



Total soil organic carbon increases but becomes more labile after afforestation in China's Loess Plateau

Qingyin Zhang^a, Xiaoxu Jia^{b,c}, Xiaorong Wei^a, Mingan Shao^{a,b,c}, Tongchuan Li^a, Qiang Yu^{a,c,d,*}

^a State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, Yangling 712100, China

^b Key Laboratory of Ecosystem Network Observation and Modeling, Institute of Geographic Sciences and Natural Resources Research, Chinese Academy of Sciences, Beijing 100101, China

^c College of Resources and Environment, University of Chinese Academy of Sciences, Beijing 100190, China

^d School of Life Sciences, University of Technology Sydney, P.O. Box 123, Broadway, NSW 2007, Australia



ARTICLE INFO

Keywords:

Afforestation
Cropland
Labile soil organic carbon
Loess Plateau

ABSTRACT

Afforestation of cropland is recommended as an effective approach to enhance soil organic carbon (SOC) sequestration and labile organic C fractions. However, the stabilization of SOC and its labile organic C fractions on the Loess Plateau is largely unknown. Our objective was to quantify total SOC concentration and labile organic C fractions in the 0–20 cm soil depth for four land use types on the Loess Plateau, including cropland and three afforested areas (composed of *R. pseudoacacia* forests, *P. tabuliformis* forests, and *R. pseudoacacia* + *P. tabuliformis* mixed forests). Total SOC concentration, particulate organic C (POC), dissolved organic C (DOC), microbial biomass C (MBC), and potassium permanganate-oxidizable C (KMnO₄-C) were measured. Carbon management index (CMI) was also calculated. Afforestation showed a significant positive effect on total SOC and labile organic C fractions, compared with cropland. Afforestation with *R. pseudoacacia*, *P. tabuliformis*, and *R. pseudoacacia* + *P. tabuliformis* significantly increased POC by 57.4%, 22.2%, and 44.4% in the 0–5 cm soil layer; and similar increases were observed in the 5–10 cm and 10–20 cm layers. Similar trends to those observed for POC in response to afforestation were also seen for DOC, MBC, and KMnO₄-C. Afforestation with *R. pseudoacacia* resulted in the highest total SOC concentrations and labile organic C fractions among the three afforestation treatments. These findings suggested that although afforestation can significantly promote total SOC accumulation, especially with *R. pseudoacacia*, SOC may become more labile following afforestation in the future.

1. Introduction

Forests are among the most productive and ecologically valuable ecosystems in the world (Richter et al., 1999; Foley et al., 2005; Miles and Kapos, 2008). They provide critical ecosystem services, including carbon (C) storage, biodiversity conservation, water purification, and erosion control (Lal, 2005; Metz et al., 2007; Canadell and Raupach, 2008). One of the most important ecological functions of forests is their role in global C cycles (Houghton and Hackler, 1999; Six et al., 2002; Guo and Gifford, 2002). Soil organic C (SOC) is the largest C stock in the terrestrial ecosystem (Batjes, 1996). Soil organic C stocks are controlled by the balance between C inputs and outputs from soils, and afforestation may influence C input as well as output fluxes from ecosystems (Guo and Gifford, 2002; Lal, 2004). Reports from around the world indicate that afforestation can increase soil C sequestration by simultaneously decreasing C loss from decomposition and erosion (Laganière et al., 2010; Xiao et al., 2015; Deng et al., 2016; Li et al.,

2018). However, some studies have reported negligible effects of afforestation on C sequestration. (Rytter, 2016; Chen et al., 2017). The positive effects of afforestation are particularly important in arid and semi-arid regions where fragile ecosystems can suffer severe soil degradation and erosion.

Because soil C accumulation plays an important role in climate change mitigation, numerous studies have reported the dynamics of total organic C and labile organic C fractions (e.g. dissolved organic C (DOC), microbial biomass C (MBC), particulate organic C (POC), and potassium permanganate-oxidizable C (KMnO₄-C)) with regard to afforestation in different regions (Blair et al., 1995; Wang et al., 2009; Shang et al., 2018; Bargali et al., 2018; Kooch et al., 2019; Pang et al., 2019). Soil total C generally has been reported to increase following afforestation. For example, Martens et al. (2003) found that soil C accumulated at an average rate of 0.62 Mg ha⁻¹ y⁻¹ during cropland conversion to forest in Central America. Globally, Post and Kwon (2000) found that the average rate of soil C accumulation for forest

* Corresponding author at: State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, Yangling 712100, China.
E-mail address: yq@nwfau.edu.cn (Q. Yu).

established on cropland was $0.34 \text{ Mg ha}^{-1} \text{ y}^{-1}$. Guo and Gifford (2002) concluded that soil C stocks significantly increased when cropland was converted to tree plantation (+18%) and secondary forest (+53%). However, Vesterdal et al. (2002) observed that afforestation on cropland did not lead to an increase in soil organic C over a 30-yr period, but led to a redistribution of SOC in the soil profile. Some findings about the changes of soil labile organic C following afforestation have also been reported. For instance, in a temperate forest, Kooch et al. (2019) reported that a tree plantation showed higher POC and DOC than a mixed natural forest. Natural restoration resulted in markedly higher SOC and labile organic C than artificial afforestation in the karst regions of southwestern China (Pang et al., 2019). Bargali et al. (2018) reported that mixed grassland-pine forest was better in sustaining the soil MBC than pure oak or pine forest. They indicated that tree species is a key factor in determining SOC fractions along with vegetation succession. The inconsistencies of soil C dynamics observed in these studies are likely a result of multiple factors, including climate, soil type, soil depth, and tree species (Paul et al., 2002; Lal, 2004; Laganière et al., 2010; Li et al., 2012). Even though several studies have considered the changes of total organic C and labile organic C in pure forest types or different plantations, some recent studies comparing pure vs. mixed stands obtained different results for labile organic C (Shang et al., 2018; Bargali et al., 2018; Kooch et al., 2019; Pang et al., 2019). Moreover, few studies have assessed the possible effects of different tree species mixtures in China's Loess Plateau.

