

Accumulation of soil organic C and N in planted forests fostered by tree species mixture

Yan Liu¹, Pifeng Lei^{1,2}, Wenhua Xiang¹, Wende Yan^{1,2}, and Xiaoyong Chen^{2,3}

¹Faculty of Life Science and Technology, Central South University of Forestry and Technology, Changsha 410004, Hunan, China

²National Engineering Laboratory for Applied Technology of Forestry & Ecology in South China, Central South University of Forestry and Technology, Changsha 410004, Hunan, China

³Division of Science, College of Arts and Sciences, Governors State University, University Park, Illinois 60484, USA

Correspondence to: Pifeng Lei (pifeng.lei@outlook.com)

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Abstract. With the increasing trend of converting monocultures into mixed forests, more and more studies have been carried out to investigate the admixing effects on tree growth and aboveground carbon storage. However, few studies have considered the impact of mixed forests on belowground carbon sequestration, particularly changes in soil carbon and nitrogen stocks as a forest grows. In this study, paired pure Pinus massoniana plantations, Cinnamomum camphora plantations and mixed Pinus massoniana-Cinnamomum camphora plantations at ages of 10, 24 and 45 years were selected to test whether the mixed plantations sequestrate more organic carbon (OC) and nitrogen (N) in soils and whether this admixing effect becomes more pronounced with stand ages. The results showed that tree species identification, composition and stand age significantly affected soil OC and N stocks. The soil OC and N stocks were the highest in mixed Pinus-Cinnamomum stands compared to those in counterpart monocultures with the same age in the whole soil profile or specific soil depth layers (0–10, 10–20 and 20–30 cm) for most cases, followed by Cinnamomum stands and Pinus stands with the lowest. These positive admixing effects were mostly nonadditive. Along the chronosequence, the soil OC stock peaked in the 24-year-old stand and was maintained as relatively stable thereafter. The admixing effects were also the highest at this stage. However, in the topsoil layer, the admixing effects increased with stand ages in terms of soil OC stocks. When comparing mixed Pinus-Cinnamomum plantations with corresponding monocultures within the same age, the soil N stock in mixed stands was 8.30, 11.17 and 31.45 % higher than the predicted mean value estimated from counterpart pure species plantations in 10-, 24- and 45-year-old stands, respectively. This suggests that these admixing effects were more pronounced along the chronosequence.

1 Introduction

Soil carbon is more stable than that stored in plants, which makes soil carbon more resistant to disturbance (Cunningham et al., 2015). Organic carbon from forest soils accounts for 70-73% of global soil organic carbon (Six et al., 2002), and a notable portion of global forests are plantations. Thereby, plantations play an important role in soil carbon sequestration and mitigating the global atmospheric carbon budget (Cunningham et al., 2015). However, considering the problems caused by pure plantations (Peng et al., 2008), nowadays planting mixed forest with different species in one stand is becoming more and more popular in worldwide plantation management (Felton et al., 2016; Oxbrough et al., 2012) for the potential positive effects (Knoke et al., 2007). Mixed forests, compared with monocultures, are generally characterized by the sustainability to resist disturbance, potential for higher yield and better ecological services (Grime, 1998; Knoke et al., 2007). It is generally accepted that mixed forests exert favorable effects over monocultures by two main mechanisms: the complementary effect and the selection effect (Isbell et al., 2009; Grossiord et al., 2014). The former is explained by inducing facilitation or easing interspecific competition and enhancing resource use efficiency by niche partitioning. The latter is explained by increasing the possibilities of including highly productive species to increase the yield. Though a lot of work has been done to study the positive effects of biodiversity on ecological function (Balvanera et al., 2006; Marquard et al., 2009), attention is rarely given to soil organic carbon (Vesterdal et al., 2013) and nitrogen stocks, which are closely related to global climate change. The limited research shows that the soil OC and N stocks in mixed stands do not necessarily exceed those of corresponding pure stands (Berger et al., 2002; Forrester et al., 2012; Wiesmeier et al., 2013; Wang et al., 2013; Cremer et al., 2016). Most previous studies did not consider the relative portion of the component species (Berger et al., 2002; Wang et al., 2013; Cremer et al., 2016), which may result in an underestimation of admixing effects. For example, admixing effects could also exist because of the small portion of higher-production species, which leads to low expected production even if the production of a twospecies mixed stand is between that of two corresponding pure stands.

