaggregate-associated OC stocks varied with vegetation restoration, decreasing by 24% in GL and 26% in SL and increasing by 8% in SAF and 24% in AF. The decrease in mesoaggregate amount was higher than the increase in mesoaggregate-associated OC concentrations, decreasing mesoaggregate-associated OC stocks. By contrast, the increase in mesoaggregate-associated OC concentrations was higher than the decrease in mesoaggregate amount, resulting in the increase in mesoaggregate-associated OC stocks. Therefore, the increase in SOC stocks was primarily

contributed by OC accumulation in macroaggregates following vegetation restoration.

4.3 Controlling factors of OC accumulation along vegetation restoration

We found no significant correlation between OC stocks in total soil and in all soil aggregates (Table 3). Stronger correlations between OC stocks in total soil and macroaggregate-

and microaggregate-associated OC stocks than those between mesoaggregate-associated OC and OC stocks in total soil were found in the present study. These results indicated that OC stocks in total soil were more dependent on OC stocks in macro- and microaggregates than those in mesoaggregates. The relationships between aggregate-associated EOC stocks and soil EOC stocks and between LFOC stocks and soil LFOC stocks were also greater than those between aggregate-associated and SOC stocks (Table 3). This finding

Table 3 Relationships between OC accumulations in aggregates and total soils and exchangeable Ca

		SOC stocks	Calcium
Aggregate-associated SOC concentrations	5–10 mm		0.733*
	2–5 mm		0.708*
	1–2 mm		0.714*
	0.25–1 mm		0.746**
	< 0.25 mm		0.769**
SOC concentrations			0.722*
Aggregate-associated SOC stocks	5–10 mm	0.896**	0.756**
	2–5 mm	0.778**	–
	1–2 mm	–	–
	0.25–1 mm	–	–
	< 0.25 mm	0.679*	–
SOC stocks			0.710*
Aggregate-associated EOC concentrations	5–10 mm		0.739**
	2–5 mm		0.685*
	1–2 mm		0.658*
	0.25–1 mm		0.640*
	< 0.25 mm		0.734*
EOC concentrations			0.704*
Aggregate-associated EOC stocks	5–10 mm	0.926**	0.802**
	2–5 mm	0.900**	–
	1–2 mm	0.820**	–
	0.25–1 mm	0.870**	–
	< 0.25 mm	0.837**	–
EOC stocks			0.710*
Aggregate-associated LFOC concentration	5–10 mm		0.754**
	2–5 mm		0.688*
	1–2 mm		0.634*
	0.25–1 mm		0.677*
	< 0.25 mm		0.752**
LFOC concentrations			0.711*
Aggregate-associated LFOC stocks	5–10 mm	0.935**	0.789**
	2–5 mm	0.906**	–
	1–2 mm	0.703*	–
	0.25–1 mm	0.758**	–
	< 0.25 mm	0.944**	0.602*
LFOC stocks			0.696*

Note: “—” indicates that the correlation is not significant ($P > 0.05$). ** means significant at $P < 0.01$, * means significant at $P < 0.05$. Ca means exchangeable calcium content in total soils

suggested that EOC and LFOC stocks are more sensitive than SOC stocks along with vegetation restoration. Exchangeable calcium is the key factor that affects soil OC dynamics in a few studies in karst regions (Yang et al. 2016; Xiao et al. 2017). Our study also found significantly positive correlations between exchangeable calcium contents and OC concentrations and stocks in total soil in aggregate sizes (Table 3). Limestone addition or increased calcium level in soil can also promote SOC sequestration (Kaiser et al. 2011; Morris et al. 2007). This observation showed OC stabilization via soil organic matter interaction with mineral surfaces through polyvalent cation bridges (Kaiser et al. 2011; von Lützow et al. 2006), and calcium and magnesium are the major cations in neutral and alkaline soils (Xiao et al. 2017). Thus, soil calcium may be a key factor that affects OC dynamics in aggregates in the present study. However, the relationships between mesoaggregate-associated OC stocks and exchangeable calcium contents and between SOC and EOC stocks in the microaggregates and exchangeable calcium contents were insignificant (Table 3). Therefore, vegetation restoration increases soil exchangeable calcium levels, enhances soil aggregates and OC concentrations in soil aggregates, and facilitates OC stock accumulation in macroaggregates (predominantly) and microaggregates, leading to OC accumulation in total soils.

5 Conclusions

The results of our study showed that vegetation restoration considerably affected the amount of soil aggregates, OC concentrations, and OC stocks in total soil and soil aggregates. Changes in OC concentrations and stocks can be more pronounced in the liable carbon fraction than in total SOC. Vegetation restoration increased soil macroaggregate amounts (5–10 mm) and OC concentrations in aggregates and resulted in OC accumulation in the total soil and macroaggregates. The increase in SOC was mainly attributed to OC accumulation in macroaggregates. The increase in macroaggregate-associated OC stocks was due to increases of macroaggregate amount and macroaggregate-associated OC concentrations. Exchangeable calcium also affected soil OC accumulation in total soils and soil aggregates.

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