China implemented the "Grain for Green" programs with the objective of restoring degraded cropland to forest and grassland. *R. pseudoacacia* was chosen as one of the tree species because of its N_2 -fixation capability, an important pathway for atmospheric N_2 input into terrestrial ecosystems. Additionally, the species *P. tabuliformis* (non- N_2 -fixing) was also used as a prominent artificial afforestation forest in the Loess Plateau. Consequently, this project has significantly increased total organic C accumulation in the soil profile and enhanced total organic C storage for over a decade on the Loess Plateau (Zhang et al., 2013; Deng et al., 2014, 2017). However, it is not clear whether SOC has become more labile as total SOC increased following afforestation. Mixed plantations that included N_2 -fixing trees species are thought to achieve greater soil C sequestration through improved growth compared with monocultures (Forrester et al., 2006). Similarly, Sayyad et al. (2006) found that foliar N concentrations increased significantly for a non- N_2 -fixing tree species in mixed cultivation with an N_2 -fixing species. Although there have been some studies investigating the effects of N_2 -fixing species on plant nutrition and total SOC, a consensus on which tree species (N_2 -fixing species, non- N_2 -fixing species, or mixed forest) has a more significant effect on soil labile organic C fractions has yet to be achieved. Such information is essential for determining the

success of afforestation and restoration programs in the Loess Plateau of China.

Considering the traits of these species in the Loess Plateau, our objective was to test the following two hypotheses: (1) afforestation leads to a substantial gain in total organic C, but SOC may become more labile in the three surface soil layers (0–5 cm, 5–10 cm and 10–20 cm); (2) *R. pseudoacacia* forests will result in greater carbon benefits due to its ability to fix atmospheric nitrogen compared with *P. tabuliformis* forests. To test our hypotheses, total organic C and labile organic C fractions (e.g., DOC, MBC, POC, and $\text{KMnO}_4\text{-C}$) in the three soil layers designated above were examined in three forests (*R. pseudoacacia*, *P. tabuliformis*, *R. pseudoacacia* + *P. tabuliformis* mixed forest) and compared with those same soil carbon fractions from cropland sites.

2. Materials and methods

2.1. Study area

This study was carried out at the Yehe National Forestry Center of Fufeng County, Shaanxi Province, China (34.55°N , 107.90°E ; 1080 m a.s.l.). The study area is part of the Qishui watershed which belongs to the hilly and gully zone of the Loess Plateau (Zhang et al., 2019). The study site features a temperate, semi-humid climate, with a mean annual temperature of 11.5°C and a mean annual precipitation of 592 mm, 70% of which occurs during the growing season from June to September. The soil is mainly Gleyic Phaeozems (World Reference Base for Soil Resources), with soil texture of 11% sand, 20% clay, and 69% silt (Zhang et al., 2018). In the study area, there are deciduous broad-leaf and evergreen coniferous forests characterized by *R. pseudoacacia* Linn. and *P. tabuliformis* Carr., which dominate artificial afforestation forests. Maize (*Zea mays* L.) and wheat (*Triticum aestivum* L.) are the predominant crops in this region.

2.2. Soil sampling and processing

The three forests in our study were: (i) *R. pseudoacacia* forests, 20 years old; (ii) *P. tabuliformis* forests, 28 years old; and (iii) *R. pseudoacacia* + *P. tabuliformis* mixed forests, 25 years old. A cropland site, harvested once a year, was used as the control because the three afforested areas had been converted from croplands. For the cropland area, both nitrogen and phosphorus (urea + P_2O_5 , dry form) were applied at the rates of $100 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ and $50 \text{ kg P ha}^{-1} \text{ yr}^{-1}$ annually at the end of May. Three blocks (distance of 500 m from each other) with similar climatic conditions, soil texture (Table 1), and parent material were randomly selected in the cropland area and each afforested area, which may minimize confounding site factor effects.

Table 1

Physical and chemical soil properties in June in 2018 (0–5, 5–10, and 10–20 cm soil layers) for cropland and three afforestation areas (RF: *Robinia pseudoacacia* forest; PF: *Pinus tabuliformis* forest; RPF: *Robinia pseudoacacia* and *Pinus tabuliformis* forest). Different lowercase letters in the same soil layer indicate a significant difference at $P < 0.05$ among the different land-use types. Values represent mean \pm SD for sample size of $n = 6$.

Land use	Soil layers (cm)	Sand (%)	Silt (%)	Clay (%)	BD (g cm^{-3})	Total organic C concentration (g kg^{-1})	Total organic C stocks (kg m^{-2})
Cropland	0–5	10.2(0.3)	68.9(3.5)	20.9(1.2)	1.28(0.1)	11.5(1.1)c	7.36(0.05)b
	5–10	11.6(0.4)	69.6(4.1)	18.8(0.9)	1.36(0.1)	8.4(0.8)b	5.71(0.04)b
	10–20	11.8(0.5)	70.5(2.9)	17.7(1.4)	1.38(0.1)	7.6(0.6)b	10.49(0.06)b
RF	0–5	7.6(0.4)	70.9(3.7)	21.5(1.8)	1.18(0.2)	21.8(1.3)a	12.86(0.09)a
	5–10	9.8(0.8)	68.1(2.4)	22.1(2.0)	1.29(0.1)	12.9(1.2)a	8.32(0.07)a
	10–20	12.1(0.9)	69.3(4.1)	18.6(1.2)	1.42(0.1)	9.3(0.7)a	13.77(0.09)a
PF	0–5	8.8(0.7)	74.8(5.6)	16.4(1.2)	1.23(0.1)	14.5(0.9)b	8.92(0.10)b
	5–10	9.2(0.8)	73.5(4.1)	17.3(1.4)	1.38(0.1)	8.8(0.8)b	5.93(0.07)b
	10–20	10.7(0.8)	71.9(6.5)	17.4(1.3)	1.45(0.1)	8.7(0.4)a	11.17(0.07)b
RPF	0–5	8.2(0.7)	73.1(5.8)	18.7(1.1)	1.19(0.1)	19.1(1.7)a	11.36(0.08)a
	5–10	9.5(0.6)	72.8(4.9)	17.7(0.9)	1.26(0.1)	12.6(1.1)a	7.94(0.05)a
	10–20	11.8(1.1)	71.2(5.2)	17(0.8)	1.42(0.1)	9.2(0.8)a	13.06(0.10)a

Three replicated plots (5 m × 5 m for cropland, 20 m × 20 m for the three afforested areas) were randomly chosen in each block.

