Received: 6 January 2022 | Accepted: 30 March 2022 | Published online: 7 July 2022

DOI: 10.1002/saj2.20422

Soil Science Society of America Journal

SPECIAL SECTION: MOLLISOLS DEGRADATION AND EVOLUTION UNDER DIFFERENT MANAGEMENT PRACTICES AND CLIMATE CHANGE

Long-term grassland restoration exerts stronger impacts on the vertical distribution of labile over recalcitrant organic carbon fractions in Mollisols

Xiangxiang Hao[1] | **Xiao-Zeng Han**[1] | **Na Li**[1,2] | **Wanying Lei**[1,2] | **Xu Chen**[1] | **Baoshan Xing**[3]

[1]Key Laboratory of Mollisols Agroecology, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Harbin 150081, PR China

[2]Univ. of the Chinese Academy of Sciences, Beijing 100039, China

[3]Stockbridge School of Agriculture, Univ. of Massachusetts, Amherst, MA 01003, USA

Correspondence
Na Li, Key Laboratory of Mollisols Agroecology, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Harbin 150081, PR China.
Email: bluena820@163.com, nal@iga.ac.cn

Assigned to Associate Editor Scott Chang.

Abstract

Land use change influences soil organic carbon (SOC) distribution in soil matrix. Conversion of cropland to grassland or forestland has the potential to sequester more carbon and influence the carbon allocation. Soil carbon sequestration happens within different physical and chemical fractions. This study aimed to investigate the vertical distribution of SOC fractions under 29-yr of cropland, grassland, and forestland of a typical Mollisol and to derive specific factors for SOC fraction changes associated with land use conversion. Soil samples within 100 cm soil depth were fractionated by density and humic methods. Biomass of litter residues and plant roots were also measured. Together with plant root biomass, the contents of total SOC and organic carbon (OC) fractions decreased along soil profile under three land uses. At each soil depth, total SOC and OC fractions followed the same order of grassland > forestland > cropland except for higher OC contents in heavy fraction, fulvic acid, and humin in cropland than those in forestland at the 0–10 cm layer. Compared with forestland and cropland, 29-yr grassland exerted stronger effects on the profile distribution of labile OC (water-soluble and light OC fractions) than recalcitrant OC (humic and heavy fractions), significances were observed at 0–80 cm layers for the labile OC and at 0–40 cm layers for the recalcitrant OC ($P < .05$). Larger root biomass and better soil structure in grassland might facilitate such changes. Grassland restoration, with greater C sequestration potential and larger liable OC contents, may help improve soil quality better than forestland and cropland in this Mollisol region.

Abbreviations: FAC, fulvic acid carbon; fLF, free light fraction; HA, humic acid; HAC, humic acid carbon; HF, heavy fraction; HFC, heavy fraction carbon; HM, humin fraction; HMC, humin carbon; HS, humic substance; LFC, light fraction carbon; OC, organic carbon; oLF, occluded light fraction; SOC, soil organic carbon; SOM, soil organic matter; WSOC, water soluble organic carbon.

1 | INTRODUCTION

Soil organic carbon (SOC) is an essential component of soil, which is assessed by the balance between C inputs through plant residues and the decomposition of soil organic matter (SOM) into the atmosphere. Land use patterns or soil managements have often been proposed as primary factors that control the SOC changes (David et al., 2009; Gregory et al., 2016; Guo & Gifford, 2002). When soils under native vegetation were converted to cropland, a depletion of the SOC would occur because of reduced OC inputs and enhanced decomposition of SOM after intensive physical disturbance (Poeplau et al., 2011). By contrast, large amounts of OC were captured in soil after grassland or forest restoration (Crème et al., 2018; Ding et al., 2017) in response to additional plant residues (H. Sheng et al., 2015), root biomass (Dietzel et al., 2017), and decreased SOC decomposition caused by less soil perturbation (Six et al., 2000). Land use change is acknowledged as a measure to mitigate climate change if the depleted cropland pools are refilled with carbon (Poeplau & Don, 2013). However, the conclusions about the effects of land use changes on the SOC fractions often vary under different environment conditions. In some cases, grassland restoration leads to a higher proportion of stabilized SOC than forests in temperate regions (Poeplau & Don, 2013; Yamashita et al., 2006). While in some other regions, perennial grassland restoration could highly increase the labile fractions of SOC than in forestland (Liu et al., 2014). The direction and magnitude of changes in SOC during land use changes are regulated by the amount and quality of aboveground biomass and root residue returned (Laganière et al., 2010) as well as the natural conditions lying in (Albaladejo et al., 2012).

Most studies investigating the SOC changes under different land use patterns have mainly limited to a depth of surface 20–30 cm, where the soils harbor the highest SOC contents (Ding et al., 2017; Hou et al., 2010; Lorenz & Lal, 2005). This involves a lack of knowledge about the response of different SOC fractions in subsoil to land use changes. The SOC in subsoil, accounting for more than half of total soil C stock (Jobbágy & Jackson, 2000), has been assumed to be old and recalcitrant (Salomé et al., 2010). However, a recent study found that SOC in subsoil was more susceptible to land use and climate changes than the topsoil (Luo et al., 2020). Moreover, there is growing evidence that SOC dynamics in the subsoil were also affected by land use changes (Poeplau & Don, 2013; M. Sheng et al., 2020), while comprehensively studies regarding the response of SOC and its fractions in subsoil to land use changes remain scarce.

Soil carbon fractionation protocol can help unveil subtle changes in specific compartments of soil matrix where the overall effect may be hidden when bulk soil is considered (Duval et al., 2018; M. Sheng et al., 2020). Different physical or chemical fractions of SOC represent different source and stability of SOC under different land use patterns (Li et al., 2020). For example, physically isolated density fractions could be used to determine the contributions of labile, predominantly plant-derived OC (i.e., light fraction carbon [LFC]), and stable, mostly microbially processed OC (i.e., heavy fraction carbon [HFC]) (John et al., 2005). The chemical fractions of SOC, based on their solubility in water (i.e., water soluble OC [WSOC]), aqueous acid, and alkaline media (i.e., humic substance [HS]), could be used to represent different decomposed stage of SOM (Guimarães et al., 2013). Recent studies reported that these OC fractions decreased in subsoil compared with surface soil under different land uses (Beniston et al., 2014; Hurisso et al., 2013; H. Sheng et al., 2015). The OC fractions with different availability and stability varied with land use regimes. Higher OC contents within macroaggregates (M. Sheng et al., 2020), humic fractions (Hurisso et al., 2013; Kotzé et al., 2016), organo-mineral complex (Hou et al., 2010), and microbial residues (Ding et al., 2017) were generally found in perennial grassland or forest soils compared with cropland. However, to date, only a few studies have focused on the effects of long-term contrasting land use patterns on the profile allocation of different OC fractions.

Previous studies argued that plant roots (including exudates) contributed to a large amount of OC inputs in subsoil (Dietzel et al., 2017; Kong & Six, 2010), which seemed to be more effective than shoot residue in building SOM (Sokol et al., 2019). Additional organic inputs were delivered into subsoils through root litter and exudates, bioturbation or mechanical incorporation of surface litters, and transport of dissolved OC from the surface layer (Rumpel & Kögel-Knabner, 2011). Many studies have demonstrated that the SOC dynamics and stabilization differed among grassland, forestland, and cropland, and these discrepancies may be caused by distinctly changes in root biomass, morphology, distribution, and turnover in these ecosystems (Guo et al., 2007; Six et al., 2000). A better understanding of the inter relationships of root inputs with SOC dynamics after long-term

Core Ideas

- Vertical distribution of SOC fractions under 29-yr land use regimes was studied.
- 29-yr grassland restoration accumulated more liable SOC fractions in 0–80 cm depth.
- Recalcitrant OC fractions at 0–40 cm depth were affected by land use changes.

land use patterns will be essential if whole-profile assessments of SOC changes are adopted to estimate C sequestration potential.

