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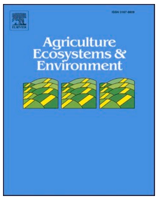

Dynamics of new- and old- soil organic carbon and nitrogen following afforestation of abandoned cropland along soil clay gradient

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ABSTRACT

Disentangling the changes of newly input and original organic carbon (OC) and nitrogen (N) after agricultural land-use transition is crucial for understanding the mechanisms behind soil OC and N dynamics in terrestrial ecosystems. However, these dynamics have received little attention so far due to the lack of an approach separating them. In this study, we determined the natural abundance of ^{13}C and ^{15}N in soils from afforested forestlands and adjacent croplands, the forestlands were established at different times (10, 20 and 30 years) at four sites with different soil textures across the Loess Plateau of China. The objective was to explore the changes of new- and old- OC and N along with soil clay gradient, and to examine the proportions of these changes in total OC and N along with the clay content. We showed that the conversion of cropland to forestland increased total- and new- OC and N stocks by 0.69 kg m^{-2} and 0.10 kg m^{-2}, but decreased old OC and N stocks by -0.20 kg m^{-2} and -0.04 kg m^{-2}, respectively. When averaged across afforestation age, the proportions of new OC and N increased, but the proportions of old OC and N decreased with the clay content. In the soils that have a higher clay content, the accumulation of N rates were greater than that of OC in the new stock, and the loss rates of N were higher than that of OC in the old stock after afforestation. In contrast, in the soils that have a lower clay content, the accumulation rates of N were lower than that of OC in the new stock, and the loss rates of N were smaller than the loss of OC in the old stock. These findings indicated that soil texture regulates the changes of OC and N in new and old soil organic matter pools following cropland afforestation.

1. Introduction

As essential determinants of terrestrial ecosystems, soil organic carbon (C) and nitrogen (N) cycles are strongly coupled with each other in terrestrial ecosystems (Marañón-Jiménez et al., 2019; Soussana and Lemaire, 2014). The interaction between C and N can reflect whether organic carbon (OC) accumulation is sustainable in the long run (Tian et al., 2017). Generally, the input of N is positively correlated with the accumulation of OC (Huang et al., 2020; Zhang et al., 2020). Shift in agricultural land use has important impacts on soil C and N by changing input, decomposition and turnover of C and N (Karhu et al., 2011; Li et al., 2014, 2015). After converting cropland to forestland, the original OC and N that derived from previous cropland will be lost, and the newly input OC and N that derived from the afforested forest will accumulate (Hu et al., 2016). Nevertheless, the decomposition of original organic material (derived from cropland) would be stimulated due to the priming effects (Brant et al., 2006; Kuzyakov et al., 2000) or would be decreased due to physical protection by increased aggregation of soil particles (Barthès et al., 2008; Six et al., 2004). Furthermore, the accumulation of new inputs of OC and N varies with the original OC and N levels. For example, when the OC and N contents in the original soil are high, the plants in the soil grow vigorously and produce more aboveground and underground biomass (Foster et al., 2003), and new OC and N returned to the soil may increase correspondingly (Zhao et al., 2015).

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These variations in new- and old- soil OC and N hinder our comprehensive understanding of the sequestration of C and N (Huang et al., 2020). Such knowledge is imperative to better predict the dynamics response of soil OC and N cycles and their feedbacks to land-use change.

The impact of agricultural land-use changes on C and N stocks depend on the duration of land-use change, tree species used, climate (temperature and precipitation), and edaphic factors (Deng et al., 2014; Han et al., 2019). Numerous studies indicated that duration time is a critical factor influencing soil C and N sequestration (Mujuru et al., 2014; Li et al., 2019). For example, it has been showed that the C and N stocks were either minimally affected within decades of afforestation and significantly increased at the later stage of afforestation (Chen et al., 2013; Li et al., 2012) or continuously increased with the time of afforestation (Chen et al., 2018; Li et al., 2019; Qiu et al., 2010). This knowledge has advanced our understanding of soil C and N cycles in restored forest systems. Given that soil OC and N in afforested ecosystems are composed of old OC and N that derived from previous ecosystems and the newly input OC and N that derived from afforested ecosystems, the response of ecosystems to the old and new inputs regulates the changes of total soil OC and N (Deng et al., 2016). However, the limitations of the technique to separate new OC and N from old OC and N under field conditions restrict our insights into the mechanisms underlying the changes of OC and N and their precise prediction. In the past two decades, researchers have used changes in ^{13}C to separate the sources of OC following land-use shifts between C_3 and C_4 plants (Blagodatskaya et al., 2011; Santos et al., 2020; Wei et al., 2012), providing essential understanding of how new and old OC changes. There are also limited reports separating the old and new N in soils after land-use transition between legume and nonlegume plants (Wei et al., 2014). Moreover, the synchronical shifts in OC and N after land-use change, as well as the differences in C-N interactions in old and new SOM pools, have not been examined, but this is urgently needed due to the important effects of C-N interactions on most terrestrial ecosystems (Han et al., 2020; Luo et al., 2004, 2006; Reich et al., 2006).

Soil texture plays essential role in influencing the response of soil OC and N to agricultural land use change (Gonçalves et al., 2017; Zhou et al., 2019). High clay contents soils are more capable of sequestrating OC and N (Iranmanesh and Sadeghi, 2019; Kong et al., 2009; Wan et al., 2018). To date, several studies have examined the impact of soil texture on OC and N in different land-use types. For example, Gonçalves et al. (2017) found that the quantity of oxide minerals in the clay fraction controls the stabilization and sequestration of soil OC in agricultural ecosystems. Kong et al. (2009) showed that soils with higher clay contents have larger OC accumulation capacities and rates. This positive effect of fine particles on OC accumulation has been due to the physicochemical protection of SOM by clay fractions (Vogel et al., 2015). However, it is unclear how soil texture determines the response of new- and old- OC and N stocks to the agricultural land-use change, which limits our understanding of total OC and N changes and assessment of the direction and magnitude of OC and N feedback in forest systems.

Herein, we presented the results of OC and N in newly input and old SOM pools along afforestation chronosequence at four sites that varied significantly in the soil clay content across the Loess Plateau. At each site, black locust with three afforestation age (10, 20, 30 years) was chosen, and adjacent lands were selected as controls to compare the effects of agricultural land use change. We determined the natural abundances of ^{13}C and ^{15}N in bulk soil of different afforestation age to distinguish the new- and the old- OC and N. We attempted to evaluate how new- and old- OC and N contents changed and how the C/N ratio of the new stock, old stock and total stock changed along the soil clay gradients after agricultural land use change. We hypothesized the following: (i) The new OC and N stock would increase, but with the extension of time after land use conversion, the old OC and N stock may decrease, as the input of fresh organic materials would provide more OC and N with increasing time (Hu et al., 2016; Kuzyakov et al., 2000). (ii) The proportions of new OC and N would be higher at sites with a high clay content because soil clay more easily absorbs new organic matter material due to its large specific surface area and many reactive sites (Vogel et al., 2015). Conversely, the proportions of old OC and N would be relatively lower at sites with high clay contents. (iii) The effect of soil clay on C/N would vary with old and new SOM pools because the newly input SOM was mainly derived from the C_3 legume plant, while the old SOM was mainly derived from the residual C_4 nonlegume crops after the conversion of cropland to forestland.