Soil samples were taken using a 5.0 cm diameter auger in each plot in June 2018 from five locations (including the four corners and the center). This was the time just before *R. pseudoacacia* trees shed their leaves. Each soil sample was divided into three layers (0–5 cm, 5–10 cm, and 10–20 cm) and the five samples from each layer were composited into one sample. After roots and other plant debris were removed, the samples were divided into two parts. One part of the fresh soil sample was sieved moist (< 2 mm) and kept at 4 °C for DOC and MBC analysis conducted within two weeks of sampling. The other portion of each soil sample was air-dried and kept at room temperature for analysis of SOC concentration, POC, and KMnO₄-C. Three additional soil cores per plot were taken from the three soil layers using stainless steel cylinders (5 cm inner diameter and 5 cm height) for bulk density (BD) analysis. The main physical and chemical properties of the soil for the four land use types are shown in Table 1. In addition, three sampling points were randomly selected in each plot using a 1 m × 1 m quadrat for the aboveground biomass measurements of understory vegetation on the same date. All of the biomass samples were then oven-dried at 70 °C to a constant weight and weighed to determine aboveground biomass.

2.3. Soil analysis and data calculation

Soil particle-size distributions were determined using the laser-diffraction method with a Mastersizer 2000 particle-size analyzer (Malvern Instruments, Worcestershire, UK). Total SOC was determined by the dichromate wet oxidation method (Nelson and Sommers, 1982), which may result in smaller values than the dry combustion method. However, this difference will not affect our comparative study among the afforestation treatments. For DOC, the moist soil was extracted with 50 mL of 0.5 mol L⁻¹ K₂SO₄ for 1 h. The extracts were filtered through a 0.45 μm membrane filter and analyzed using a Multi 3100 N/C analyzer (Analytik Jena, Germany) (Jones and Willett, 2006). MBC was measured by the fumigation-extraction method (Vance et al., 1987). In brief, extracted C concentration was determined using a Multi 3100 N/C analyzer (Analytik Jena, Germany). Extracted C concentration was calculated as: MBC (mg kg⁻¹) = (fumigated C-non-fumigated C)/0.38 (Vance et al., 1987). POC was measured as described by Cambardella and Elliott (1992). In brief, 20 g of air-dried soil and 70 mL Na hexametaphosphate (5 g L⁻¹) were shaken on a centrifuge for 18 h. The soil suspension was poured over a 53-μm sieve and the retained coarse fraction was rinsed with a weak stream of distilled water. All materials remaining on the sieve were washed into a dry dish, oven dried at 60 °C for 48 h, and ground to determine C content. KMnO₄-C was measured as described by Blair et al. (1995), and the change in concentration of KMnO₄ was used to estimate the content of C oxidized, assuming that 1.0 mmol L⁻¹ of MnO₄⁻ was consumed (Mn⁷⁺ → Mn²⁺) in the oxidation of 0.75 mmol L⁻¹ (9.0 mg) of carbon.

The carbon management index (CMI) provides a reliable measure of the change in soil C dynamics of an experimental object relative to a reference object (Blair et al., 1995). CMI was determined as described by Blair et al. (1995). In our study, this index was calculated for the three afforestation areas using a reference sample value (the cropland) as follows:

$$\text{CMI} = \text{Carbon pool index(CPI)} \times \text{Lability index(LI)} \times 100$$

$$\text{CPI} =$$

$$\frac{\text{Experimental sample total organic C}}{(\text{g kg}^{-1})/\text{Reference sample total organic C}(\text{g kg}^{-1})}$$

$$\text{LI} = \frac{\text{Lability of C(L)in experimental sample soil/Lability of C(L)in reference soil}}{\text{L} = \text{KMnO}_4 - \text{C}/(\text{total organic C} - \text{KMnO}_4 - \text{C})}$$

$$\text{L} = \text{KMnO}_4 - \text{C}/(\text{total organic C} - \text{KMnO}_4 - \text{C})$$

2.4. Statistical analysis

SPSS ver. 18.0 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis of the data. At each soil layer, the effects of afforestation on soil BD, total SOC concentrations, organic C stocks, labile organic C fractions (POC, KMnO₄-C, MBC, and DOC), CMI, and SI were analyzed using one-way ANOVA. Two-way ANOVA was performed to examine the differences in total SOC concentrations, organic C stocks, labile organic C fractions (POC, KMnO₄-C, MBC, and DOC), with afforestation and soil layer as two fixed factors. Multiple comparisons of the means were performed using the least significant difference test ($P < 0.05$).

3. Results

3.1. Total organic C concentrations and organic C stocks

Afforestation significantly affected total SOC concentrations and organic C stocks (Table 1). The total SOC concentrations were highest in the upper 0–5 cm soil layer and then decreased with increasing soil layer (Table 1). Afforestation with *R. pseudoacacia* and the mixed-species forest had significantly higher total organic C concentrations and stocks compared with cropland and *P. tabuliformis*. Furthermore, afforestation with *R. pseudoacacia*, and the mixed-species forest resulted in significant increases in total organic C concentrations in the 5–10 cm soil layer compared with cropland. Generally, no significant differences in total organic C concentrations were observed among the afforestation treatments with *R. pseudoacacia*, *P. tabuliformis*, and mixed stand in the 10–20 cm soil layer (Table 1). Soil organic C stocks showed similar patterns as seen for total organic C concentrations.

3.2. Labile organic C fractions

Results for the 0–5 cm soil layer are described in the following paragraphs whereas results for the 5–10 cm and 10–20 cm layers are not repeated as they generally showed patterns similar to the 0–5 cm layer or no significant differences.