Studies of monocultures have suggested that soil organic carbon stocks in the upper soil layer fluctuate in the early stage of afforestation until reaching a new equilibrium that depends on the rates of litter input and decomposition (Paul et al., 2002; Tremblay et al., 2006; Sartori et al., 2007). The soil nitrogen stock increases with increasing stand age in the upper soil layer because of cumulative biological fixation, litterfall, recycling from deeper mineral soil via root mortality and even atmospheric N deposition (Hume et al., 2016). However, the changes in soil organic carbon and nitrogen stocks at different stand ages are rarely quantified and poorly understood, and this is especially true in the context of comparing pairwise mixed forests with pure forest stands along the chronosequence.

In this study we investigated the soil organic carbon and nitrogen stocks of *Pinus massoniana* and *Cinnamomum camphora* pure stands as well as mixed stands at 10, 24 and 45 years old. All the soil samples were taken from the 0–10, 10–20 and 20–30 cm soil layers in the above-mentioned stands in the Hunan Province in China. We hypothesized that (1) the soil organic carbon and nitrogen stocks under mixed stands are higher than those under corresponding monocultures within the same age in the whole soil profile, and (2) these positive admixing effects become more pronounced along the chronosequence.

2 Materials and methods

2.1 Study site

Three parallel forest stands at different stand ages were selected, comprised of mixed *P. massoniana* and *C. camphora* plantations, pure *P. massoniana* plantations and pure

C. camphora at 10, 24 and 45 years old. Some Pinus elliottii were spotted in between and were treated as P. massoniana because of the similarity in growth and biological characteristics (we grouped them as "Pinus" thereafter). Three 20×20 m plots were established in each of the forest stands, including mixed Pinus-Cinnamomum stands and corresponding pure species stands (Pinus and Cinnamomum) at the age of 24 and 45 years in March 2013 in the Botanical Garden in the Hunan Province, China (28°06' N, 113°02' E); this amounted to 18 plots. Due to the lack of young stands in the Botanical Garden, we selected another site located in the Taolin forestry station (28°55' N, 113°03' E) in the Hunan Province, China. This site was used as a nursery and abandoned in 2000; it was replanted in 2003. Three plots for each of the three plantation types at 10 years old were set up with same method mentioned above, but with a size of 12×12 m constrained by the smaller patches there. In total, our study included 27 plots consisting of mixed Pinus-Cinnamomum plantations and corresponding monocultures at ages of 10, 24 and 45 years (Table 1). These two sites are 200 km in distance from each other with similar climates and soil types. The regional climate is typical midrange subtropical monsoonal with a mean annual air temperature of 17.2°, mean annual precipitation of 1422 mm in the Botanical Garden and a mean annual air temperature of 16.9°. Mean annual precipitation is 1353.6 mm in the Taolin forestry station. The soil is well-drained clay-loam red soil developed from slate parent rock and classified as Alliti-Udic Ferrosols according to the World Reference Base for Soil Resources (Institute of Soil Science, Chinese Academy of Science, 2001). The depth of the soil layer is deeper than 80 cm, but the content of the soil humus was not rich and pH values range from 4.12 to 4.86. The forests remained unmanaged and all soil-forming factors had remained constant since forest establishment owning to either the foundation of the Botanical Garden or the young age of the forests. Here we used two sites for this experiment due to the difficulties of finding one field site with all these plantations in gradients of forest ages. The principle of field site selection here was to put three plantation types of the same age in one site. Therefore, we think these two sites are suitable for our experiment since our purpose here was to evaluate the admixing effects on soil C and N stocks by comparing mixed forests and corresponding monocultures at the same age rather than comparing soil C and N in forests at different ages. For more detailed information about the experimental site and soil conditions, refer to Wen et al. (2014).