Mollisols in northeast China were degraded severely because of extensive cultivation and anthropogenic activities (Han & Li, 2018). The soil degradation inevitably threatened the national food security. Natural restorations, such as conversion cropland into grassland or forestland, were proven to be effective measures in improving carbon stock and soil quality in the surface layer in this region (Ding et al., 2017; Hou et al., 2010; M. Sheng et al., 2020). A deeper investigation of OC distribution in whole-soil profile after introduction of natural perennials and forest from cropping system needs to be carried out. In this study, a field experiment established in 1985 was used to observe SOC distribution in whole-soil profile under long-term contrasting land use patterns. The fields of the three land use types are closely adjacent and have totally the same edaphic condition, climate, and history of land use, which providing a scarce opportunity to compare the differences of SOC directly (Supplemental Figure S1). The SOC contents in bulk soil, as well as in density and humic fractions, were quantified in 0–100 cm soil depth, where 95% of plant roots were distributed. The aims of this study were to reveal the vertical distribution of SOC contents and OC fractions after long-term contrasting land use patterns of a typical Mollisol and to derive specific factors for carbon fraction changes associated with land use conditions. We hypothesized that conversion from cropland to grassland or forestland caused redistribution of SOC in soil matrix and in soil profile under long-term land use regimes because of continuous organic residues and root interplay effects.

2 | MATERIALS AND METHODS

2.1 | Study site and experimental design

The field experiment was established at the National Observation Station of Hailun Agroecology System, Heilongjiang Province, China (47°27′ N, 126°55′ E). The experimental site is located in the central region of typical Mollisols in northeastern China. The region has a temperate continental monsoon climate. The mean annual precipitation is 550 mm, with >80% of precipitation distributed from May through September. The mean annual temperature is 1.5 °C, with the lowest mean monthly temperature of −23 °C in January and the highest of 21 °C in July. The frost-free period is ∼120 d. The soil is classified as Udolls with a texture class of silty clay according to the USDA soil taxonomy (Soil Survey Staff, 2010) developed from loamy loess. The soil texture is 16.4% sand, 40.6% silt, and 43.0% clay. The soil at the start of the experiment in 1985 had a pH of 6.10 (soil/water = 1:2.5) and contained 31.5 g C kg^{-1} soil and 2.26 g N kg^{-1} soil (Table 1; Figure 1).

The study site was a grassland before reclamation ca. 1885. During the period of cultivation, the cropping system is interannual rotation between wheat (*Triticum aestivum* L.) and soybean [*Glycine max* (L.) Merrill.]. No fertilizer was applied from 1885 to 1944, and farm manure was applied as fertilizer from 1945 to 1964. Nitrogen fertilizer was used from 1965 to 1984. In 1985, a field experiment was setup by dividing the initial cropland into three contrasting land use sections: a naturally restored grassland (104 × 16 m); an artificial spruce forest (485 × 57 m), both without any fertilizer or soil tillage; and a cropland (120 × 5.6 m) under continuous soil tillage (Supplemental Figure S1). The dominant species in the grassland were annual bluegrass (*Poa annua* L.), common horsetail (*Equisetum arvense* L.), and frost grass (*Spodiopogon sibiricus* Trin.) when soil samples were taken. In the artificial spruce forest, spruce trees (*Picea koraiensis* Nakai) were planted in lines with a planting density of 2,100 trees ha^{-1}. The planting space was approximately 2.1 × 2.4 m. In 2013, the height and base diameter of the trees were approximately 4.5 m and 46 cm, respectively. There were only few short herbs plants and scattered mosses in the understory of the artificial spruce forest. The cropland was maintained as continuously tilled arable soil without any chemical fertilizer under a 3-yr wheat–maize (*Zea mays* L.)–soybean rotation since 1985. All crop straw was removed after harvest. The soil was ridged by rotary tillage to a depth of 20 cm after harvest in autumn each year. The selected soil properties in 0–20 cm at the beginning of the experiment and in the 29th year under different land use patterns are presented in Table 1.

2.2 | Soil sampling

The precipitation often peaks in the middle of August in this region. There is too much water in the deep soil, especially at 1.5-m soil depth, after crop harvest. Therefore, soil samples were collected at the end of July 2013, during which time the plant growth was the most vigorous, and the plant biomass also peaked under the three land use patterns. Sampling was carried out on a large field scale. In order to carry out statistical analysis, three sampling points with an area of 1 m^2 were randomly selected within each land use field representing three replicates. In the forestland, the sampling points were arranged on the diagonal point of four adjacent trees. A soil profile was dug to 1.0 m depth at each sample point. Before soil sampling, litter layers in grassland and forestland were carefully removed from the surface by hand. The cropland was maize during soil sampling. Soils were sampled at 0–10, 10–20, 20–40, 40–60, 60–80, and 80–100 cm depth. In all, 54 soil samples were collected (three treatments × six soil

TABLE 1 Selected soil properties in 0–20 cm in the initial soil and soils under different land use patterns after 29 yr

Soil properties	Initial soil (1985 year)	Grassland	Forestland	Cropland
Total N, g kg^{-1}	2.26 (0.08) b	2.81 (0.10) a	2.23 (0.07) b	2.16 (0.06) c
Total P, g kg^{-1}	0.84 (0.05) a	0.82 (0.03) a	0.79 (0.01) a	0.82 (0.03) a
Total K, g kg^{-1}	24.1(2.2) a	21.7 (1.5) a	20.5 (1.1) a	20.2 (1.9) a
Available N, g kg^{-1}	224 (3.9) b	249 (2.0) a	189 (1.6) c	163 (2.4) d
Olsen P, mg kg^{-1}	25.8(1.0) a	20.6 (1.7) b	18.2 (0.9) bc	17.3 (1.2) c
Extractable K, mg kg^{-1}	191 (15) a	169 (10) ab	156 (14) bc	144 (9) c
pH (H_2O)	6.10 (0.06) ab	6.40 (0.18) a	5.90 (0.03) b	6.01 (0.07) b
Sand (50–2,000 μm), %	16.4 (1.6) a	15.4 (1.0) a	15.3 (1.1) a	14.1 (1.7) a
Silt (2–50 μm), %	40.6 (0.8) a	42.2 (1.6) a	41.2 (1.3) a	41.6 (2.2) a
Clay (<2 μm), %	43.0 (2.4) a	42.4 (2.4) a	43.5 (1.7) a	44.4 (3.8) a

Note. Standard errors are reported in brackets ($n = 3$). Different letters indicate significant differences at $P < .05$ among initial soil in 1985 and land uses.

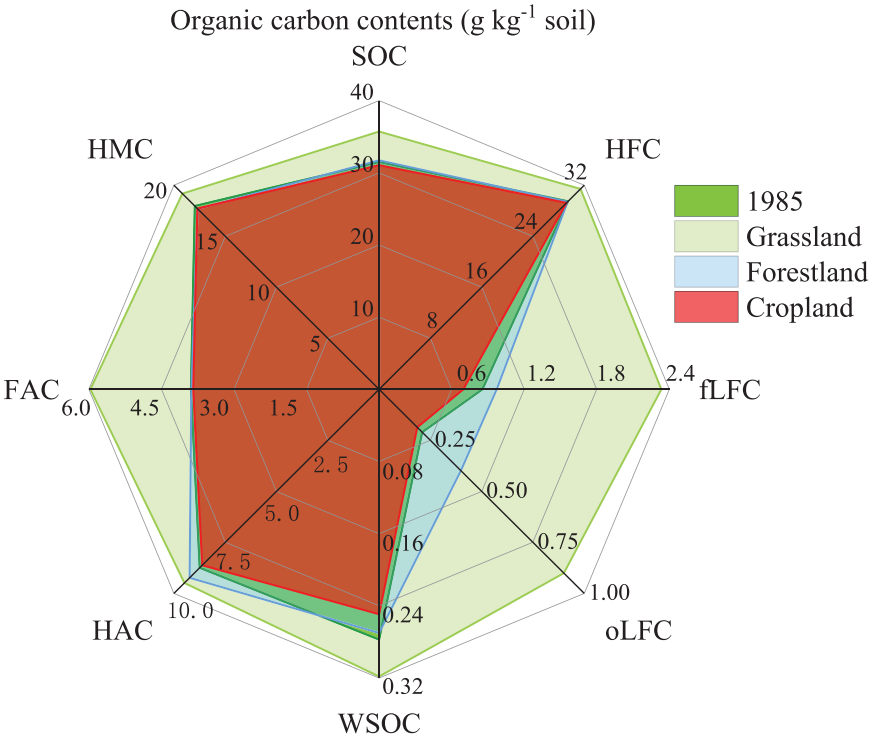

FIGURE 1 Organic carbon contents and carbon fractions of bulk surface soil (0–20 cm) in initial soil in 1985 and three field soils in 2013. Different letters indicate significant differences at $P < .05$ among land uses. SOC, soil organic carbon; fLFC, free light fraction carbon; oLFC, occluded light fraction carbon; HFC, heavy fraction carbon; WSOC, water-soluble organic carbon; HAC, humic acid carbon; FAC, fulvic acid carbon; HMC, humin carbon

layers × three replicates). In addition, five 100-cm^{-3} columns at each soil depth were sampled to determine soil bulk density after drying soil cores at 105 ˚C for 48 h. After removal of visible roots, segments, and fresh litter materials, soil samples were homogenized, sieved (<2 mm), and air dried prior to SOC fractionation and further analysis. The initial soils at 0–20 cm depth for the establishment of the experiment were collected with three replicates in 1985 then air dried and stored in dark brown glass bottles. The initial soil was analyzed together with the field soils sampled in 2013.