POC accounted for the largest proportion of labile C fractions (Fig. 1a). The three afforestation treatments significantly increased POC in the 0–5 cm soil layer compared with cropland. In comparison with the cropland alone, POC extracted from the *R. pseudoacacia*, *P. tabuliformis*, and the mixed stand was significantly increased by 57.4%, 22.2%, and 44.4% in the 0–5 cm, respectively. KMnO₄-C concentrations showed a pattern similar to POC among all land use types. KMnO₄-C in the 0–5 cm soil layer was highest for *R. pseudoacacia* (Fig. 1b). Afforestation with *R. pseudoacacia*, *P. tabuliformis*, and the mixed stand resulted in higher levels of KMnO₄-C compared with cropland, with concentrations in the 0–5 cm soil layer that were 2.3, 1.5, and 2.1 times higher than observed for cropland. MBC in the 0–5 cm layer was different among the three afforestation areas and cropland, with a significantly lower value observed for cropland than for the three afforestation areas ($P < 0.01$, Fig. 2a). MBC comprised the smallest proportion (1.6–3.1%) of labile C fractions. The three afforestation areas had significantly higher DOC compared with cropland ($P < 0.01$, Fig. 2b). The proportion of DOC varied from 3.1 to 5.2% of labile C fractions. The impacts of different afforestation compositions on the proportion of DOC were similar to the other organic C fractions described above, with the highest concentration observed for *R. pseudoacacia* and the lowest concentration observed for cropland.

3.3. Carbon management index

There were significant differences between the three afforestation areas and cropland in each soil layer for CMI, CPI, and LI (Table 2). Changes in CMI under the three afforestation treatments and cropland increased in the order of cropland < *P. tabuliformis* < mixed stand < *R. pseudoacacia*, with values ranging from 100 to 244 in the

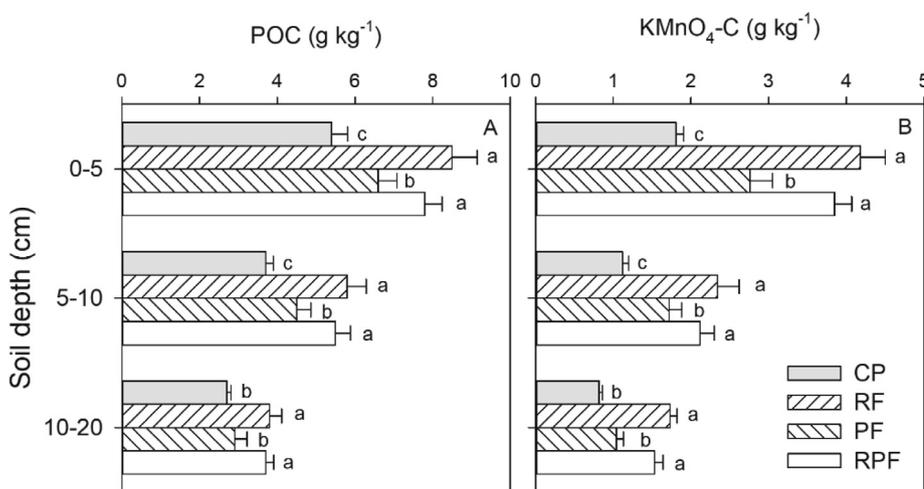


Fig. 1. Soil particulate organic carbon content (POC) (a) and potassium permanganate-oxidizable carbon (KMnO₄-C) (b) in the 0–5, 5–10, and 10–20 cm soil layers under different land-use types. Error bars represent standard deviations (n = 6). Different lowercase letters in the same soil layer indicate a significant difference at P < 0.05 among the different land-use types. CP: cropland; RF: *Robinia pseudoacacia* forest; PF: *Pinus tabuliformis* forest; RPF: *Robinia pseudoacacia* and *Pinus tabuliformis* forest.

0–5 cm soil layer. Generally, afforestation with *R. pseudoacacia* resulted in the highest CMI value in each soil layer, indicating more labile C fraction.

4. Discussion

4.1. Effect of afforestation on soil total organic SOC

The distribution of total SOC concentration and stocks was affected by land use types (Zhao et al., 2014; Tong et al., 2016; Chen et al., 2018). In this study, total SOC concentration and stocks decreased with soil depth and were usually highest in the surface soil. Given that topsoil is the primary provider of soil nutrients and water for uptake by plant roots (Chen et al., 2016), this observation is not surprising. It was also found that afforestation led to a significant increase in total organic C, supporting our hypothesis. The average total SOC concentrations and stocks decreased in the order of *R. pseudoacacia* > *P. tabuliformis* > mixed stand > cropland. On the one hand, these differences could rationally be ascribed to diverse amounts of understory vegetation biomass returned to the soil (Wang et al., 2007; Kooch et al., 2019). For example, the *R. pseudoacacia* sites had the greater understory vegetation biomass amounts with the greater soil total organic C pool, while the *P. tabuliformis* sites had the lower understory vegetation biomass amounts, and consequently the lower total SOC (Fig. 3). On the other hand, deciduous broadleaf trees produce litter which is decomposed more rapidly than that of coniferous tree species (Wang et al., 2007; Sreekanth et al., 2013; Chen et al., 2018). Our study suggested that evergreen species utilize a more conservative carbon-use strategy than

Table 2

Changes in carbon management index (CMI) at different soil layers under three afforestation treatments. Values represent mean ± SD for sample size of n = 6. Different lowercase letters in the same soil layer indicate a significant difference at P < 0.05 among the different land-use types. RF: *Robinia pseudoacacia* forest; PF: *Pinus tabuliformis* forest; RPF: *Robinia pseudoacacia* and *Pinus tabuliformis* forest. CPI: carbon pool index; LI: lability index.