In 1980, the Chinese government started the afforestation program in the Loess Plateau of China (Chen et al., 2007; Deng et al., 2014). Leguminous C_3 trees or shrubs were widely planted on degraded or abandoned cropland to restore degraded ecosystems and to conserve soil and prevent water loss (Qiu et al., 2010). In most cases, the crops planted in croplands on the Loess Plateau were nonleguminous C_4 crops (mainly maize and/or millet). The conversion from nonleguminous C_4 crops to leguminous C_3 crops lead to major changes in soil ^{13}C and ^{15}N isotopic signature (Rong et al., 2020). These variations in vegetation types allowed us to distinguish the old and newly input OC and N simultaneously by measuring soil ^{13}C and ^{15}N abundance and allowed us to study their dynamics following cropland afforestation in different sites. We addressed how agricultural land use change affect newly input and old soil OC and N across a soil clay gradient, which was not examined previously.

2. Materials and methods

2.1. Field investigation and sampling

We conducted this research on the Loess Plateau (100°54’–114°33’ E, 33°43’–41°16’ N, 1800–2148 m a.s.l.) in China (Fig. 1). We selected four sites (Fufeng, Binxian, Yan’an and Shenmu from south to north) across the Loess Plateau, across which soil texture and nutrient status varied significantly. The averaged clay contents in Shenmu, Yan’an, Binxian and Fufeng sites were 12.4%, 14.0%, 19.2% and 26.3%, respectively (Table 1). At each site, afforested forestlands with different stand ages (10, 20 and 30 years) were chosen, and an adjacent cropland (0 year) was regarded as a control. We used the space-for-time substitution approach to determine the time of afforestation (Walker et al., 2010). In each site, we selected the afforested land that were established around 1984, 1994 and 2004, respectively, to compose an afforestation chronosequence. The time of afforestation was determined by interviewing the forestry bureau and was identified by the tree-ring dating technique. The forested soil was planted with black locust (*Robinia pseudoacacia* Linn.) at the Yan’an, Binxian, and Fufeng sites and with peashrubs (*Caragana korshinskii* Kom.) at the Shenmu site. Both black locust and peashrub are leguminous C_3 trees. At previous cropland, the cropping system was winter wheat (*Triticum aestivum* L.) and summer maize (*Zea mays* Linn.) rotation in Fufeng, millet (*Panicum miliaceum* L.) or summer maize monoculture in Shenmu, and winter wheat or summer maize monocropping in Binxian and Yan’an. Maize and millet are non-leguminous C_4 crops. The differences in such plant assembling enabled us to identify the newly input and old OC and N in soils (Rong et al., 2020). Since the 1970s, the cropland has not actually received manure fertilizer, and after the 1970s, chemical N and P fertilizers were added to cropland at rates of 360 kg ha^{-1} and 135 kg ha^{-1} each year, respectively (Ge et al., 2019).

In September 2014, three replicate plots (10 m × 10 m in size) were designated in afforested forestlands and adjacent cropland at each site. Within the same site, we randomly selected seven representational points in each plot that has similar geographical conditions. All the sampling plots have similar slope gradients, directions, and positions. The slopes of sampling plots are all close to or smaller than 5°. The plots of forestlands and adjacent agricultural land were all located in the north slopes. These sampling points were located at a distance of at least 1.0 m from the trunks of black locust or the main stems of the peashrubs and were at least 50 m from the border between the two land-uses to

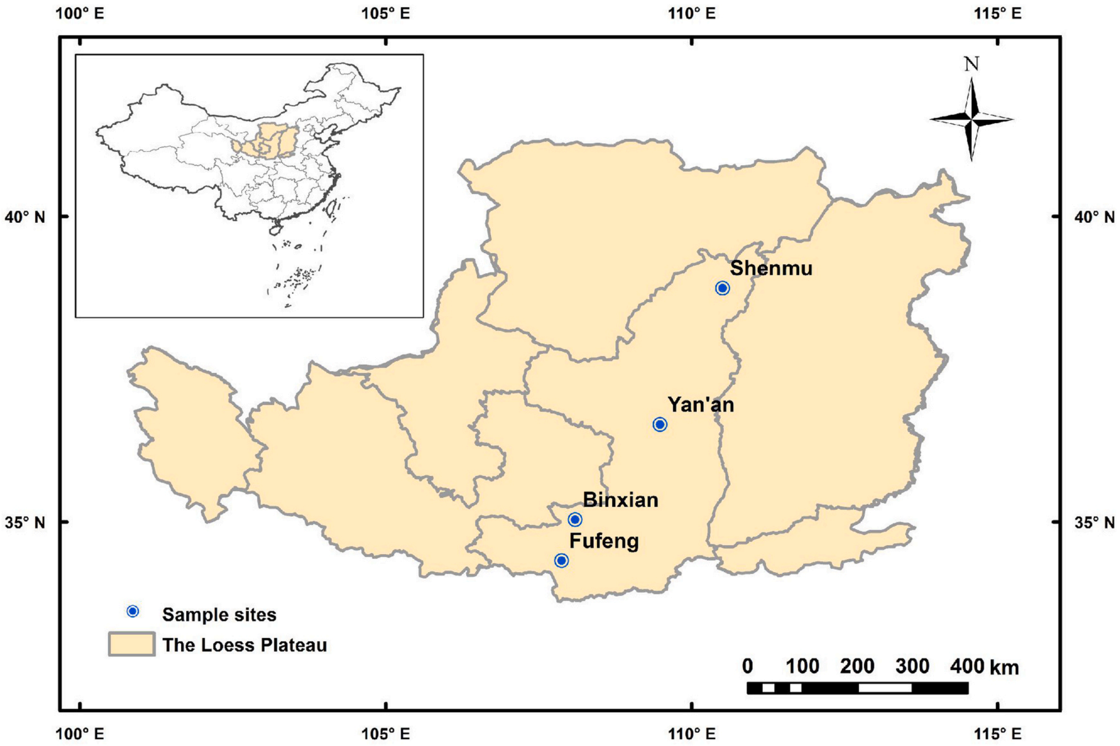

Fig. 1. Location of the four study sites across China's Loess Plateau.

Table 1
Geographical, climatic and soil conditions of the four study sites across China's Loess Plateau.

Metrics	Shenmu	Yan'an	Binxian	Fufeng
Longitude (E)	110°21′–110°23′	109°14′–110°07′	107°47′–108°22′	107°54′–107°58′
Latitude (N)	38°46′–38°51′	36°11′–37°02′	34°51′–35°17′	34°32′–34°35′
Altitude (m)	1039	1200	1108	1008
MAP (mm)	437	550	579	592
MAT (°C)	8.4	9.4	9.7	12.4
AI	0.31	0.56	0.37	0.66
BD (g cm^{-3})	1.43 ± 0.09	1.12 ± 0.18	1.31 ± 0.89	1.26 ± 0.13
Clay content (%)	12.43 ± 0.55	13.99 ± 0.41	19.17 ± 0.32	26.32 ± 0.58
Silt content (%)	16.38 ± 0.68	26.26 ± 0.57	32.42 ± 0.72	40.23 ± 0.74
Sand content (%)	71.17 ± 1.09	59.72 ± 0.59	48.38 ± 0.72	33.43 ± 0.93
SOM (g kg^{-1})	3.58 ± 0.69	6.13 ± 1.59	19.75 ± 1.32	32.84 ± 3.31
Soil type	Cambisols	Cambisols	Cambisols	Anthrosols

Note: Soils are classified according to the Food and Agriculture Organization classification system, 2006; MAP: mean annual precipitation; MAT: mean annual temperature; AI: aridity index, which is the ratio of precipitation to potential evapotranspiration; BD: bulk density. SOM: the amount of organic matter.

minimize the impacts of forestland on cropland or vice versa. The depth of the organic layer in forestland varied with the age of forest, and there was no organic layer in the cropland and afforested lands. We collected mineral soils from 0–10 to 10–20 cm soil depths in each plot by using a soil corer and then mixed them to make composite samples. These soil samples were air-dried for the analysis of soil metrics. Additionally, we collected 3 replicated samples from each soil layer (0–10, 10–20 cm depth) in each plot using a stainless-steel ring cutter (5 cm diameter × 5 cm height) to measure soil bulk density (BD).