2.2 Soil sampling and laboratory analyses

Four soil samples were randomly collected in each plot using a metal corer (10 cm in diameter) from three depths: 0–10, 10–20, 20–30 cm. Each was treated as one individual sample, then air-dried and sieved through a 0.25 mm sieve. We collected the soil samples down to 30 cm of depth as the soils in this area were susceptible to environmental varia-

| Table 1. Stand characteristics in pure species Pinus stands, pure Cinnamomum stands and mixed Pinus-Cinnamomum stands at age 10, 24 |
|---|
| and 45 years (mean \pm standard deviation). |

| Stand | Age | Species | Density (nha ⁻¹) | Diameter at breast height (cm) | Height (cm) | Basal area $(m^2 ha^{-1})$ |
|----------------------------------|-----|------------|---------------------------------|-----------------------------------|------------------|----------------------------|
| | 10 | Pinus | 2592 | 9.38 ± 3.26 | 5.28 ± 3.97 | 20.06 |
| Pinus stands | 24 | Pinus | 2050 | 14.18 ± 4.34 | 12.86 ± 6.52 | 35.37 |
| | 45 | Pinus | 600 | 21.40 ± 5.30 | 12.47 ± 1.88 | 22.84 |
| Cinnamomum stands | 10 | Cinnamomum | 2708 | 7.77 ± 2.60 | 5.99 ± 1.25 | 14.26 |
| | 24 | Cinnamomum | 900 | 17.02 ± 6.52 | 13.71 ± 2.74 | 23.46 |
| | 45 | Cinnamomum | 800 | 21.06 ± 6.73 | 13.24 ± 2.29 | 30.63 |
| Mixed Pinus–Cinnamomum stands | 10 | Pinus | 902 | 7.64 ± 1.82 | 4.73 ± 0.82 | 4.37 |
| | | Cinnamomum | 1689 | 8.14 ± 2.81 | 7.2 ± 0.73 | 9.83 |
| | 24 | Pinus | 267 | 19.88 ± 5.06 | 12.35 ± 1.64 | 7.80 |
| | | Cinnamomum | 592 | 15.27 ± 5.92 | 11.41 ± 3.13 | 12.45 |
| | 45 | Pinus | 250 | 19.69 ± 4.10 | 12.37 ± 2.60 | 7.91 |
| | | Cinnamomum | 325 | 20.94 ± 8.54 | 13.75 ± 2.79 | 12.91 |

tions and sensitive to the carbon input by litter and fine roots, which matches our purposes of assessing the admixing effects on soil OC and N stocks over time (Wang et al., 2013; Cremer et al., 2016). Soil organic carbon was determined by the wet combustion method through the oxidization of potassium bichromate (Walkley–Black method) followed by titration with 0.5 N ferrous ammonium sulfate solution by using a diphenylamine indicator. Soil total nitrogen was measured using the semimicro Kjeldahl method digested with a mixture of H_2SO_4 , K_2SO_4 , CuSO₄ and Se (Institute of Soil Science, Chinese Academy of Science).

2.3 Data analysis

Soil OC stock (tha⁻¹) and N stock (tha⁻¹) at different soil depths in different stands were calculated with the following formula: $stock = bulk density \cdot depth \cdot [OC or N concentra$ tion]. The effects of the experimental factors, species composition, stand age and soil depth on soil OC, N concentrations and soil OC and N stocks were tested by means of threeway analysis of variance (ANOVA). Differences in soil OC or N concentrations and stocks among mixed P. massoniana and C. camphora stands, pure P. massoniana stands and pure Cinnamomum stands within the same age stages and soil layers were analyzed by using one-way ANOVA followed by a Tukey's test. In order to detect whether the admixing effects were additive or nonadditive, an alternative analytical method was used as suggested by Ball et al. (2008). Expected values of OC and N stock in mixed stands at different stand ages in different soil depths were calculated by adjusted values based on basal area in monocultures with the following formula:

expected value = $(Ba_{p.mix}/Ba_{p.pure})$ •Stock_{p.pure}

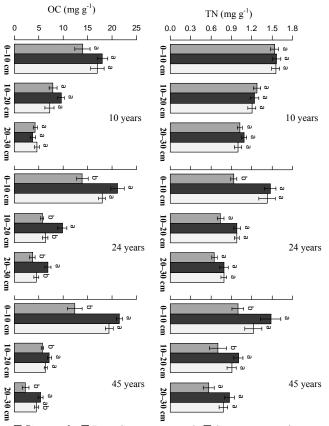
+ $(Ba_{c.mix}/Ba_{c.pure}) \cdot Stock_{c.pure}$,

where Bap,mix is the basal area of Pinus in the mixed stand, Bap.pure is the basal area of Pinus in the pure Pinus stand, Bac.mix is the basal area of C. camphora in the mixed stand, Bac.pure is the basal area of Cinnamomum in the Cinnamomum pure stand, Stock_{p.pure} is the soil OC or N stock under the pure Pinus stand and Stock_{c.pure} is the soil OC or N stock under the pure Cinnamomum stand. All the calculations above are conducted in the same given stand age (10, 24 or 45 years old). These expected values for OC or N stock were then compared with observed values for each individual sample that were measured experimentally in mixed stands with the following formula: (observed-expected)/expected. For soil OC and N stocks at specific soil depths at given stand ages, 95% confidence intervals (CI) were calculated with the above formula. If the CIs for mixtures did not cross y = 0, the admixing effect was considered nonadditive (Ball et al., 2008). Otherwise, we regard the admixing effect as additive. All the statistical analyses were conducted with statistical software from the R project (R 3.0.2; R Development Core Team, 2013).

3 Results

3.1 Soil organic carbon and nitrogen stocks

The three-way ANOVA indicated that the forest stand types, soil depths and their interactions exerted significant influence on the soil OC and N concentrations, while the age effects were not significant for soil OC concentration (Table 2). When compared with parallel forest stands with the

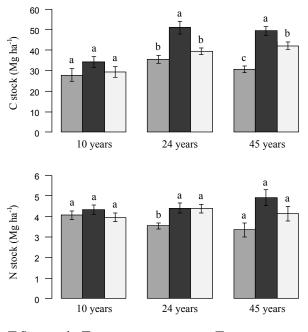


■ Pinus stands ■ Pinus-Cinnamomum stands □ Cinnamomum stands

Figure 1. Soil organic carbon (OC) concentration and nitrogen (N) concentration in pure *Pinus*, *Cinnamomum* and mixed *Pinus–Cinnamomum* stands in 0–10, 10–20 and 20–30 cm of soil depth at the age of 10, 24 and 45 years. Error bars indicate standard errors. Different letters indicate significant differences among different stands within the same soil profile and age stage (p < 0.05).

same age, soil OC and N concentrations were the highest in *Pinus–Cinnamomum* mixed stands for almost all cases in the whole soil profile or in the specific soil layers, but significant differences were only detected in 24- and 45-year-old stands (P < 0.05) (Fig. 1).

Total soil OC stock was highest in *Pinus–Cinnamomum* mixed stands (44.86 Mg ha⁻¹) compared to corresponding monocultures at the same stand age in all the soil profiles investigated followed by the evergreen broad-leaved *Cinnamomum* stand (36.37 Mg ha⁻¹), and the conifer *Pinus* stand showed the lowest values (31.51 Mg ha⁻¹). Significant differences were detected in 24- and 45-year-old stands but not in 10-year-old stands (P < 0.05). Along the chronosequence, soil OC stocks increased with increasing stand age in these three forest types with a mean value of 30.50, 41.96 and 43.85 Mg ha⁻¹ in 10-, 24- and 45-year-old stands, respectively (Fig. 2). When we take a closer look at the stratification distribution of the soil OC stocks in these three forest types with a single stand stands.