	Soil layers (cm)	Cropland	RF	PF	RPF
CMI	0–5	100b	244(18)a	229(17)a	242(26)a
	5–10	100b	272(22)a	225(12)a	243(19)a
	10–20	100b	310(24)a	275(20)a	305(22)a
CPI	0–5	1.00c	1.74(0.1)a	1.33(0.1)b	1.23(0.1)b
	5–10	1.00c	1.42(0.1)a	1.13(0.1)bc	1.36(0.1)ab
	10–20	1.00b	1.43(0.1)a	1.24(0.1)ab	1.16(0.1)b
LI	0–5	1.00c	1.40(0.1)b	1.72(0.1)ab	1.96(0.1)a
	5–10	1.00b	1.92(0.2)a	1.98(0.2)a	1.79(0.1)a
	10–20	1.00b	2.16(0.1)a	2.20(0.2)a	2.60(0.2)a

deciduous species. In addition, in line with our second hypothesis, soil C was significantly greater in the presence of N₂-fixing *R. pseudoacacia* than with *P. tabuliformis*. This was indicated by greater total SOC and labile organic C fractions in the soil surface layer (Table 1, Figs. 1, 2). It has been postulated that enhanced soil N availability in the N₂-fixing forest may increase total SOC and labile organic C fractions compared with the non-N₂-fixing forest (Butterbach-Bahl and Dannenmann, 2012).

The observation that both total SOC and labile organic C fractions in

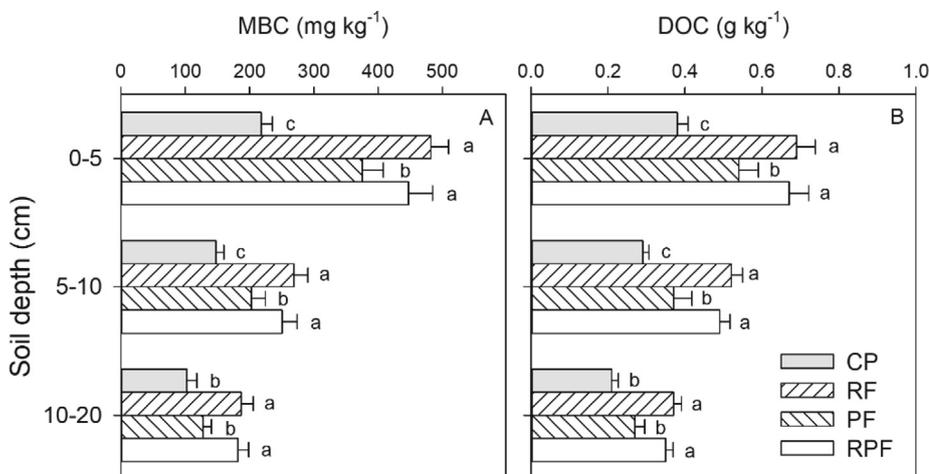


Fig. 2. Soil microbial biomass carbon content (MBC) (a) and dissolved organic carbon (DOC) (b) in the 0–5, 5–10, and 10–20 cm soil layers under different land-use types. Error bars represent standard deviations (n = 6). Different lowercase letters in the same soil layer indicate a significant difference at P < 0.05 among the different land-use types. CP: cropland; RF: *Robinia pseudoacacia* forest; PF: *Pinus tabuliformis* forest; RPF: *Robinia pseudoacacia* and *Pinus tabuliformis* forest.

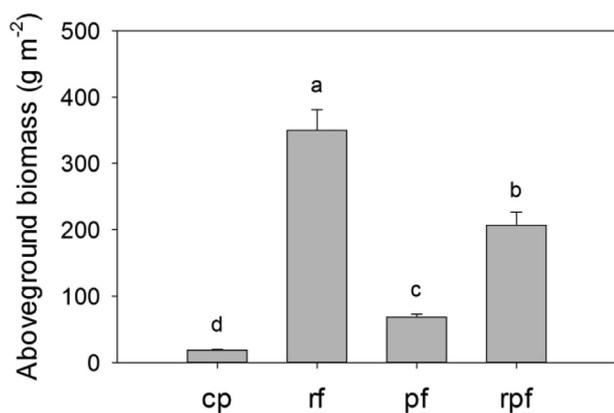


Fig. 3. Understory vegetation biomass under different land-use types in August 2018. Error bars represent standard deviations ($n = 6$). Significant differences among land-use types are indicated by different lowercase letters ($P < 0.05$). CP: cropland; RF: *Robinia pseudoacacia* forest; PF: *Pinus tabuliformis* forest; RPF: *Robinia pseudoacacia* and *Pinus tabuliformis* forest.

the mixed stand were greater than in cropland but similar to the *R. pseudoacacia* stand indicated a non-synergistic effect of *R. pseudoacacia* intercropped with *P. tabuliformis* on SOC accumulation. Indeed, there is considerable potential for intercropping N_2 -fixing species with woody non- N_2 -fixing species to enhance SOC accumulation following afforestation (Yuan et al., 2016; Du et al., 2019). This may be explained by several factors, such as non- N_2 -fixing species and soil texture (Wang and Xue, 1994). Therefore, the potential of intercropping with another locally and ecologically-important N_2 -fixing species (*C. korshinskii*) with non- N_2 -fixing species (*S. cheilophila* or *P. orientalis*) should be further studied.

4.2. Effect of afforestation on soil labile organic C fractions

Soil labile organic C fractions differed among different afforestation treatments and soil depths. Afforestation with different species compositions on the Loess Plateau positively increased POC, $KMnO_4$ -C, MBC, and DOC concentrations in the 20-cm soil profile, compared with cropland (Figs. 1, 2). It has been widely accepted that afforestation markedly increases soil labile organic C fractions (Deng et al., 2013; Sierra et al., 2013) directly or indirectly, which is consistent with our findings. There are several reasons for this observation. First, understory vegetation litter inputs may directly contribute to the soil labile organic C fractions (Fig. 3). Second, the positive microbial activities in the afforestation areas could increase the conversion of plant litter organic matter into labile forms of organic C (Poirier et al., 2013; Whalen et al., 2014). This result suggests that afforestation not only increased total SOC but also resulted in more soil labile organic C fractions (Table 1, Figs. 1, 2).