2.2. Laboratory analyses

The air-dried soil samples were ground to pass through a 2.0 mm sieve for analysis of soil particle size. A small fraction of the < 2.0 mm sample were ground to pass through a 0.25 mm sieve for analysis of the OC, N contents. Soil texture was analyzed using the sieve pipette method (Day, 1965). Soil OC and N contents were analyzed with the Walkley and Black method and the Kjeldahl method, respectively (Page et al., 1982). Soil bulk density (BD) was determined based on the original volume and dry mass (dried at 105 °C) of each soil core using the soil bulk sampler method (Jia et al., 2005). The natural abundances of ^{13}C and ^{15}N were measured with an isotope ratio mass spectrometer (C/N Isotope, Sercon Ltd., Cheshire, UK) at the University of California, Davis.

2.3. Data and statistical analyses

The C and N isotope ratios were expressed relative to the international PDB limestone standard (Balesdent and Mariotti, 1996). By convention, the ^{13}C and ^{15}N abundances in a sample were expressed in delta-units ($\delta^{13}C$ and $\delta^{15}N$, ‰) according to the following equation (Tiunov, 2007):

$$\delta^{n}E = \left[\left(R_{sample} - R_{standard}\right)/R_{standard}\right] \times 1000 \quad (1)$$

where E is the element (e.g., C or N), n is the weight of the heavier isotope, and R is the ratio of the heavy to light isotopes.

The $\delta^{13}C$ and $\delta^{15}N$ values of OC and N were used to calculate the proportions of new C and N (f_{new}, OC or N derived from the current forestland) for bulk soil. The δ^{13}C and δ^{15}N of SOM were used to calculate the proportions of new OC and N (f_{new}). The proportions of old OC and N (f_{old}) were subtracted from 1. This calculation was done using the mass balance equation described by Balesdent and Mariotti (1996):

$$f_{new} = (\delta_{new} - \delta_{old})/(\delta_{veg} - \delta_{old}) \quad (2)$$

$$f_{old} = 1 - f_{new} \quad (3)$$

where δ_{new} is the δ^{13}C or δ^{15}N value (‰) of the bulk soils in afforested soil, δ_{old} is the δ^{13}C or δ^{15}N value (‰) of the bulk soils in cropland soils (assuming that in the last > 30 years, there has been no shift in the ratio between C_3/C_4 input in the cropland soil), and δ_{veg} is the δ^{13}C or δ^{15}N value (‰) of the forest litter.

The stocks of soil OC and N (kg m^{-2}) in bulk soils were calculated as:

$$\text{OC stock} = \frac{D \times BD \times OC}{100} \quad (4)$$

$$\text{N stock} = \frac{D \times BD \times N}{100} \quad (5)$$

where D is the thickness (cm) of the soil depth, BD is the bulk density (g cm^{-3}), and OC and N are the OC and N contents (g kg^{-1}), respectively, of the 0–10 or 10–20 cm soil depths.

The new- and old- OC and N stocks in the bulk soils were calculated by multiplying the current total OC and N stocks by the corresponding proportions of new- and old- soil OC and N in the bulk soil. The details are as follows:

$$\text{New OC } stock = \text{OC } stock \times f_{new\ OC} \quad (6)$$

$$\text{Old OC } stock = \text{OC } stock \times f_{old\ OC} \quad (7)$$

$$\text{New N } stock = \text{N } stock \times f_{new\ N} \quad (8)$$

$$\text{Old N } stock = \text{N } stock \times f_{old\ N} \quad (9)$$

Where OC or N stock are the stocks of soil OC and N (kg m^{-2}) in bulk soils, $f_{new\ oc}$ and $f_{old\ oc}$ are the corresponding proportions of new- and old- soil OC in the bulk soil, respectively, and $f_{new\ N}$ and $f_{old\ N}$ are the corresponding proportions of new- and old- soil N in the bulk soil, respectively.

First, we tested the data for normality and homogeneity of variance. The logarithmic or square root transformation were used to stabilize the distribution to approximate normality when the data did not exhibit a normal distribution. Three-way analysis of variance (ANOVA) was carried out to test the effects of site, afforestation age and soil depth on soil δ^{13}C and δ^{15}N and the stocks of OC and N in bulk soils and new- and old- SOM pools. Linear regressions were conducted to examine the correlations of OC and N in bulk soils and different pools as well as their changes after afforestation relative to the soil clay content. All analyses were performed in R v3.6.1 and JMP Pro 13.0 (SAS Institute Inc. Cary, NC, USA).