□ Pinus stands □ Pinus-Cinnamomum stands □ Cinnamomum stands

Figure 2. Total soil organic carbon (OC) and nitrogen (N) stocks in pure *Pinus*, *Cinnamomum* and mixed *Pinus–Cinnamomum* stands in 0–30 cm of soil depth at the age of 10, 24 and 45 years. Error bars indicate standard errors. Different letters indicate significant differences among different stands with the same age (p < 0.05).

OC stocks decreased significantly with increasing soil depth and a similar pattern: the highest OC stock in mixed stands over monocultures was observed within given soil layers at given stand ages. The over-performance of OC stock in Pinus-Cinnamomum mixed stands compared to the counterpart monoculture stands with the same age was mainly attributed to the top 10 cm of soil depth (Fig. 3). In 0-10 cm soil layers, OC stocks under mixed stands showed no significant differences compared with individual stands in the young stands. Those under mixed stands significantly exceeded the Pinus individual stands in middle-aged stands and finally exceeded both individual stands in the oldest stands. In the 10–20 cm soil layers, both 10- and 45-year-old stands exerted no significant differences in soil OC stock among three stand types, while in 24-year-old stands the mixture had a significantly higher soil OC stock than the monocultures. In the 20-30 cm soil layers, mixed stands showed similar soil OC stock patterns when compared with corresponding monocultures, and in 24-year-old stands the mixture showed a significantly higher soil OC stock than two pure Pinus and Cinnamomum stands.

Soil depth and stand type, but not stand age, exerted significant effects on soil N stock in mineral soil (Table 2). Within the same stand, the soil N stocks decreased with soil depth, and among them, the superficial soil layers (0-10 cm) exhibited significantly higher soil N stocks than the other two

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| Factor | OC concentration | | N concentration | | OC stock | | N stock | |
|-----------------------------------|------------------|---------|-----------------|---------|----------|---------|---------|---------|
| | F value | P value | F value | P value | F value | P value | F value | P value |
| Stand | 24.30 | <0.0001 | 11.89 | <0.0001 | 36.73 | <0.0001 | 15.17 | <0.0001 |
| Age | 0.00 | 0.9942 | 60.53 | <0.0001 | 35.99 | <0.0001 | 0.54 | 0.5814 |
| Depth | 699.57 | <0.0001 | 205.33 | <0.0001 | 489.87 | <0.0001 | 65.62 | <0.0001 |
| Stand \times age | 2.07 | 0.1283 | 10.50 | <0.0001 | 2.87 | 0.0236 | 3.92 | 0.0042 |
| Stand \times depth | 8.61 | 0.0002 | 2.39 | 0.0937 | 5.53 | 0.0003 | 1.15 | 0.3354 |
| Age \times depth | 2.00 | 0.1588 | 0.05 | 0.8163 | 16.79 | <0.0001 | 4.18 | 0.0027 |
| Stand \times age \times depth | 0.15 | 0.8579 | 0.73 | 0.4834 | 1.03 | 0.4142 | 0.67 | 0.7217 |

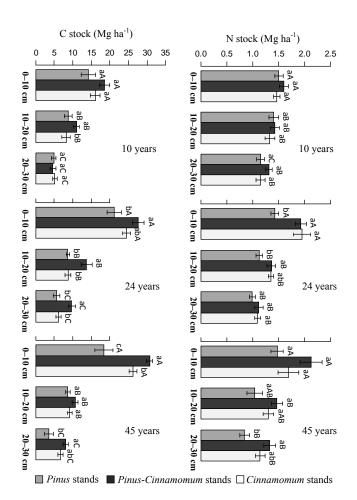


Figure 3. Soil organic carbon (OC) and nitrogen (N) stocks in pure *Pinus, Cinnamomum* and mixed *Pinus–Cinnamomum* stands in 0–10, 10–20 and 20–30 cm of soil depth at the age of 10, 24 and 45 years. Error bars indicate standard errors. Different small letters indicate significant differences among different stands within the same soil profile and age stage (p < 0.05). Different capital letters indicate significant differences among different soil layers within the same stand at a given stand age (p < 0.05).