The CMI provides an indication of changes in the labile C dynamics of soil systems (Kalambukattu et al., 2013). Blair et al. (1995) reported that the actual CMI values make no sense, but relative differences in CMI values reflect how different management practices impact soil systems. In our study, CMI was highest under the three afforestation treatments and significantly larger than for cropland. *R. pseudoacacia* resulted in a higher CMI than *P. tabuliformis* and the mixed stand (Table 2). Our results agree with those from a long-term investigation by Blair et al. (2006). Tirol-Padre and Ladha (2004) explained that CMI variations observed under different soil management practices could be attributed to changes in organic matter quality, thus affecting the lability of C to $KMnO_4$ oxidation. The $KMnO_4$ -C concentration was also significantly affected by the forest expansion onto cropland, with the highest value for *R. pseudoacacia* and the lowest value for cropland (Fig. 1b). Similar concentrations of soil $KMnO_4$ -C have also been reported under long-term vegetation restoration (Zhao et al., 2014,

2015). Since CMI values indicate if an ecological system is in decline or being rehabilitated (Blair et al., 1995), the CMI values for cropland indicated deterioration, while afforestation fostered rehabilitation, especially for the *R. pseudoacacia* forest. Therefore, afforestation with *R. pseudoacacia* led to increases in $KMnO_4$ -C as a percentage of total SOC and consequently increased CMI values compared with the agricultural management practices associated with crop production. These indices explain why afforestation not only increases soil total organic C, but also results in greater soil labile organic C fractions.

In addition, the POC level was substantially lower in cropland than in the three afforestation areas (Fig. 1a). POC contains decomposing plant litters with short-term turnover periods, which is consistent with the characteristics of total organic C concentrations (Feller and Beare, 1997). The three afforestation treatments resulted in the highest levels of POC because they produced the most plant residues. MBC and DOC are produced by the decomposition of soil organic matter mainly driven by soil microbes (Marschner and Bredow, 2002). Although they account for only a small proportion of total organic C (generally 1.6–3.1% for MBC and 3.1–5.2% for DOC) in the forest soils of our study, these two quantities are considered good indicators of the soil nutrient cycle (Moharana et al., 2012; Benbi et al., 2015). Significant increases in MBC and DOC were observed after afforestation, suggesting that forest expansion onto cropland had positive effects on the activity of microorganisms, probably by providing an available source of C substrate (Yang et al., 2012).

5. Conclusions

Our results clearly indicated that afforestation significantly increased total SOC concentrations, organic C stocks, labile organic C fractions (POC, $KMnO_4$ -C, MBC, DOC), and CMI compared with cropland in the Loess Plateau. The results also showed that afforestation with *R. pseudoacacia* resulted in greater carbon benefits than the other two afforestation treatments. Moreover, afforestation with *R. pseudoacacia* produced more total SOC and labile organic C fractions than either *P. tabuliformis*, the mixed stand, or cropland, mainly due to its ability to fix N_2 . Although afforestation increased total SOC, it also resulted in more soil labile organic C fractions in China's Loess Plateau.

CRedit authorship contribution statement

Qingyin Zhang: Conceptualization, Data curation, Software, Visualization, Writing - original draft. **Xiaoxu Jia:** Formal analysis, Funding acquisition, Project administration, Resources, Supervision, Validation. **Xiaorong Wei:** Data curation, Writing - review & editing. **Mingan Shao:** Funding acquisition. **Tongchuan Li:** Investigation, Methodology. **Qiang Yu:** Conceptualization, Writing - review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

This research was supported by the Fundamental Research Funds for the Central Universities (2452018088), the National Natural Science Foundation of China (41530854 and 41390461), the International Partnership Program of the Chinese Academy of Sciences (161461KYSB20170013). We thank the editors and reviewers for the constructive comments on the manuscript.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://>

doi.org/10.1016/j.foreco.2020.117911.