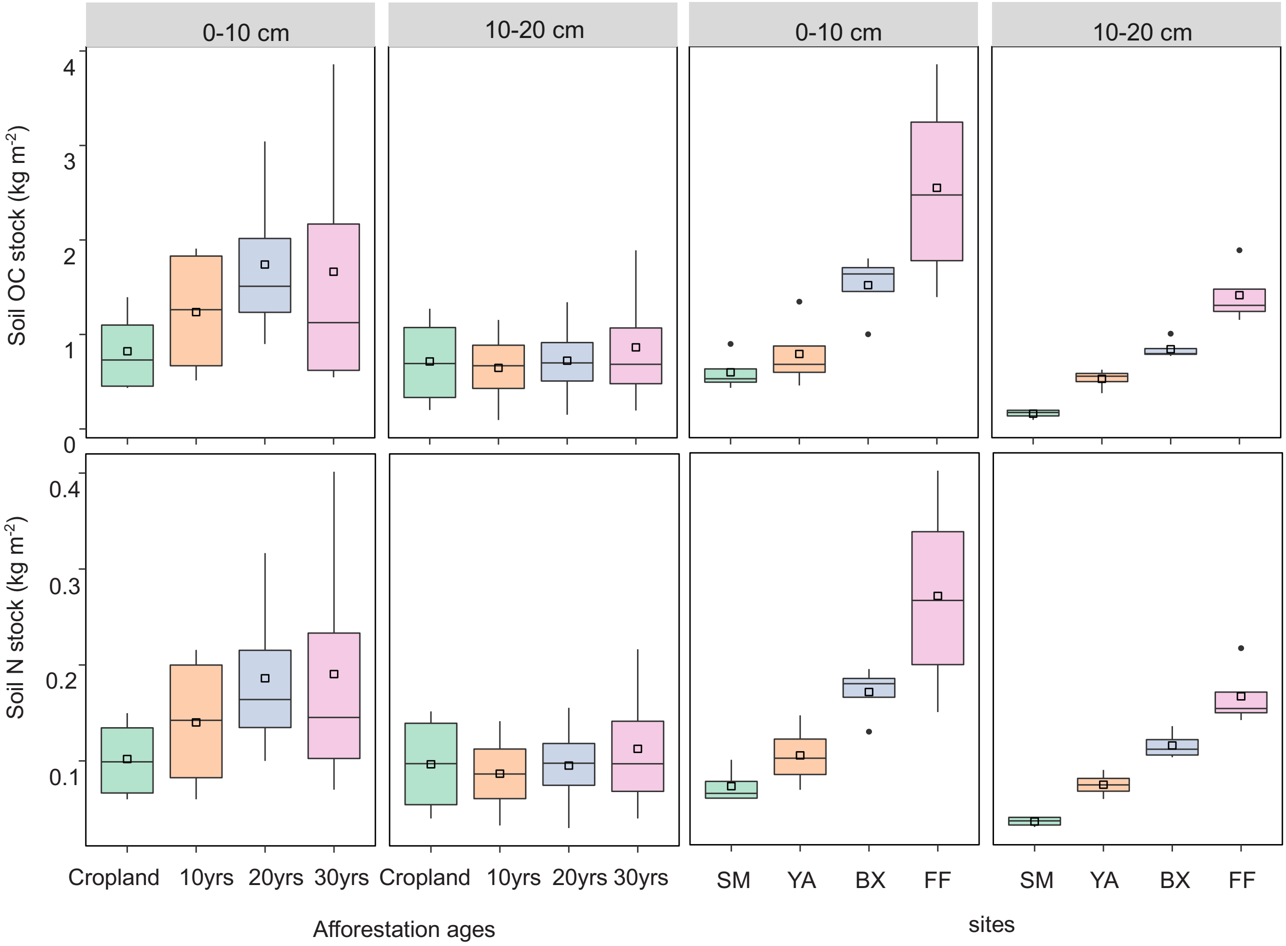

Fig. 2. Soil total organic carbon (OC) and nitrogen (N) stocks averaged across sites (left) and across afforestation ages (right). Lines and squares within boxes represent the median and mean values, respectively. The 10 yrs, 20 yrs and 30 yrs are afforestation ages. SM: Shenmu; YA: Yan'an; BX: Binxian; FF: Fufeng.

3. Results

3.1. Variations in total organic carbon and nitrogen

The total OC stock ranged from 0.09 kg m^{-2} to 3.86 kg m^{-2}, and the total soil N stock ranged from 0.03 kg m^{-2} to 0.40 kg m^{-2} across the four sites (Figs. 2 and S1). When averaged across sites, converting cropland to forestland significantly increased the total OC and N stocks. After 10, 20, and 30 years of land-use transformation, in topsoil, the total OC stock increased by 50.4%, 111.7%, and 102.4%, and the total N stock increased by 38.9%, 83.8%, and 89.0%, respectively. Moreover, the increases in total OC and N were higher in topsoil (1.54 kg m^{-2} and 0.17 kg m^{-2}, $P < 0.01$) than in subsoil (0.74 kg m^{-2} and 0.10 kg m^{-2}, $P > 0.05$), averaged across site and afforestation age.

The increases in the total OC and N stocks varied significantly with site and soil depth (Table 2, Fig. 2). In topsoil, the total OC and N averaged across the afforestation age (10, 20, 30 years) in Fufeng, Binxian, Yan'an and Shenmu increased by 110.6% and 107.8%, 69.3% and 42.6%, 97.1% and 70.7%, 50.4% and 36.3%, respectively. In subsoil, the total OC and N in Fufeng and Yan'an increased by 14.9% and 12.8%, 53.7% and 33.5%, respectively, whereas those in Binxian and Shenmu decreased by 21.7% and 19.8%, 26.8% and 11.2%, respectively (Fig. S1).

3.2. The proportions of new- and old- organic carbon and nitrogen

The proportions of new OC and N (forest-derived, f_{new}) ranged from 21.9–65.6% to 18.5–85.8%, respectively (Figs. 3 and S2). Afforestation increased the f_{new} but decreased the f_{old} at both soil depths. Moreover, f_{new} was higher while the f_{old} was lower in topsoil than in subsoil across afforestation ages and sites (Fig. 3). For instance, in topsoil, new OC and N were 50.3% and 33.1%, 56.5% and 47.1%, and 54.6% and 55.4% in the 10-, 20-, and 30-year forestlands, respectively. In subsoil, the new OC and N were 34.8% and 21.7%, 40.5% and 19.0%, and 48.4% and 41.1%, respectively (Figs. 3 and S2).

The f_{new} of N varied significantly among sites ($P < 0.001$), but that of OC was not affected by site ($P > 0.05$). When averaged across the two depths and afforestation treatments, f_{new} increased but f_{old} decreased along the Shenmu, Yan'an, Binxian and Fuxian site sequences (Fig. 3).

3.3. Dynamic patterns of new and old organic carbon and nitrogen stocks

The new OC and N stocks are at the range in 0.03–2.53 kg m^{-2} for OC and 0.01–0.33 kg m^{-2} for N, and the old OC and N stocks are at the range in 0.06–1.32 kg m^{-2} for OC and 0.03–0.13 kg m^{-2} for N (Fig. 4). The new- and old- OC and N stocks were influenced by afforestation age, soil depth, and site ($P < 0.05$, Table 2), except for the old OC stock, which was similar among the afforestation ages ($P > 0.05$, Table 2).

Converting cropland to forestland increased the new OC and N stocks but decreased the old OC and N stocks when averaged across the sites, and these changes were higher in topsoil than in subsoil (Figs. 4 and S3). Within 30 years of transformation, the respective new OC and N stocks increased by 1.00 kg m^{-2} and 0.13 kg m^{-2} in topsoil and by 0.38 kg m^{-2} and 0.06 kg m^{-2} in subsoil, whereas the old OC and N decreased by 0.16 kg m^{-2} and 0.04 kg m^{-2} in topsoil and by 0.23 kg m^{-2} and 0.04 kg m^{-2} in subsoil (Figs. 4 and S3). When averaged across afforestation age, the OC and N in new and old stocks and their changes after converting cropland to forestland were greater at the Fufeng and Binxian sites but smaller at the Shenmu and Yan'an sites (Fig. 4).

3.4. Effect of agricultural land use change on soil organic carbon and nitrogen stocks across a soil clay gradient

After agricultural land use changes, total OC and N stocks in both soil layers increased with soil clay content, the changes of total OC and N stocks increased with the clay content in topsoil ($P < 0.05$) but not in subsoil (Fig. 5, $P > 0.05$). Moreover, the proportions of new OC and N also increased significantly, whereas the proportions of old OC and N significantly decreased with soil clay in topsoil (Fig. 6). These results showed that the effects of agricultural land use change on total soils, and new and old stocks were dependent on clay content. Therefore, soil texture regulates the response of soil OC and N to agricultural land use change.

3.5. The coupled organic carbon and nitrogen dynamics after agricultural land use change across a clay gradient

There was a positive correlation between OC and N in total soils, and the new and old stocks in the 0–20 cm soil layers ($P < 0.05$, Table 3), indicating coupled C and N dynamics and synchronous sequestration of OC and N in total soils and new input pools or the old stock in each vegetation restoration year after agricultural land use changes. Moreover, the C/N in old stocks increased with the age of afforestation, indicating a greater loss of old N than old OC with increasing afforestation age (Table 3).

When examined across the four sites, the C/N ratio in the total stock had a positive relationship with clay content, and the relationship was

Table 2
Results of variance analysis (F and P values) for the effects of afforestation age (A), site (S) and soil depth (D) on soil organic carbon (OC) and nitrogen (N) stocks and their changes following afforestation.