2.3 | Root and litter sampling

During soil sampling, root samples were collected simultaneously at each soil depth using 8-cm-diam. soil cores with five replicates. In the grassland, the sample points were selected randomly. In the forestland, the root sampling points were same as the soil samples, which were ~1.6 m away from the tree stems. In the cropland, samples of maize root were collected right over the crop after the aboveground parts were cut. The soil cores were stored at 4 ˚C until measurements. Cores were washed with tap water to remove adhering soil and

organic debris. All root samples were oven dried at 75 ˚C until constant weight. Root biomass in each depth was calculated using the following formula:

$$\text{Root biomass}\,(\text{tha}^{-1}) = \text{dry weight of roots per core}\,(\text{g}) \times 10^{-6}/\pi[8/2\,(\text{cm})]^2 \times 10^8$$

Litter material of grassland and forestland were collected from five randomly selected squares (50 × 50 cm), and all dead and detached vegetation was collected down to soil surface being careful to exclude mineral soil. For the cropland, the aboveground stubble and shoot residue left on the ground was collected after harvest from three random sites (1.5 m^2 in each site). Litters were placed in paper bags, transported to the laboratory, oven dried to a constant mass at 65 ˚C, and weighed.

2.4 | Laboratory analyses

Density fractions of SOC was obtained following Llorente et al. (2010). Briefly, 10-g soil sample (<2 mm) was placed in a 100-ml centrifuge tube with 50 ml of sodium iodide (NaI) solution ($d = 1.8$ g cm^{-3}). The tube was gently turned upside down by hand five times. After centrifugation at 2,000 × g for 30 min, the supernatant was passed through a 0.45-μm membrane filter into a millipore vacuum unit. The separation method was repeated three times. Then, particles on the membrane were collected, washed with deionized water, and considered as the free light fraction (fLF, $d < 1.8$ g cm^{-3}). The residue remaining in the tube was then added to 50 ml NaI. The tube was placed in an ice bath and sonicated at 300 J ml^{-1} for 15 min with a probe-type ultrasonic disintegrator. The floating material was the occluded light fraction (oLF, $d < 1.8$ g cm^{-3}) protected by soil aggregates, then recovered by centrifugation, filtered, and washed in the same way as the fLF. The leftover soil in the centrifuge tube was washed with distilled water until the water became clear and was used as the heavy fraction (HF, $d > 1.8$ g cm^{-3}). All fractions were dried at 50 ˚C, weighed, ground in a mortar and pestle, and analyzed for C.

Water-soluble organic carbon was extracted using hot water (Gregorich et al., 2003). In short, 5 g of air-dried soil samples (<2 mm) were placed in a 100 ml centrifuge tube with 50 ml deionized water, then the tubes were placed in a water bath at 80 ˚C for 16 h. The extracts were centrifuged at 2,000 × g for 20 min and filtered through 0.45-μm glass-fiber, microfiltration membrane. The total organic C concentrations of the filtered solution were measured using a liquid total organic carbon analyzer (Elementar Analysensysteme GmbH).

The extraction of humic substance was preformed according to the method described by Stevenson (1994). In short, 50 ml 0.1 mol L^{-1} NaOH and 0.1 mol L^{-1} $Na_4P_2O_7$ solution (50:50, v/v) was added into the remaining soils after the extraction of WSOC, then the tubes were placed in a water bath at 70 ˚C for 1 h. The supernatant solution was centrifuged at 2,000 × g for 15 min, then the humic extractable substances was collected. The humic extractable substance was acidified to pH 1.0 with 0.5 mol L^{-1} H_2SO_4, the precipitated fraction was acid-insoluble humic acid (HA), the solution was fulvic acid, the two fractions were separated via centrifugation at 3,500 × g for 15 min. The HA was redissolved with 0.05 mol L^{-1} NaOH. The residue soil was the humin fraction (HM). The C contents of humic extractable substances (HEC) and HA (HAC) was determined by the $K_2Cr_2O_7$ oxidation method. Carbon content of fulvic acid (FAC) and humin (HMC) were calculated using the following formulas:

$$\text{FAC} = \text{HEC} - \text{HAC}$$

$$\text{HMC} = \text{SOC} - \text{WSOC} - \text{HEC}$$

Total C and N contents in bulk soil and light fractions were determined using a Vario EL CHN elemental analyzer (Heraeus Elementar Vario EL). The Mollisols were free of carbonates, and thus, the total C content was equivalent to SOC content. Available N was determined according to Page et al. (1982). Total P was extracted by sodium carbonate and Olsen P by 0.5 M sodium bicarbonate and were detected by molybdenum-blue colorimetry (Olsen & Sommers, 1982). Total K was extracted by HF-$HClO_4$, and extractable K by 1 M ammonium acetate and were determined by flame photometry (Jones, 1973). Soil pH was determined in a 1:2.5 soil/water suspension. Soil particle size distribution was analyzed by the sieve–pipette method.

2.5 | Statistical analysis

Before analysis, all variables were checked for normal distribution (Kolmogorove Smirnov test) and homogeneity (Levene's test). Data of OC contents in soil density fractions were transformed to natural logarithms to achieve homogeneity. Each field was considered an experimental unit, therefore, replicated data were averaged by fields for analysis. One-way analysis of variance with Turkey honestly significant difference as a post hoc was used for means of SOC and OC fractions among the three field treatments at $P < .05$. Relationships of SOC and OC fractions with root biomass in 0–100 cm soil profile were examined using linear regression. Statistical analyses were performed using software package SPSS 19.0 for Windows.

3 | RESULTS

3.1 | Selected soil properties in surface 0–20 cm layer

Compared with the initial soil in 1985, the soil pH, total N, and available N contents increased, while the Olsen P decreased significantly after 29-yr natural grassland conversion ($P < .05$). The total P, total K, and extractable K decreased in grassland but without significance ($P > .05$). The 29-yr continuous cropland and spruce forest soils had obvious declines in soil pH, available N, Olsen P, and extractable K contents ($P < .05$) but had slight decreases in total N, total P, and total K contents in the surface 0–20 cm soil depth. The weight percentage of particle-size fractions was similar under 1985 initial soil and different land use patterns, with an average value of 43.2% for the <2 μm clay fraction, 41.4% for the 2–50 μm silt fraction, and 15.3% for the 50–2,000 μm sand fraction (Table 1). Compared with the initial soil in 1985, 29-yr continuous cropland and spruce forest did not cause obvious changes in the contents of SOC and OC fractions in the surface 0–20 cm depth (Figure 1) with the exceptions of significant decline of WSOC in cropland and increases of fLFC and oLFC in forestland ($P < .05$). Surprisingly, converting cropland to grassland significantly increased the contents of SOC and OC fractions ($P < .05$) with relatively higher increments of labile OC fractions (fLFC, oLFC) than the recalcitrant OC fractions. The fLFC and oLFC contents increased by 173–329% ($P < .05$) (Figure 1).

3.2 | Stocks of root and litter biomass

Root biomass showed decreases along soil profile in all land use patterns (Figure 2a). In all soil depths, root biomass was highest in grassland followed by spruce forestland and lowest in cropland except for slightly higher root biomass in forestland in 10–20 cm soil layer. The total root biomass within 100-cm soil profile was significantly higher in grassland (12.7 t ha^{-1}) than that in forestland (10.0 t ha^{-1}) and cropland (1.8 t ha^{-1}) (Figure 2a). Grassland also had significantly higher litter biomass (9.48 t ha^{-1}) than forestland (2.39 t ha^{-1}). The litter biomass in cropland was relatively low (0.92 t ha^{-1}) than that in grassland and forestland (Figure 2b).