References

- Bargali, K., Manral, V., Padalia, K., Bargali, S.S., Upadhyay, V.P., 2018. Effect of vegetation type and season on microbial biomass carbon in Central Himalayan forest soils, India. *Catena* 171, 125–135.
- Batjes, N.H., 1996. Total carbon and nitrogen in the soils of the world. *Eur. J. Soil Sci.* 47, 151–163.
- Benbi, D.K., Brar, K., Toor, A.S., Singh, P., 2015. Total and labile pools of soil organic carbon in cultivated and undisturbed soils in northern India. *Geoderma* 237, 149–158.
- Blair, N., Faulkner, R.D., Till, A.R., Poulton, P.R., 2006. Long-term management impacts on soil C, N and physical fertility Part I: Broadbalk experiment. *Soil Tillage Res.* 91, 30–38.
- Blair, G.J., Lefroy, R.D.B., Lisle, L., 1995. Soil carbon fractions based on their degree of oxidation, and the development of a carbon management index for agriculture systems. *Aust. J. Agric. Res.* 46, 1459–1466.
- Butterbach-Bahl, K., Dannenmann, M., 2012. Soil carbon and nitrogen interactions and biosphere-atmosphere exchange of methane and nitrous oxide. In: Lal, R., Lorenz, K., Hüttl, R.F., Schneider, B.U., von Braun, J. (Eds.), *Re-carbonization of the Biosphere-Ecosystems and the Global Carbon Cycle*. Springer, Dordrecht.
- Cambardella, C.A., Elliott, E.T., 1992. Particulate soil organic-matter changes across a grassland cultivation sequence. *Soil Sci. Soc. Am. J.* 56, 777–783.
- Canadell, J.G., Raupach, M.P., 2008. Managing forests for climate change mitigation. *Science* 320, 1456–1457.
- Chen, L.L., Mu, X.M., Yuan, Z.Y., Deng, Q., Chen, Y.L., Yuan, L.Y., Ryan, L.T., Kallenbach, R.L., 2016. Soil nutrients and water affect the age-related fine root biomass but not production in two plantation forests on the Loess Plateau, China. *J. Arid Environ.* 135, 173–180.
- Chen, L.L., Deng, Q., Yuan, Z.Y., Mu, X.M., Kallenbach, R.L., 2018. Age-related C:N: P stoichiometry in two plantation forests in the Loess Plateau of China. *Ecol. Eng.* 120, 14–22.
- Chen, Y.Q., Yu, S.Q., Liu, S.P., Wang, X.L., Zhang, Y., Liu, T., Zhou, L.X., Zhang, W.X., Fu, S.L., 2017. Reforestation makes a minor contribution to soil carbon accumulation in the short term: evidence from four subtropical plantations. *For. Ecol. Manage.* 384, 400–405.
- Deng, L., Wang, K.B., Chen, M.L., Shangguan, Z.P., Sweeney, S., 2013. Soil organic carbon storage capacity positively related to forest succession on the Loess Plateau, China. *Catena* 110, 1–7.
- Deng, L., Liu, G.B., Shangguan, Z.P., 2014. Land use conversion and changing soil carbon stocks in China's 'Grain-for-Green' Program: a synthesis. *Glob. Change Biol.* 20, 3544–3556.
- Deng, L., Wang, G.L., Liu, G.B., Shangguan, Z.P., 2016. Effects of age and land-use changes on soil carbon and nitrogen sequestrations following cropland abandonment on the loess plateau, china. *Ecol. Eng.* 90, 105–112.
- Deng, L., Liu, S.G., Dong, G.K., Peng, C.H., Sweeney, S., Shangguan, Z.P., 2017. Past and future carbon sequestration benefits of China's grain for green program. *Glob. Environ. Chang.* 47, 13–20.
- Du, B.G., Pang, J.Z., Hu, B., Allen, D.E., Bell, T.L., Pfautsch, S., Netzer, F., Dannenmann, M., Zhang, S.X., Rennenberg, H., 2019. N₂-fixing black locust intercropping improves ecosystem nutrition at the vulnerable semi-arid Loess Plateau region, China. *Sci. Total Environ.* 688, 333–345.
- Feller, C., Beare, M.H., 1997. Physical control of soil organic matter dynamics in the tropics. *Geoderma* 79, 69–116.
- Foley, J.A., DeFries, R., Asner, G.P., Barford, C., Bonan, G., Carpenter, S.R., Chapin, F.S., Coe, M.T., Daily, G.C., Gibbs, H.K., Helkowski, J.H., Holloway, T., Howard, E.A., Kucharik, C.J., Monfreda, C., Patz, J.A., Prentice, I.C., Ramankutty, N., Snyder, P.K., 2005. Global consequences of land use. *Science* 309, 570–574.
- Forrester, D.I., Bauhus, J., Cowie, A.L., Vanclay, J.K., 2006. Mixed-species plantations of *Eucalyptus* with nitrogen-fixing trees: a review. *For. Ecol. Manage.* 233, 211–230.
- Guo, L.B., Gifford, R.M., 2002. Soil carbon stocks and land use change: a meta analysis. *Glob. Change Biol.* 8, 345–360.
- Houghton, R.A., Hackler, J.L., 1999. Emissions of carbon from forestry and land-use change in tropical Asia. *Global Change Biol.* 5, 481–492.
- Jones, D.L., Willett, V.B., 2006. Experimental evaluation of methods to quantify dissolved organic nitrogen (DON) and dissolved organic carbon (DOC) in soil. *Soil Biol. Biochem.* 38, 991–999.
- Kalambukattu, J.G., Singh, R., Patra, A.K., Arunkumar, K., 2013. Soil carbon pools and carbon management index under different land use systems in the Central Himalayan region. *Acta Agric. Scan. Sect. B Soil Plant Sci.* 63, 200–205.
- Kooch, Y., Sanji, R., Tabari, M., 2019. The effect of vegetation change in c and n contents in litter and soil organic fractions of a northern Iran temperate forest. *Catena* 178, 32–39.
- Laganière, J., Angers, D.A., Paré, D., 2010. Carbon accumulation in agricultural soils after afforestation: a meta-analysis. *Global Change Biol.* 16, 439–453.
- Lal, R., 2004. Soil carbon sequestration impacts on global climate change and food security. *Science* 304, 1623–1627.
- Lal, R., 2005. Forest soils and carbon sequestration. *For. Ecol. Manage.* 220, 242–258.
- Li, D.J., Niu, S.L., Luo, Y.Q., 2012. Global patterns of the dynamics of soil carbon and nitrogen stocks following afforestation: a meta analysis. *New Phytol.* 195, 172–181.
- Li, D.J., Wen, L., Yang, L.Q., Luo, P., Xiao, K.C., Chen, H., Zhang, W., He, X.Y., Chen, H.S., Wang, K.L., 2018. Dynamics of soil organic carbon and nitrogen following agricultural abandonment in a karst region. *J. Geophys. Res. Biogeosci.* 122, 230–242.
- Marschner, B., Bredow, A., 2002. Temperature effects on release and ecologically relevant properties of dissolved organic carbon in sterilised and biologically active soil samples. *Soil Biol. Biochem.* 34, 459–466.
- Martens, D.A., Reedy, T.E., Lewis, D.T., 2003. Soil organic carbon content and composition of 130-year crop, pasture and forest land-use managements. *Global Change Biol.* 10, 65–78.
- Metz, B., Davidson, O.R., Bosch, P.R., Dave, R., Meyer, L.A., 2007. Contribution of Working Group III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, UK and New York, NY, USA.