Variations	S	A	D	S × A	S × D	A × D	S × A × D
	F						
Total OC stock	76.85	33.16	62.03	11.57	4.88	17.92	3.80
Total N stock	86.22	29.42	48.63	11.34	5.46	14.40	3.45
New OC proportion	0.38	39.16	2.71	0.13	0.69	0.14	0.27
New N proportion	11.28	72.96	11.65	5.48	0.75	2.86	0.60
New OC stock	18.45	47.30	36.63	10.31	7.15	10.50	4.14
Old OC stock	41.53	4.05	8.57	1.44	0.78	0.38	0.26
New N stock	120.50	264.80	105.64	64.17	25.41	43.51	17.05
Old N stock	20.21	21.36	4.96	5.93	0.92	0.00	0.31
	P						
Total OC stock	**<0.001**	**<0.001**	**<0.001**	**0.001**	**0.016**	**0.001**	**0.031**
Total N stock	**<0.001**	**<0.001**	**<0.001**	**0.001**	**0.009**	**0.002**	**0.042**
New OC proportion	0.771	**<0.001**	0.119	0.941	0.569	0.711	0.845
New N proportion	**0.001**	**<0.001**	**0.004**	**0.009**	0.536	0.110	0.625
New OC stock	**<0.001**	**<0.001**	**<0.001**	**0.001**	**0.003**	**0.005**	**0.024**
Old OC stock	**<0.001**	0.061	**0.010**	0.269	0.524	0.548	0.856
New N stock	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**<0.001**
Old N stock	**<0.001**	**0.001**	**0.041**	**0.006**	0.453	0.954	0.817

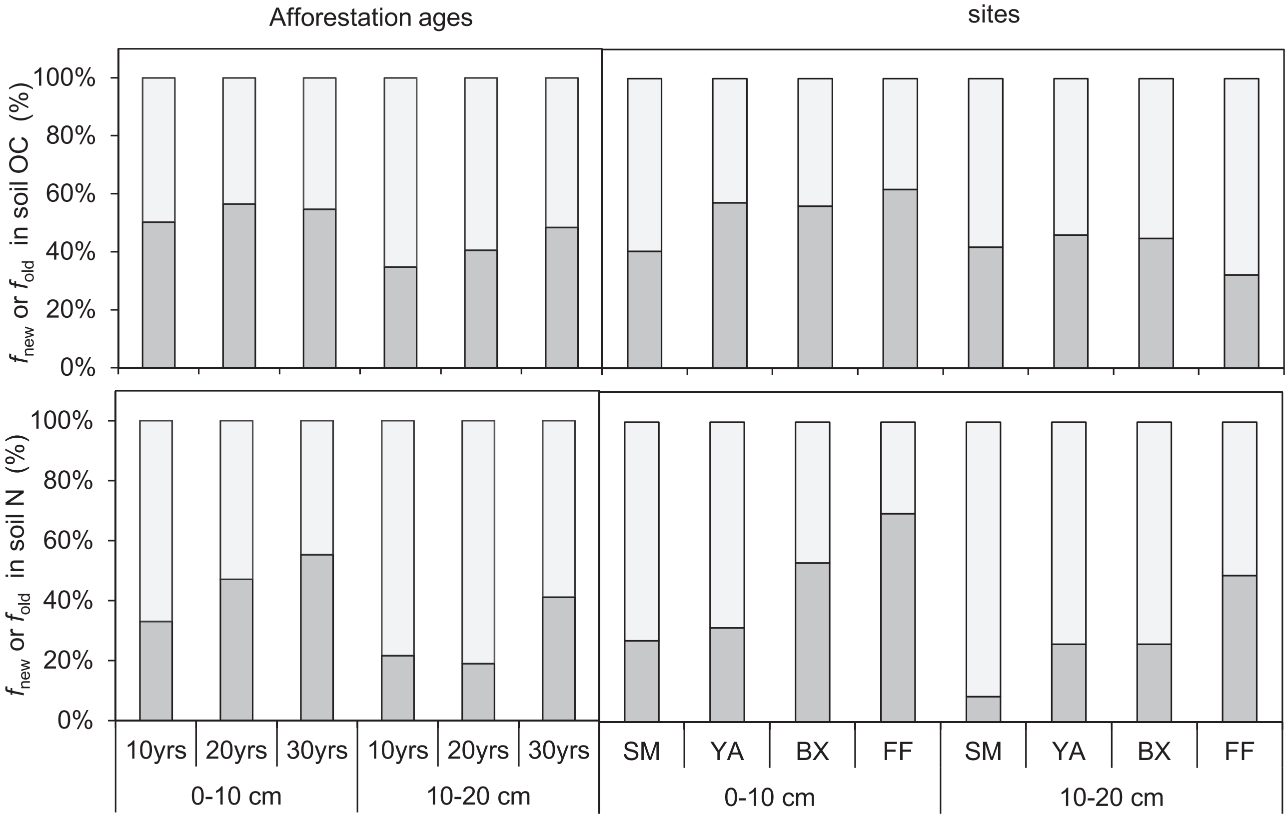

Fig. 3. The proportions of new- (f_{new}) and old- (f_{old}) soil organic carbon (OC) or nitrogen (N) averaged across the sites (left) and across afforestation ages (right) in the 0–10 cm and 10–20 cm depths. Columns in dark gray and light gray are the proportions of new- and old- soil OC or N, respectively. The 10 yrs, 20 yrs and 30 yrs are afforestation ages. SM: Shenmu; YA: Yan'an; BX: Binxian; FF: Fufeng.

determined by the relationship of the C/N ratio in old stock to soil clay (Fig. 7). With the increase in the clay content, the C/N in the new stock decreased (with a slope of -0.34, $R^2 = 0.2621$, $P = 0.0888$), but that in the old stock increased (with a slope of 0.49, $R^2 = 0.4675$, $P = 0.0142$) (Fig. 7). Therefore, clay has a greater ability to protect old C than old N and to accumulate more new N than new C.

4. Discussion

In support of our three hypotheses, after agricultural land use changes, the new OC and N stocks increased, whereas the old OC and N stocks decreased or remained unchanged. The proportions of new OC and N increased, but the old OC and N decreased across the increasing of soil clay content. Most importantly, our results revealed that the C/N ratio of the new stock decreased, whereas the C/N ratio of the old stock increased across a clay gradient after the conversion of cropland to forestland, suggesting a contrasting regulation effect of soil clay on the C/N between old and new pools after agricultural land use changes.

4.1. Effects of agricultural land use change on the total, new and old organic carbon and nitrogen stocks

We observed that total OC and N stocks increased significantly in topsoil at the first 20 years of converting cropland to forestland, with the extension of the land-use change time, the OC and N stocks slowly increased and remained at a stable level (Fig. 2). A potential explanation is that the litter biomass and live aboveground biomass significantly increased in surface soil when the cropland was abandoned and forest vegetation began to grow, resulting in a rapid increase in new OC and N (Lozano-García et al., 2016; Wang et al., 2016). However, as the stand age advanced, the amount of new OC and N inputs gradually decreased, the input and output of SOM became balanced, and the increasing rates of OC and N decreased relative to the initial stages of land use change (Deng et al., 2016; Paul et al., 2002; Zhang et al., 2015). This result is in agreement with some previous research (Abegaz et al., 2020; Li et al., 2019; Wang et al., 2016; Zhang et al., 2015), but differed from the finding recorded by Santos et al. (2020), who found that the C stocks decreased after about 6 years of eucalypt planting, this may be related to the duration of land-use change and tree species (Han et al., 2019; Li et al., 2019).

Our findings go further, showing that new soil OC and N increased but old soil OC and N declined rapidly during the first stages (< 10 years) after converting cropland to forestland, followed by a relatively stable state. These phenomena could be related to the litter input from previous cropland ceased after agricultural land use change, and soil OC (derived from previous litter) would be decomposed by microorganisms and soil enzymes (Benoit et al., 2015). Simultaneously, more fresh organic materials (from forestland) would continue to increase (Richter et al., 1999). Additionally, at first decades of vegetation restoration, the conservation of SOM was not restored or reconstructed well (Zhang et al., 2015), and old OC and N were easily lost at early stages (< 10 years) following cropland afforestation. Together, these results highlighting the vital importance of the new OC and N in supplementing OC and N after agricultural land use change.

Prior work investigated the proportions of new and old OC responses to the conversion of cropland to forestland, finding that the proportion of new soil C increased while old soil C exhibited opposite patterns (Deng et al., 2016; Zhang et al., 2015). Here, we further explored the changes in the proportions of new and old N and quantified the stocks of new- and old- OC and N after agricultural land use change. Our findings revealed that the proportions of new and old N had the same pattern as that of new and old OC, providing more useful information about the OC and N dynamics (Chalk et al., 2019).