3.3 | Distribution of SOC and carbon fractions in soil profile after land use changes

After 29-yr of land use changes, the contents of total SOC and OC fractions mostly followed the same trends as grassland > forestland > cropland along the whole-soil profile. Total SOC contents were significantly higher at 0–10 and 20–40 cm depth in grassland ($P < .05$) (Figure 3a), and the fLFC and oLFC contents were significantly higher at 0–80 cm depth in grassland soil than those in forest and cropland soils ($P < .05$) (Figure 3b,c). The WSOC and HAC contents were significantly higher in grassland and forestland soils than those in cropland in 0–80 and 0–60 cm soil depths, respectively (Figure 3e,g). The FAC contents were significantly higher in grassland than those in forestland and cropland in 0–60 cm soil depth ($P < .05$) (Figure 3f). The HFC and HMC in soil profile followed the same changes as total SOC under three land use patterns (Figure 3d,h).

In all soil layers, the HF and HM fractions contained a large proportion of OC in all field soils, accounting for 88.8–99.3 and 38.0–58.0% of the total SOC, respectively (Table 2). Similar to the variations of OC contents, the proportions of fLFC and oLFC fractions in total SOC both decreased with soil depth, and the proportions in 0–60 cm soil layers in grassland were higher than the other two land use patterns. However, for the proportion of HFC to SOC, differences among land use patterns existed along the 0–80 cm depths, and grassland had a significantly lower proportion of HFC than the other two land use patterns. Unlike the decreasing trends of WSOC contents in soil profiles, the proportions of WSOC in total SOC showed the opposite changes but no differences among land use patterns. The proportions of FAC in total SOC in forestland were lowest at 0–10 and 20–60 cm soil layers compared with those of grassland and cropland soils. The proportions of HAC and HMC in total SOC did not obviously change along soil profiles under all the three land uses (Table 2).

The stocks of SOC and its fractions were positively linearly correlated with root biomass in all 0–100 cm soil profiles, significant relationships were found in total SOC, WSOC, HA, and HF fractions ($P < .05$) (Figure 4).

4 | DISCUSSION

The aim of this study was to illustrate the OC allocation regimes along soil profile under 29-yr contrasting land use patterns of cropland, natural grassland, and spruce forestland in a Mollisols region. Only the surface 0–20 cm arable soil in 1985 used for the establishment of the experiment was available, hence it was impossible to determine the vertical changes of SOC after land use changes. By comparing the contents of SOC and OC fractions in the initial soil in 1985 with the surface soils under 29-yr of different land use patterns, we can evaluate the effects of land use regimes on soil properties in the surface soil where changes are most likely to occur. The Mollisols studied here have been under cropping for >100 yr. It was obvious that 29-yr continuous cropping did not cause significant changes of SOC, total N, and subfractions of SOC in the surface 0–20 cm soil (Table 1; Figure 2). The SOC

TABLE 2 Proportions of carbon fraction to total soil organic carbon in soil profile under different land uses

Soil depth	Land use types	Density fractions			Chemical fractions			
		fLF	oLF	HF	WSOC	FAC	HAC	HMC
cm		%						
0–10	Grassland	8.31 a	2.86 a	88.83 c	0.92 a	16.93 a	25.94 b	56.21 a
	Forestland	4.55 b	1.67 b	93.78 b	0.83 b	12.11 b	30.17 a	56.89 a
	Cropland	3.18 c	0.60 c	96.22 a	0.78 c	12.32 b	28.93 a	57.97 a
10–20	Grassland	5.00 a	2.13 a	92.87 c	0.88 a	18.30 a	30.22 a	50.60 b
	Forestland	1.79 b	0.83 b	97.38 b	0.89 a	13.64 b	30.95 a	54.53 ab
	Cropland	1.40 b	0.60 b	98.00 a	0.85 a	13.78 b	29.52 a	55.85 a
20–40	Grassland	1.73 a	0.94 a	97.33 c	0.99 a	16.83 ab	32.98 a	49.20 a
	Forestland	1.46 b	0.70 b	97.83 b	1.06 a	14.84 b	33.59 a	50.52 a
	Cropland	0.60 c	0.46 c	98.94 a	1.05 a	20.34 a	35.70 a	42.90 a
40–60	Grassland	1.05 a	0.64 a	98.31 b	1.12 a	22.98 a	33.95 a	41.96 ab
	Forestland	0.58 b	0.44 ab	98.98 a	1.17 a	16.12 b	33.95 a	48.76 a
	Cropland	0.48 b	0.26 b	99.26 a	1.16 a	23.45 a	37.33 a	38.05 b
60–80	Grassland	0.94 a	0.37 ab	98.69 b	1.28 a	21.54 a	28.94 a	48.24 a
	Forestland	0.45 b	0.46 a	99.09 a	1.30 a	20.79 a	29.03 a	48.88 a
	Cropland	0.49 b	0.22 b	99.29 a	1.25 a	23.94 a	33.03 a	41.78 a
80–100	Grassland	0.66 a	0.35 a	99.00 a	1.54 a	19.96 a	29.99 a	48.51 a
	Forestland	0.48 a	0.35 a	99.17 a	1.50 a	18.69 a	30.03 a	49.79 a
	Cropland	0.54 a	0.27 a	99.19 a	1.68 a	23.55 a	34.69 a	40.08 a

Note. fLFC, free light fraction carbon; oLFC, occluded light fraction carbon; HFC, heavy fraction carbon; WSOC, water-soluble organic carbon; HAC, humic acid carbon; FAC, fulvic acid carbon; HMC, humin carbon. Different letters indicate significant differences at $P < .05$ at each soil depth among land uses.

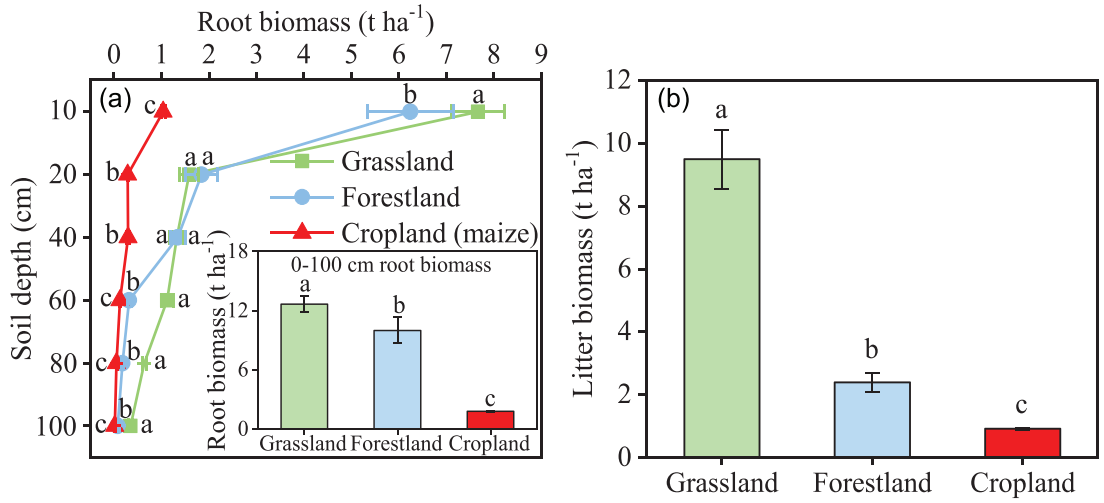

FIGURE 2 Stock of (a) root biomass and (b) surface litter biomass under different land uses. The litter biomass of cropland was the average of 1-yr value of three crops (wheat, soybean, and maize). Error bars indicate standard error of the mean for each depth ($n = 3$). Different letters indicate significant differences at $P < .05$ at each soil depth among land uses