- Miles, L., Kapos, V., 2008. Reducing greenhouse gas emissions from deforestation and forest degradation: global land-use implications. *Science* 320, 1454–1455.
- Moharana, P.C., Sharma, B.M., Biswas, D.R., Dwivedi, B.S., Singh, R.V., 2012. Long-term effect of nutrient management on soil fertility and soil organic carbon pools under a 6-year-old pearl millet-wheat cropping system in an Inceptisol of subtropical India. *Field Crop Res.* 136, 32–41.
- Nelson, D.W., Sommers, L.E., 1982. Total carbon and organic matter. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.), *Methods of Soil Analysis. Part 2: Chemical and Microbiological Properties*. American Society of Agronomy, Madison, Wisconsin, pp. 539–557.
- Pang, D.B., Cui, M., Liu, Y.G., Wang, G.Z., Cao, J.H., Wang, X.R., Dan, X.Q., Zhou, J.X., 2019. Responses of soil labile organic carbon fractions and stocks to different vegetation restoration strategies in degraded karst ecosystems of southwest China. *Ecol. Eng.* 138, 391–402.
- Paul, K.I., Polglase, P.J., Nyakuengama, J.G., Khanna, P.K., 2002. Change in soil carbon following afforestation. *Forest Ecol. Manage.* 168, 241–257.
- Poirier, V., Angers, D.A., Rochette, P., Whalen, J.K., 2013. Initial soil organic carbon concentration influences the short-term retention of crop-residue carbon in the fine fraction of a heavy clay soil. *Biol. Fertil. Soils* 49, 527–535.
- Post, W.M., Kwon, K.C., 2000. Soil carbon sequestration and land-use change: processes and potential. *Global Change Biol.* 6, 317–327.
- Richter, D.D., Markewitz, D., Trumbore, S.E., Wells, C.G., 1999. Rapid accumulation and turnover of soil carbon in a re-establishing forest. *Nature* 400, 56–58.
- Rytter, R.M., 2016. Afforestation of former agricultural land with Salicaceae species-initial effects on soil organic carbon, mineral nutrients, C: N and pH. *For. Ecol. Manage.* 363, 21–30.
- Sayyad, E., Hosseini, S.M., Mokhtari, J., Mahdavi, R., Jalali, S.G., Akbarinia, M., Tabari, M., 2006. Comparison of growth, nutrition and soil properties of pure and mixes stands of *Populus deltoides* and *Alnus subcordata*. *Silva Fennica* 40, 27–35.
- Shang, W., Wu, X.D., Zhao, L., Yue, G.Y., Zhao, Y.H., Qiao, Y.P., Li, Y.Q., 2018. Seasonal variations in labile soil organic matter fractions in permafrost soils with different vegetation types in the central Qinghai-Tibet Plateau. *Catena* 137, 670–678.
- Sierra, M., Martínez, F.J., Verde, R., Martín, F.J., Macías, F., 2013. Soil-carbon sequestration and soil-carbon fractions, comparison between poplar plantations and corn crops in south-eastern Spain. *Soil Tillage Res.* 130, 1–6.
- Six, J., Feller, C., Denef, K., Ogle, S.M., Moraes, S.C.J., Albrecht, A., 2002. Soil organic matter, biota and aggregation in temperate and tropical soils-effects of no-tillage. *Agronomie* 22, 755–775.
- Sreekanth, N.P., Shanthi, P.V., Padmakumar, B., Thomas, A.P., 2013. Soil carbon alterations of selected forest types as an environmental feedback to climate change. *Int. J. Environ. Sci. Technol.* 3, 1516–1530.
- Tirol-Padre, A., Ladha, J.K., 2004. Assessing the reliability of permanganate-oxidizable carbon as an index of soil labile carbon. *Soil Sci. Soc. Am. J.* 68, 969–978.
- Tong, X.G., Han, X.H., Wu, F.Q., Zhao, F.Z., Ren, C.J., Li, J., 2016. Change in carbon storage in soil physical fractions after afforestation of former arable land. *Soil Sci. Soc. Am. J.* 80, 1098–1106.
- Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil microbial biomass C. *Soil Biol. Biochem.* 19, 703–707.
- Vesterdal, L., Ritter, E., Gundersen, P., 2002. Change in soil organic carbon following afforestation of former arable land. *Forest Ecol. Manage.* 169, 137–147.
- Wang, X., Jia, Y., Li, X., Long, R., Ma, Q., Li, F., Song, Y., 2009. Effects of land use on soil total and light fraction organic, and microbial biomass C and N in a semi-arid ecosystem of northwest China. *Geoderma* 153, 285–290.
- Wang, Q.K., Wang, S.L., Fan, B., Yu, X.J., 2007. Litter production, leaf litter decomposition and nutrient return in *Cunninghamia lanceolata* plantations in south China: effect of planting conifers with broadleaved species. *Plant Soil* 297, 201–211.
- Wang, Z.L., Xue, Z.D., 1994. Ecological dividing of growing fitted area of *Robinia pseudoacacia* forest in Loess Plateau. *Res. Soil Water Conserv.* 1, 43–47.
- Whalen, J.K., Gul, S., Poirier, V., Yanni, S.F., Simpson, M.J., Clemente, J.S., Feng, X., Grayston, S.J., Barker, J., Gregorich, E.G., Angers, D.A., Rochette, P., Janzen, H.H., 2014. Transforming plant carbon into soil carbon: process-level controls on carbon sequestration. *Can. J. Plant Sci.* 94, 1065–1073.
- Xiao, Y., Huang, Z.G., Lu, X.G., 2015. Changes of soil labile organic carbon fractions and their relation to soil microbial characteristics in four typical wetlands of Sanjiang Plain, Northeast China. *Ecol. Eng.* 82, 381–389.
- Yang, X.Y., Ren, W.D., Sun, B.H., Zhang, S.L., 2012. Effects of contrasting soil management regimes on total and labile soil organic carbon fractions in a loess soil in China. *Geoderma* 177–178, 49–56.
- Yuan, Z.Q., Yu, K.L., Guan, X.K., Fang, C., Li, M., Shi, X.Y., 2016. *Medicago sativa* improves soil carbon sequestration following revegetation of degraded arable land in a semi-arid environment on the loess plateau, china. *Agr. Ecosyst. Environ.* 232, 93–100.
- Zhang, C., Liu, G.B., Xue, S., Sun, C.L., 2013. Soil organic carbon and total nitrogen storage as affected by land use in a small watershed of the Loess Plateau. *China. Eur. J. Soil Biol.* 54, 16–24.
- Zhang, Q.Y., Shao, M.A., Jia, X.X., Zhang, C.C., 2018. Understory vegetation and drought effects on soil aggregate stability and aggregate-associated carbon on Loess Plateau in

- China. Soil Sci. Soc. Am. J. 82, 106–114.
- Zhang, Q.Y., Shao, M.A., Jia, X.X., Wei, X.R., 2019. Changes in soil physical and chemical properties after short drought stress in semi-humid forests. *Geoderma* 338, 170–177.
- Zhao, Y.G., Liu, X.F., Wang, Z.L., Zhao, S.W., 2015. Soil organic carbon fractions and sequestration across a 150-yr secondary forest chronosequence on the loess plateau, china. *Catena* 133, 303–308.
- Zhao, F.Z., Yang, G.H., Han, X.H., Feng, Y.Z., Ren, G.X., 2014. Stratification of carbon fractions and carbon management index in deep soil affected by the grain-to-green program in China. *PLoS One* 9, e99657.