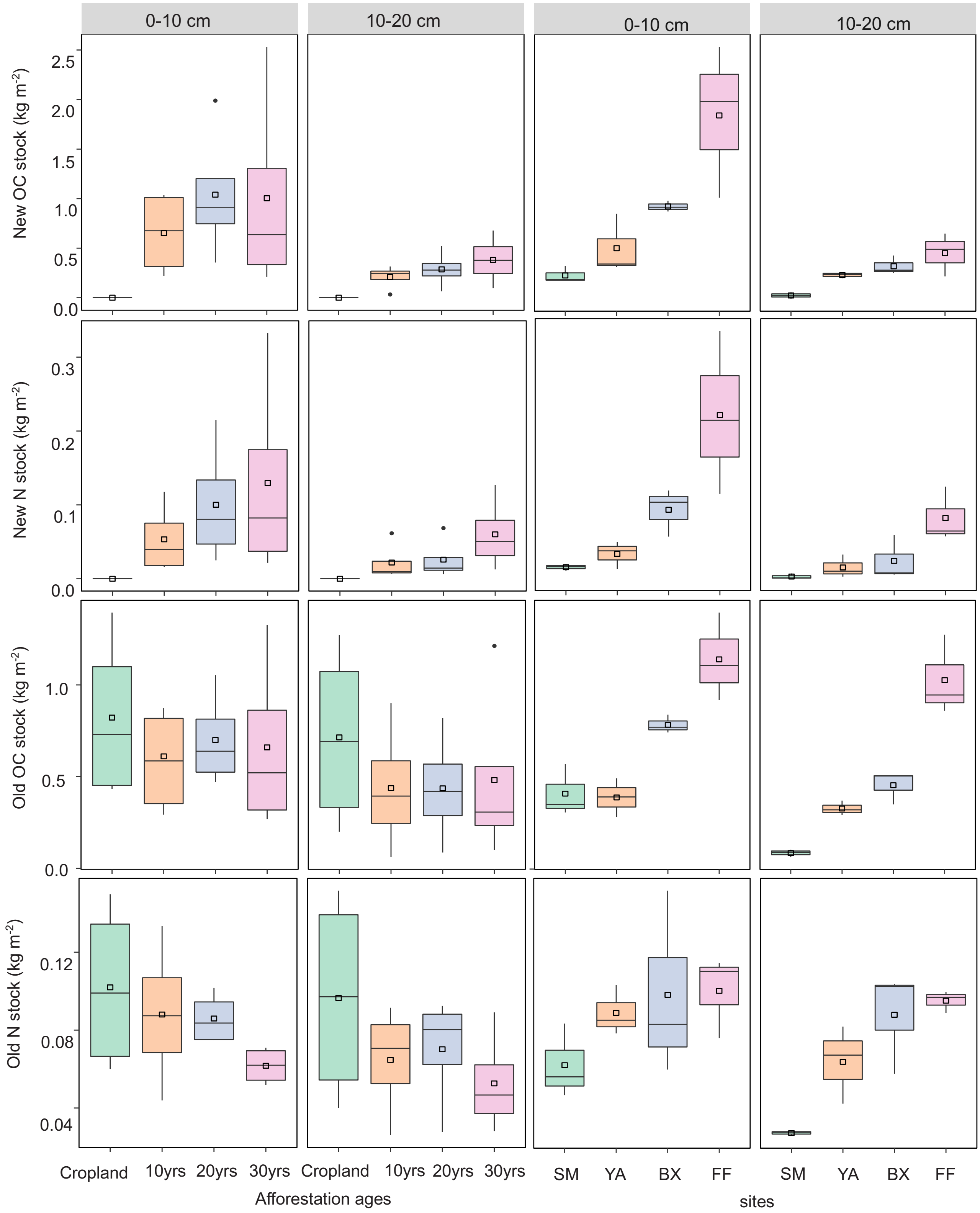

Fig. 4. New- and old- organic carbon (OC) and nitrogen (N) stocks averaged across the sites (left) and afforestation ages (right). Lines and squares within boxes represent the median and mean values, respectively. The 10 yrs, 20 yrs and 30 yrs are afforestation ages. SM: Shenmu; YA: Yan'an; BX: Binxian; FF: Fufeng.

4.2. Effect of agricultural land use change on the total-, new- and old-organic carbon and nitrogen across the clay gradient

Our findings showed that the dynamics of OC and N in total soils and new and old pools were dependent on clay content, indicating soil OC and N contents were influenced by soil texture after agricultural land use change (Gami et al., 2009; Mayer, 1994; Telles et al., 2003). Generally, the stocks of OC and N increase with the clay content (Iranmanesh and Sadeghi, 2019; Whisler et al., 2016; Zhou et al., 2019). Some previous studies also reported that soil with more silt and clay had a higher