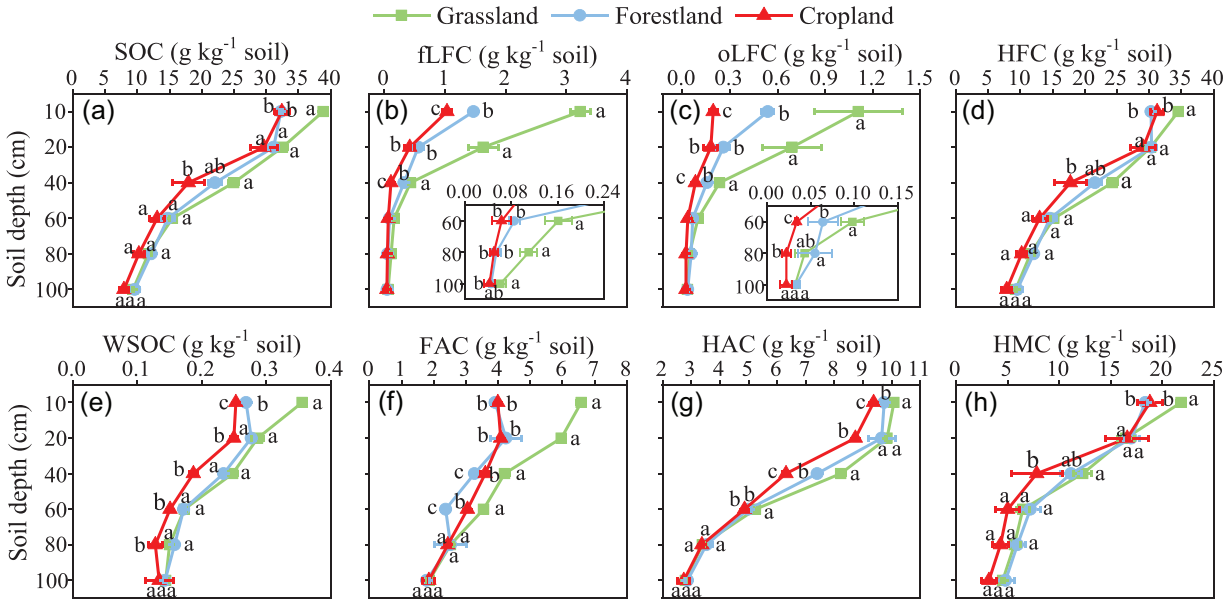

FIGURE 3 Contents of soil organic carbon (SOC) and its fractions along soil profile under different land use patterns. Error bars indicate standard error of the mean for each depth ($n = 3$). Different letters indicate significant differences at $P < .05$ at each soil depth among land uses. fLFC, free light fraction carbon; oLFC, occluded light fraction carbon; HFC, heavy fraction carbon; WSOC, water-soluble organic carbon; HAC, humic acid carbon; FAC, fulvic acid carbon; HMC, humin carbon

content decreased by 1.4% after 29-yr continuing cropping compared with the SOC content in 1985 (31.5 g kg^{-1}) with a reduction rate of 0.05% per year. This rate is relatively low when compared with the averaged value reported by Han and Li (2018) (0.5–2.6%) and Yang et al. (2004) (0.05–0.11%). Therefore, the cropland soil here was considered as the reference soil to further illustrate the vertical changes of SOC after 29-yr different land use patterns.

4.1 | Total SOC changes in soil profile under contrasting land use patterns

Fresh C inputs, as well as microbial biomass, were the highest near the surface (Fontaine et al., 2007) because most herbaceous plant roots were distributed in the top few centimeters of soil (Jackson et al., 1996). A higher SOC content was found in grassland than cropland and forestland in the

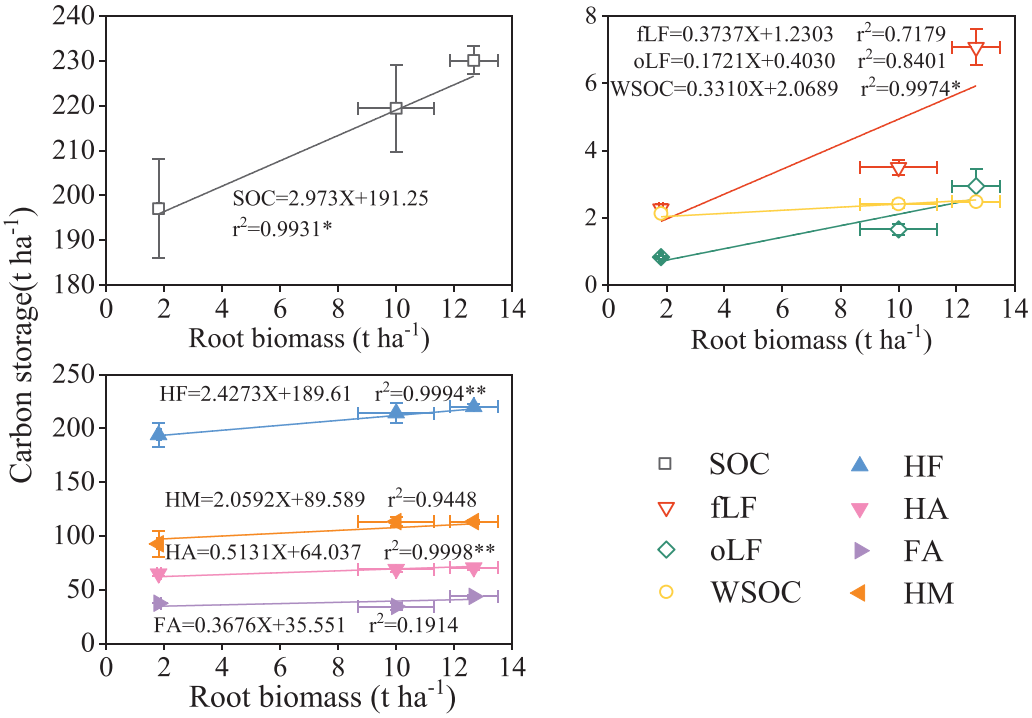

FIGURE 4 Relationships between root biomass and the stock of soil organic carbon (SOC) and its fractions in the 0–100 cm soil profile. fLFC, free light fraction carbon; oLFC, occluded light fraction carbon; HFC, heavy fraction carbon; WSOC, water-soluble organic carbon; HAC, humic acid carbon; FAC, fulvic acid carbon; HMC, humin carbon. *$P < .05$; **$P < .01$

surface soil because of large amounts of organic inputs into soil (Figure 3a). The SOC content in the 0–10 cm layer increased by 0.69% per year after 29-yr conversion from cropland to grassland, which was similar to 0.68% after 59-yr conversion from cropland to perennial grassland in the Chernozems of Russia (Kalinina et al., 2011) and 0.62 % after 40-yr conversion from cropland to perennial grassland in the U.S. Midwest (McLauchlan et al., 2006). However, the rate was still lower than the 1.03% found after 35-yr conversion from cultivated land to grassland in semiarid Mediterranean (Novara et al., 2014). The SOC content in forestland did not increase as significantly as that in grassland, showing that the SOC content increased but not significantly through the 0–100 cm soil profile after 29-yr spruce plantation compared with cropland (Figure 3a). Other studies also found little SOC change after afforestation of a cropland when pine species were used (Paul et al., 2002). The tree species has an important effect on the recovery of the SOC following afforestation because of the variability of OC inputs (De Deyn et al., 2008). Using a meta-analysis, Laganière et al. (2010) found that coniferous forest has a significantly lower capacity to accumulate SOC than broadleaf forest because of its smaller and shallower root systems. In addition, the decomposition rate of spruce root litter to soil humus was much slower than that of grass and crop plants (Guo et al., 2006; Laganière et al., 2010). These results could help explain the smaller SOC changes in forestland than those in grassland here (Figure 1). However, we cannot preclude that there will be further SOC accumulation with increasing forest age (Poeplau & Don, 2013). Moreover, soil tillage might also influence SOC changes. The cessation of tillage disturbance could affect soil aggregate-protected carbon, nutrients availability, and microclimate, thus preventing SOM decomposition (M. Sheng et al., 2020; Six et al., 2000).

The SOC in subsoil or deep soil is assumed to be more recalcitrant with longer residence time than that in the topsoil (Jenkinson et al., 2008). It is believed that subsoil has the potential to sequester more OC through higher root C inputs because of low C concentrations (Lorenz & Lal, 2005). However, our study showed that the increase of SOC in subsoil was not as straight forward (Figure 3a), because the decomposition of subsoil C with high residence time could be stimulated by addition of labile C (Fontaine et al., 2007). Some other studies found that the SOC in subsoil decreased after the conversion from cropland to grassland (Don et al., 2009; Poeplau & Don, 2013). This may be related to the differences in soil type and intrinsic soil texture. The silt and clay particles in Mollisols accounted for >70% of the whole soil (Table 1), which indicated that Mollisols had higher capacity to sequester carbon. The SOC above 20 cm was predominately controlled by root biomass, but root biomass was less of a factor below 20 cm (Dietzel et al., 2017). Hence, C dynamics in topsoil and subsoil may be controlled by different regulatory mechanisms (Salomé et al., 2010). Also, OC fractions may be

more sensitive than total SOC to unveil the long-term land use effects on C dynamics especially in soil profile.