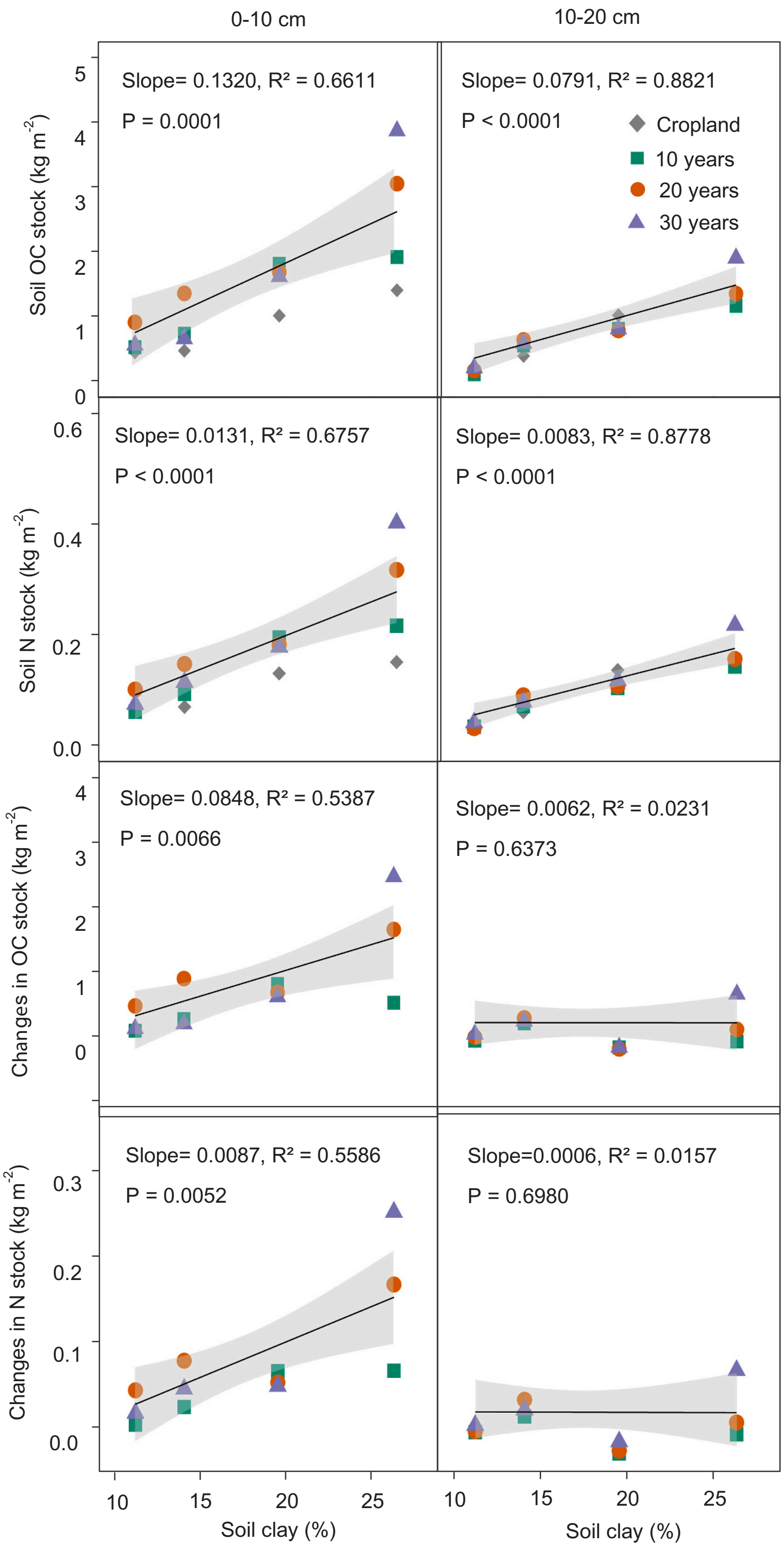

Fig. 5. Spatial distribution of soil organic carbon (OC) and nitrogen (N) stocks in each soil layer (0–10 cm and 10–20 cm) along the soil clay gradient with their 95% confidence intervals (gray-shaded areas).

sequestration capacity in topsoil of agricultural lands and forestlands (Iranmanesh and Sadeghi, 2019; Zhou et al., 2019). This regulation of soil clay on the response of OC and N can be explained by the higher amounts of organic material inputs and the physical and chemical of OC and N in fine particles (Six et al., 2002; Vogel et al., 2015). However, our results are inconsistent with some previous observations that soil texture may not be the major factor regulating SOM accumulation in grasslands (McLauchlan, 2006). These inconsistencies may be due to the influence

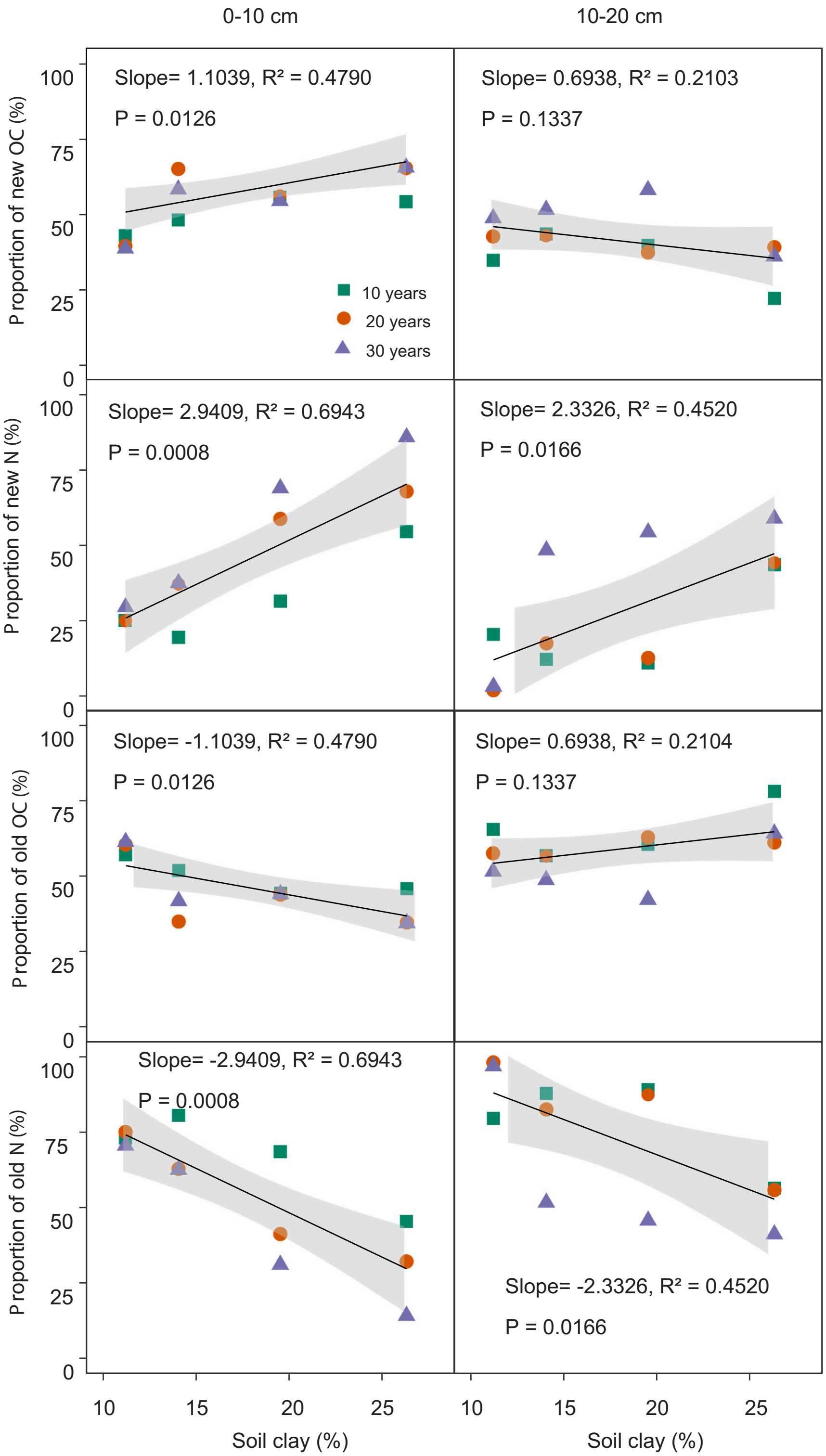

Fig. 6. The proportions of new- and old- organic carbon (OC) and nitrogen (N) (%) in each soil layer (0–10 cm and 10–20 cm) along the soil clay gradient with their 95% confidence intervals (gray-shaded areas).

of clay on C pools that did not reach C saturation a few decades after agriculture land use change (McLauchlan, 2006), which is affected by many complex environmental factors in different ecosystems. Thus, further work is therefore needed to provide more evidence about impact of soil texture on OC and N accumulation, especially at the global scale.

We further observed that the agricultural land use changes increased f_{new} but decreased f_{old} with increasing clay content. The OC and N from new forest vegetation are input into soil after the conversion of cropland to forestland, and this organic matter is easily sorbed in soils with high clay contents due to the exchange of strong ligand and polyvalent cation bridges (Gami et al., 2009; Hassink, 1994), resulting in new OC and N associated with clay soil. Moreover, a high clay content can prevent the mineralization of SOM due to physical and biochemical protection (Six et al., 2002), which contributes to the OC and N accumulations in soil

Table 3
The relationship between soil organic carbon (OC) and nitrogen (N) in 0–20 cm soil layers following afforestation. The 10 yrs, 20 yrs, and 30 yrs are forests with afforestation ages of 10, 20, and 30 years, respectively. ΔTotal stock is the change in soil OC or N stock after afforestation.

	Afforestation age	Regression equation	R^2	P
Δ Total stock (kg m^{-2})	10 yrs	OC=9.49×N+0.03	0.93	**< 0.0001**
	20 yrs	OC=9.90×N+0.04	0.98	**< 0.0001**
	30 yrs	OC=9.90×N−0.03	0.98	**< 0.0001**
New stock (kg m^{-2})	10 yrs	OC=7.80×N+0.14	0.67	**0.0129**
	20 yrs	OC=8.52×N+0.13	0.95	**< 0.0001**
	30 yrs	OC=7.40×N−0.01	0.98	**< 0.0001**
Old stock (kg m^{-2})	10 yrs	OC=7.44×N−0.06	0.64	**0.0177**
	20 yrs	OC=9.90×N−0.20	0.57	**0.0302**
	30 yrs	OC=18.09×N−0.46	0.54	**0.0359**

(Balesdent et al., 2000). Thus, the agricultural land use change increased the proportions of new OC and N across a soil clay gradient. Given that the f_{old} decreased with f_{new}, the proportions of old OC and N decreased along with clay content (Fig. 3).