4.2 | Effects of land use changes on OC fractions in soil profile

The subfractions of SOC with varied origin and availability represented different composition and stability of SOM (Li et al., 2020). Similar to the changes of SOC in bulk soil, OC fractions decreased with soil depth and followed nearly the same order of grassland > forestland > cropland in the whole 0–100 cm soil profile. Statistically, compared with the cropland, 29-yr grassland caused significant increases of labile OC contents (fLFC, oLFC, and WSOC) and FAC in 0–80 cm soil depths. The recalcitrant OC fractions, such as the HF, HA, and HM, increased in 0–40 cm soil depths ($P < .05$) (Figure 3).

The light and water-soluble fractions, mainly consisting of fresh plant-derived materials, represent labile SOC pools with rapid turnover rates. There was a steep gradient of labile OC contents and proportions with soil depth (Figure 3b,c,e; Table 2), especially in grassland soil, identifying aboveground litter being the major source of light OC fractions (Guimarães et al., 2013; M. Sheng et al., 2020). Our results partially supported the hypothesis that the OC fractions, especially the labile OC fractions, responded more sensitively than the total SOC because of land use changes. Temperate grassland allocated larger proportions of OC belowground than those observed in annual croplands (Jackson et al., 1996), which could provide abundant sources of labile OC fraction in soil (Figure 3b,c,e) (Guimarães et al., 2013). While in the spruce forest, the labile exudates from roots were more recalcitrant than those from perennials grass (Laganière et al., 2010). In addition, higher amounts of root residues and exudates in grassland and forestland could promote the formation of soil aggregates thereby increasing the oLFC that was physically protected within soil aggregates (Figure 4c) (Oades & Waters, 1991; Yamashita et al., 2006). The labile OC in subsoil or deeper soil layers consisted of materials either transferred by leaching and diffusion from topsoil or locally produced during extended biological processing of native SOM (Hassouna et al., 2010; Rumpel & Kögel-Knabner, 2011). The labile OC fractions along the soil profile in grassland were higher than those in forestland even having significantly higher WSOC and fLFC in 0–80 cm depth (Figure 4). Also, the proportions of WSOC in the surface 0–10 cm and fLFC and oLFC in 0–60 cm depths to total SOC were also higher in grassland than those in forestland and cropland soils (Table 2) as reported by John et al. (2005). Besides root effects, soil porosity and unique climatic conditions might be responsible for these differences. The content of WSOC was positively correlated with soil porosity (Mertens et al., 2007); higher porosity in grasslands could facilitate the leaching of WSOC from topsoil to deep soil along soil profile (Figure 3e). In addition, the study region was characterized by a simultaneously hot and rainy period from June to September. During this period, a strong leaching occurred in soil profile, allowing less complex and more water dissolve organic compounds to move along the soil profile more easily. Furthermore, cultivation practices in cropland were reported to decrease the amount of LFC in subsoil (H. Sheng et al., 2015) because of significantly higher root and litter decomposition rates when compared with grassland (Buyanovsky et al., 1987). Soil tillage in cropland may also help increase soil perturbation and OC decomposition thus facilitating the infiltration of more labile OC into deeper soil. Our results also showed that the LFC in cropland soil profile was 69.1 and 39.8% less than those in grassland and forestland soils, respectively.

It is worth noting that HA, HM, and HF account for more than 70–90% of SOC in each soil layer (Table 2), indicating that more OC in Mollisols was sequestrated in these recalcitrant fractions. These recalcitrant fractions were mainly mineral associated fractions with slower turnover rates and higher degrees of chemical protection; they sometimes responded slowly or were even unresponsive to soil managements. In this study, the effects of different land use patterns on these fractions mainly occurred in the surface 0–10 cm soil, indicating much closer organomineral associations and less vulnerability to mineralization with the increasing of clay content along soil profile (Hao et al., 2015; Rumpel & Kögel-Knabner, 2011). Soil management practices in cropland such as frequent soil tillage could stimulate microbial decomposition of recalcitrant OC fractions (Fontaine et al., 2007) and ultimately cause changes of SOC fractions in subsoils. In addition, deep plowing and clear cutting without fresh OC inputs could also accelerate old SOM decomposition in subsoil. Valentin et al. (2008) reported C accumulation was mainly attributed to recalcitrant C pools after land use change from arable to permanent grassland, as there were higher HFC and HMC contents in grassland.

4.3 | The underlying effects of plant litter and root biomass on OC fractions

Land use changes could alter plant production and root allocation and consequently cause redistribution of SOC in soil profiles (Guimarães et al., 2013). Beniston et al. (2014) reported that decreased inputs from roots were responsible for the reduced OC contents in bulk soil and OC fractions with soil depth. Dietzel et al. (2017) and H. Sheng et al. (2015) reported that plant root residues made substantial contributions to SOC accumulation, and the SOC contents correlated well with root biomass. This study also found that the contents of SOC and OC fractions exhibited nearly similar decreasing

patterns as root biomass along the soil depth (Figure 3), and the OC contents in bulk soil and OC fractions were positively correlated with root biomass (Figure 4). In addition, significantly higher surface litter retention and root biomass stocks in grassland explained the higher OC contents in grassland than those in forestland and cropland (Figures 2 and 3). An experiment established in Kansas, USA, also found higher litter and root biomass in grassland than in cropland soil; the root biomass at 0–40 cm depth in grassland was about six times that of adjacent wheat roots, and the SOC stock was ~1.4 times that of cropland (Beniston et al., 2014). The differences in SOC contents could be found in the upper 0–40 cm soil depth while the differences in labile OC fractions (fLFC, oLFC, WSOC, and FAC) were up to 80 cm soil depth under different land use regimes (Figure 3). Guo et al. (2007) reported that OC correlated positively with fine root length. And the fine root length was nine times higher in grassland than in pine forest soil. Furthermore, previous studies suggested that it can take ~40 yr or even a longer time to achieve new equilibrium after conversion of cropland to natural forest plantation (Guo & Gifford, 2002). However, for grassland, live fine roots seemed more degradable and available than those in forestland, which helped to explain the relatively higher OC contents in bulk soil and labile OC fractions in grassland than in forestland (Figure 3).

5 | CONCLUSIONS

This study evaluated the distribution of SOC and OC fractions in 0–100 cm soil profile of a typical Mollisol under different land use patterns. The 29-yr conversion of cropland to grassland and forestland caused relatively higher OC accumulation in soil profiles especially at the surface 0–40 cm depth, which highlighted the importance of reduced soil perturbation and continuous plant residue inputs for SOC accumulation. The labile OC fractions responded more sensitively to land use changes than the recalcitrant fractions. They were significantly higher in 0–80 cm soil depth in grassland than those in forestland and cropland because of more easily degraded plant residues and higher amounts of root biomass in the grassland. While land use effects on the recalcitrant OC fractions (HF, HA, and HM) were mainly observed in the 0–40 cm soil depths because of the high stability of these fractions. Significantly higher root biomass was found in grassland than those in forestland and cropland in the whole-soil profile, and the root biomass was highly correlated with the total SOC and OC fractions. Our results demonstrated that the vertical allocations of OC fractions changed obviously after long-term land use conversion. Long-term grassland restoration seemed to exert more effects on the labile OC than recalcitrant OC fractions through larger OC inputs from plant roots.

ACKNOWLEDGMENTS

Thank you for the anonymous reviewers for their constructive comments. This work was funded by the National Natural Science Foundation of China (41807094) Key Research Program of Frontier Sciences, CAS (ZDBS-LY-DQC017), the Strategic Priority Research Program of the CAS (XDA28010301) and the National Key Research and Development Program of China (2021YFD1500100, 2021YFD1500400). We thank all the staff at the National Observation Station of Hailun Agroecology System for their invaluable help with field management.

AUTHOR CONTRIBUTIONS

Xiangxiang Hao: Data curation; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Resources; Writing – original draft; Writing – review & editing. Xiao-Zeng Han: Conceptualization; Data curation; Investigation; Methodology; Resources; Writing – review & editing. Na Li: Data curation; Funding acquisition; Investigation; Methodology; Project administration; Resources; Supervision; Writing – original draft; Writing – review & editing. Wanying Lei: Data curation; Formal analysis; Software; Writing – original draft. Xu Chen: Investigation; Resources. Baoshan Xing: Data curation; Methodology; Writing – review & editing.

CONFLICT OF 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.

ORCID

Na Li https://orcid.org/0000-0002-5332-8216

REFERENCES

Albaladejo, J., Ortiz, R., Garcia-Franco, N., Navarro, A. R., Almagro, M., Pintado, J. G., & Martínez-Mena, M. (2012). Land use and climate change impacts on soil organic carbon stocks in semi-arid Spain. *Journal of Soils and Sediments*, *13*, 265–277. https://doi.org/10.1007/s11368-012-0617-7

Beniston, J. W., Dupont, S. T., Glover, J. D., Lal, R., & Dungait, J. A. J. (2014). Soil organic carbon dynamics 75 years after land-use change in perennial grassland and annual wheat agricultural systems. *Biogeochemistry*, *120*, 37–49. https://doi.org/10.1007/s10533-014-9980-3

Buyanovsky, G. A., Kucera, C. L., & Wagner, G. H. (1987). Comparative analyses of carbon dynamics in native and cultivated ecosystems. *Ecology*, *68*, 2023–2031. https://doi.org/10.2307/1939893

Crème, A., Rumpel, C., Le Roux, X., Romian, A., Lan, T., & Chabbi, A. (2018). Ley grassland under temperate climate had a legacy effect on soil organic matter quantity, biogeochemical signature and microbial activities. *Soil Biology & Biochemistry*, *122*, 203–210. https://doi.org/10.1016/j.soilbio.2018.04.018

David, M. B., McIsaac, G. F., Darmody, R. G., & Omonode, R. A. (2009). Long-term changes in Mollisol organic carbon and nitrogen.

Journal of Environmental Quality, *38*, 200–211. https://doi.org/10.2134/jeq2008.0132

Dietzel, R., Liebman, M., & Archontoulis, S. (2017). A deeper look at the relationship between root carbon pools and the vertical distribution of the soil carbon pool. *SOIL*, *3*, 139–152. https://doi.org/10.5194/soil-3-139-2017

De Deyn, G. B., Cornelissen, J. H. C., & Bardgett, R. D. (2008). Plant functional traits and soil carbon sequestration in contrasting biomes. *Ecology Letters*, *11*, 516–531. https://doi.org/10.1111/j.1461-0248.2008.01164.x

Ding, X. L., Qiao, Y. F., Filley, T., Wang, H., Lü, X., Zhang, B., & Wang, J. K. (2017). Long-term changes in land use impact the accumulation of microbial residues in the particle-size fractions of a Mollisol. *Biology and Fertility of Soils*, *53*, 281–286. https://doi.org/10.1007/s00374-017-1179-z

Don, A., Scholten, T., & Schulze, E. D. (2009). Conversion of cropland into grassland: Implications for soil organic-carbon stocks in two soils with different texture. *Journal of Plant Nutrition and Soil Science*, *172*, 53–62. https://doi.org/10.1002/jpln.200700158

Duval, M. E., Galantini, J. A., Martínez, J. M., & Limbozzi, F. (2018). Labile soil organic carbon for assessing soil quality: Influence of management practices and edaphic conditions. *CATENA*, *171*, 316–326. https://doi.org/10.1016/j.catena.2018.07.023

Fontaine, S., Barot, S., Barré, P., Bdioui, N., Mary, B., & Rumpel, C. (2007). Stability of organic carbon in deep soil layers controlled by fresh carbon supply. *Nature*, *450*, 277–280. https://doi.org/10.1038/nature06275

Gregorich, E. G., Beare, M. H., Stoklas, U., & St-Georges, P. (2003). Biodegradability of soluble organic matter in maize-cropped soils. *Geoderma*, *113*, 237–252. https://doi.org/10.1016/s0016-7061(02)00363-4

Gregory, A., Dungait, J., Watts, C., Bol, R., Dixon, E., White, R., & Whitmore, A. (2016). Long-term management changes topsoil and subsoil organic carbon and nitrogen dynamics in a temperate agricultural system. *European Journal of Soil Science*, *67*, 421–430. https://doi.org/10.1111/ejss.12359

Guimarães, D. V., Gonzaga, M. I. S., da Silva, T. O., da Silva, T. L., da Silva Dias, N., & Matias, M. I. S. (2013). Soil organic matter pools and carbon fractions in soil under different land uses. *Soil and Tillage Research*, *126*, 177–182. https://doi.org/10.1016/j.still.2012.07.010

Guo, L. B., & Gifford, R. M. (2002). Soil carbon stocks and land use change: A meta analysis. *Global Change Biology*, *8*, 345–360. https://doi.org/10.1046/j.1354-1013.2002.00486.x

Guo, L. B., Halliday, M. J., & Gifford, R. M. (2006). Fine root decomposition under grass and pine seedlings in controlled environmental conditions. *Applied Soil Ecology*, *33*, 22–29. https://doi.org/10.1016/j.apsoil.2005.09.004

Guo, L. B., Wang, M., & Gifford, R. M. (2007). The change of soil carbon stocks and fine root dynamics after land use change from a native pasture to a pine plantation. *Plant and Soil*, *299*, 251–262. https://doi.org/10.1007/s11104-007-9381-7

Han, X. Z., & Li, N. (2018). Research progress of black soil in Northeast China. *Scientia Geographica Sinica*, *38*, 1032–1041. https://doi.org/10.13249/j.cnki.sgs.2018.07.004

Hao, X. X., Han, X. Z., Li, L. J., Zou, W. X., Lu, X. C., & Qiao, Y. F. (2015). Profile distribution and storage of soil organic carbon in a black soil as affected by land use types. (In Chinese with English abstract.) *Journal of Applied Ecology*, *26*, 965–972. https://doi.org/10.13287/j.1001-9332.2015.0010

Hassouna, M., Massiani, C., Dudal, Y., Pech, N., & Theraulaz, F. (2010). Changes in water extractable organic matter (WEOM) in a calcareous soil under field conditions with time and soil depth. *Geoderma*, *155*, 75–85. https://doi.org/10.1016/j.geoderma.2009.11.026

Hou, X. Y., Han, X. Z., Li, H. B., & Xing, B. S. (2010). Composition and organic carbon distribution of organomineral complex in black soil under different land uses and management systems. *Communications in Soil Science and Plant Analysis*, *41*, 1129–1143. https://doi.org/10.1080/00103620903430016

Hurisso, T., Norton, J., & Norton, U. (2013). Soil profile carbon and nitrogen in prairie, perennial grass-legume mixture and wheat-fallow production in the central High Plains, USA. *Agriculture Ecosystems and Environment*, *181*, 179–187. https://doi.org/10.1016/j.agee.2013.10.008

Jackson, R. B., Canadell, J., Ehleringer, J. R., Mooney, H. A., Sala, O. E., & Schulze, E. D. (1996). A global analysis of root distributions for terrestrial biomes. *Oecologia*, *108*, 389–411. https://doi.org/10.1007/bf00333714

Jenkinson, D. S., Poulton, P. R., & Bryant, C. (2008). The turnover of organic carbon in subsoils. Part 1 Natural and bomb radiocarbon in soil profiles from the Rothamsted long-term field experiments. *European Journal of Soil Science*, *59*, 391–399. https://doi.org/10.1111/j.1365-2389.2008.01025.x

Jobbágy, E. G., & Jackson, R. B. (2000). The vertical distribution of soil organic carbon and its relation to climate and vegetation. *Ecological Applications*, *10*, 423–436. https://doi.org/10.1890/1051-0761(2000)010[0423:tvdoso]2.0.CO;2

John, B., Yamashita, T., Ludwig, B., & Flessa, H. (2005). Storage of organic carbon in aggregate and density fractions of silty soils under different types of land use. *Geoderma*, *128*, 63–79. https://doi.org/10.1016/j.geoderma.2004.12.013

Jones, J. B. Jr. (1973). Soil testing in the United States. *Communications in Soil Science and Plant Analysis*, *4*, 307–322. https://doi.org/10.1080/00103627309366451