The relationships of new- and old- OC and N dynamics to soil clay have not been reported previously. Our analysis, by separating new- and old- OC and N pools, indicating that the regulation effect of soil clay on OC and N stocks after agricultural land use change was dominated by the variations of newly input OC and N, which provides a novel perspective in understanding the role of soil texture in influencing the dynamics of OC and N after agricultural land use change.

4.3. The coupled organic carbon and nitrogen dynamics after agricultural land use change across a clay gradient

We observed significant correlations between OC and N in the total

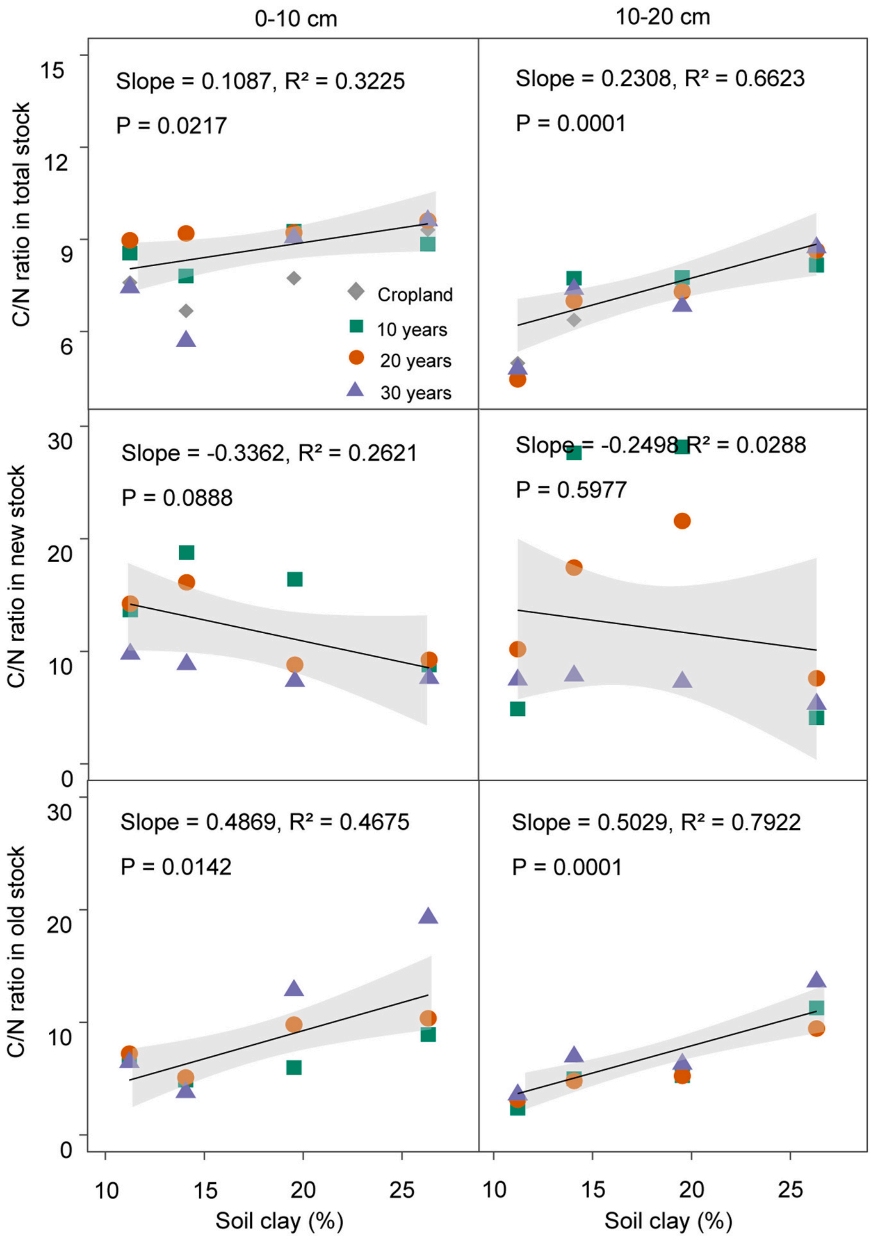

Fig. 7. Spatial distribution in the carbon/nitrogen (C/N) ratio in each soil layer (0–10 cm and 10–20 cm) along the soil clay gradient with their 95% confidence intervals (gray-shaded areas).

stock, the new stock and the old stock in each vegetation restoration year following cropland afforestation (Table 3), suggesting that the coupled OC and N dynamics were time- and soil fraction-independent after agricultural land use change. Our results are similar with the observations of the positive correlations between soil OC and N across afforestation ages and precipitation in topsoils (Han et al., 2019; Zhang et al., 2020) as well as deep soils (Tuo et al., 2018), further confirming the synergistic effect of carbon and nitrogen sequestration in response to this conversion of cropland to forestland.

There were contrasting effects of clay on C/N in old and new SOM pools after agricultural land use change (Fig. 7), indicating that clay has a greater ability to protect old C than old N and a greater ability to accumulate new N than new C. This is more related to the variations in organic materials that are input into the new and old stock and the variations in the types of soil clay (Still et al., 2003; Xu et al., 2020). The SOM in the new stock was mainly derived from C_3 legumes, which led to higher N input after decomposition (Adams et al., 2016; Alegre et al., 2004). It has been demonstrated that leguminous trees have higher N concents in their tissues, and a huge amount of organic litter N in soil would increase the proportion of soil N (Alegre et al., 2004; Xu et al., 2020). Simultaneously, during this process, more fine soil particles associated with these fresh organic materials stabilized them due to the large specific surface area in clay soil (Gami et al., 2009; Telles et al., 2003; Xu et al., 2019). Thus, the accumulating rates of N in the new pool were relatively greater than those of C. In contrast, in the old pools, the SOM was mainly derived from nonleguminous C_4 crops, which have a higher proportion of C than C_3 legumes due to the higher biomass (Still et al., 2003; Yang et al., 2019). After being incorporated into high clay soils, the fine soil particles would protect these substances from being decomposed by microbes (Six et al., 2002), resulting in greater accumulation of C compared with that of N in the old pool after agricultural land use change. This explanation was supported by our observations that the accumulation rates of N were higher than those of OC in the new stock, but the accumulation rates of OC were higher than those of N in the old stock (Fig. S4). These results suggest that the clay content should be considered when assessing the new- and old- OC and N after agricultural land use change at relatively large spatial scales.

5. Conclusions

To our knowledge, this research is the first attempt to analyze the response of new- and old- OC and N pools to agricultural land use change across soil clay gradient. We observed that agricultural land use changes increased total- and new- OC and N stocks but decreased the old pools at the four sites. We also found that agricultural land use changes increased the proportions of new OC and N with increasing soil clay content, whereas the proportions of old OC and N showed the opposite pattern, indicating shifts in the effects of soil clay on the accumulation of OC and N in the new or old SOM pools at a large spatial scale. More importantly, our results demonstrated that the regulate of soil clay on C/N was different between old and new pools after agricultural land use changes, indicating that clay might have a greater ability to protect old OC than old N but a greater ability to accumulate new N than new OC.

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.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2021.107505.

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