Kalinina, O., Krause, S. E., Goryachkin, S. V., Karavaeva, N. A., Lyuri, D. I., & Giani, L. (2011). Self-restoration of post-agrogenic chernozems of Russia: Soil development, carbon stocks, and dynamics of carbon pools. *Geoderma*, *162*, 196–206. https://doi.org/10.1016/j.geoderma.2011.02.005

Kong, A. Y. Y., & Six, J. (2010). Tracing root vs. residue carbon into soils from conventional and alternative cropping systems. *Soil Science Society of America Journal*, *74*, 1201–1210. https://doi.org/10.2136/sssaj2009.0346

Kotzé, E., Loke, P. F., Akhosi-Setaka, M. C., & Du Preez, C. C. (2016). Land use change affecting soil humic substances in three semi-arid agro-ecosystems in South Africa. *Agriculture Ecosystems and Environment*, *216*, 194–202. https://doi.org/10.1016/j.agee.2015.10.007

Laganière, J., Angers, D. A., & Paré, D. (2010). Carbon accumulation in agricultural soils after afforestation: A meta-analysis. *Global Change Biology*, *16*, 439–453. https://doi.org/10.1111/j.1365-2486.2009.01930.x

Li, N., Long, J. H., Han, X., Yuan, Y., & Sheng, M. (2020). Molecular characterization of soil organic carbon in water-stable aggregate fractions during the early pedogenesis from parent material of Mollisols. *Journal of Soils and Sediments*, *20*, 1869–1880. https://doi.org/10.1007/s11368-020-02563-w

Liu, M. Y., Chang, Q. R., Qi, Y. B., Liu, J., & Chen, T. (2014). Aggregation and soil organic carbon fractions under different land uses on

the tableland of the Loess Plateau of China. *CATENA*, *115*, 19–28. https://doi.org/10.1016/j.catena.2013.11.002

Llorente, M., Glaser, B., & Turrión, M. B. (2010). Storage of organic carbon and black carbon in density fractions of calcareous soils under different land uses. *Geoderma*, *159*, 31–38. https://doi.org/10.1016/j.geoderma.2010.06.011

Lorenz, K., & Lal, R. (2005). The depth distribution of soil organic carbon in relation to land use and management and the potential of carbon sequestration in subsoil horizons. *Advances in Agronomy*, *88*, 35–66. https://doi.org/10.1016/s0065-2113(05)88002-2

Luo, Z. K., Luo, Y. Q., Wang, G. C., Xia, J. Y., & Peng, C. H. (2020). Warming-induced global soil carbon loss attenuated by downward carbon movement. *Global Change Biology*, *26*, 7242–7254. https://doi.org/10.1111/gcb.15370

McLauchlan, K. K., Hobbie, S. E., & Post, W. M. (2006). Conversion from agriculture to grassland builds soil organic matter on decadal timescales. *Ecological Applications*, *16*, 143–153. https://doi.org/10.1890/04-1650

Mertens, J., Vanderborght, J., Kasteel, R., Pütz, T., Merckx, R., Feyen, J., & Smolders, E. (2007). Dissolved organic carbon fluxes under bare soil. *Journal of Environmental Quality*, *36*, 597–606. https://doi.org/10.2134/jeq2006.0368

Novara, A., La Mantia, T., Rühl, J., Badalucco, L., Kuzyakov, Y., Gristina, L., & Laudicina, V. A. (2014). Dynamics of soil organic carbon pools after agricultural abandonment. *Geoderma*, *36*, 191–198. https://doi.org/10.1016/j.geoderma.2014.07.015

Oades, J. M., & Waters, A. G. (1991). Aggregate hierarchy in soils. *Australian Journal of Soil Research*, *29*, 815–828. https://doi.org/10.1071/SR9910815

Olsen, S. R., & Sommers, L. E. (1982). Phosphorus. In Page A. L. (Ed.), *Methods of soil analysis: Part 2 Chemical and microbiological properties, 9.2.2* (2nd ed., pp. 403–430). ASA and ASSA. https://doi.org/10.2134/agronmonogr9.2.2ed.c24

Page, A. L., Miller, R. H., & Keeney, D. R. (1982). *Methods of soil analysis: Part 2 Chemical and microbiological properties, 9.2.2* (2nd ed., pp. 885–891). ASA and ASSA.

Paul, K. I., Polglase, P. J., Nyakuengama, J. G., & Khanna, P. K. (2002). Change in soil carbon following afforestation. *Forest Ecology and Management*, *168*, 241–257. https://doi.org/10.1016/s0378-1127(01)00740-x

Poeplau, C., & Don, A. (2013). Sensitivity of soil organic carbon stocks and fractions to different land-use changes across Europe. *Geoderma*, *192*, 189–201. https://doi.org/10.1016/j.geoderma.2012.08.003

Poeplau, C., Don, A., Vesterdal, L., Leifeld, J., Van Wesemael, B., Schumacher, J., & Gensior, A. (2011). Temporal dynamics of soil organic carbon after land-use change in the temperate zone—Carbon response functions as a model approach. *Global Change Biology*, *17*, 2415–2427. https://doi.org/10.1016/j.geoderma.2012.08.003

Rumpel, C., & Kögel-Knabner, I. (2011). Deep soil organic matter-a key but poorly understood component of terrestrial C cycle. *Plant and Soil*, *338*, 143–158. https://doi.org/10.1007/s11104-010-0391-5

Salomé, C., Nunan, N., Pouteau, V., Lerch, T. Z., & Chenu, C. (2010). Carbon dynamics in topsoil and in subsoil may be controlled by different regulatory mechanisms. *Global Change Biology*, *16*, 416–426. https://doi.org/10.1111/j.1365-2486.2009.01884.x

Sheng, H., Zhou, P., Zhang, Y., Kuzyakov, Y., Zhou, Q., Ge, T., & Wang, C. (2015). Loss of labile organic carbon from subsoil due to land-use changes in subtropical China. *Soil Biology & Biochemistry*, *88*, 148–157. https://doi.org/10.1016/j.soilbio.2015.05.015

Sheng, M., Han, X. Z., Zhang, Y. H., Long, J. H., & Li, N. (2020). 31-year contrasting agricultural managements affect the distribution of organic carbon in aggregate-sized fractions of a Mollisol. *Science Reports*, *10*, 9041. https://doi.org/10.1038/s41598-020-66038-1

Six, J., Elliott, E. T., & Paustian, K. (2000). Soil macroaggregate turnover and microaggregate formation: A mechanism for C sequestration under no-tillage agriculture. *Soil Biology & Biochemistry*, *32*, 2099–2103. https://doi.org/10.1016/s0038-0717(00)00179-6

Soil Survey Staff. (2010). *Keys to soil taxonomy* (11th ed.). USDA–Natural Resources Conservation Service. https://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs142p2_050915.pdf

Sokol, N. W., Kuebbing, S. E., Karlsen-Ayala, E., & Bradford, M. A. (2019). Evidence for the primacy of living root inputs, not root or shoot litter, in forming soil organic carbon. *New Phytologist.*, *221*, 233–246. https://doi.org/10.1111/nph.15361

Stevenson, F. J. (1994). *Humus chemistry: Genesis, composition, reactions* (2nd ed.). John Wiley and Sons Press.

Valentin, L. D. G., Kurganova, I., & Kuzyakov, Y. (2008). Carbon pool and sequestration in former arable Chernozems depending on restoration period. *Ekologija*, *54*, 232–238. https://doi.org/10.2478/v10055-008-0034-9

Yamashita, T., Flessa, H., John, B., Helfrich, M., & Ludwig, B. (2006). Organic matter in density fractions of water-stable aggregates in silty soils: Effect of land use. *Soil Biology & Biochemistry*, *38*, 3222–3234. https://doi.org/10.1016/j.soilbio.2006.04.013

Yang, X. M., Zhang, X. P., Fang, H. J., & Liang, A. Z. (2004). Changes in organic matter and total nitrogen of black soils in Jilin Province over the past two decades. *Scientia Geographica Sinica*, *24*, 710–714. https://doi.org/10.13249/j.cnki.sgs.2004.06.012

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Hao, X., Han, X.-Z., Li, N., Lei, W., Chen, X., & Xing, B. (2022). Long-term grassland restoration exerts stronger impacts on the vertical distribution of labile over recalcitrant organic carbon fractions in Mollisols. *Soil Science Society of America Journal*, *86,* 1444–1456. https://doi.org/10.1002/saj2.20422