deeper soil layers (Fig. 3). Only in the 0–10 and 10–20 cm soil layers of 24-year-old stands and the 20–30 cm soil layer of 45-year-old stands did N stock significantly differ among stand types. Pure *Cinnamomum* and *Pinus–Cinnamomum* mixed stands always had similar N stocks, which were higher than *Pinus* stands. The N stock in the topsoil of mixed stands increased along the chronosequence for all stands, except in the *Cinnamomum* stand, which marginally decreased from the 24- to the 45-year-old stand. In the 10–20 cm soil layer of the *Pinus* stand, N stock increased from the 10- to the 24-year-old stand and then stayed stable. All the other soil layer stand in the stand type (Figs. 2 and 3).

3.2 Admixing effects on soil OC and N stocks

The relative performance of soil OC and N stocks in the Pinus-Cinnamomum mixed stands was calculated to determine the additive or nonadditive effects for each stand at different soil layers and different stand ages by comparing observed soil OC or N stock values with expected values based on counterpart monocultures. In our study the mixed planting always exerted positive effects, as all the relative values of soil-sequestrated OC and N stocks were positive in all the soil profiles in the mixed stands. Almost all the CIs did not cross y = 0, suggesting that these positive admixing effects were strongly nonadditive, except the relative OC sequestration in the 20-30 cm soil layers of 10-yearold stands. In the 10-year-old stand, the over-performance of soil OC sequestration was mainly attributed to the topsoil and subsoil layers, and these positive effects were stable in the two upper soil layers and then decreased in the deepest layer. In the 24-year-old stand, it increased with soil depth. In the 45-year-old stand, the relative percentage was the highest in the deepest soil layer (20-30 cm). Overall, the percentage of over-performance in mixture was significantly higher in the 24-year-old stand than in the 10-year-old stand, and then it marginally decreased in the 45-year-old stand. For soil N stock, however, the results showed a consistent pattern

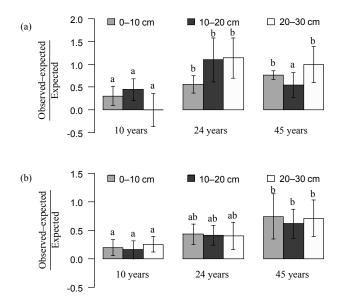


Figure 4. Investigation of additive or nonadditive interactions for soil OC stock (**a**) and N stock (**b**) in *Pinus–Cinnamonum* mixed stands in 0–10, 10–20 and 20–30 cm of soil depth at the age of 10, 24 and 45 years. Observed values were compared to expected values calculated as the average value in monocultures of *Pinus* and *Cinnamonum*. Error bars represent 95 % CI, and mixtures for which the CIs do not cross y = 0 are considered to be significantly nonadditive. Different letters indicate significant differences among different stand ages within the same soil depth profile.

that the positive effects increased with increasing stand ages (Fig. 4).

4 Discussion

4.1 Soil organic carbon and nitrogen stocks

The soil organic carbon stocks under the three stand types always followed this order: mixed Pinus-Cinnamomum stand > Cinnamomum stand \geq Pinus stand. An exception was those in the 10-20 and 20-30 cm soil layers of the 10-yearold stand, which showed slight differences. Many researchers have demonstrated the species diversity or species identity effects on soil OC and N accumulation in forests and revealed that species diversity and/or abundance of dominant tree species exerts effects on OC and N stocks through carbon input by litter and fine roots (Berger et al., 2002; Guckland et al., 2009; Wang et al., 2013; Dawud et al., 2016). In a previous study at this site, the annual litterfall was also the highest in the mixed Pinus-Cinnamomum stand followed by Pinus; Cinnamomum was the lowest (Xu et al., 2013). Therefore, the higher soil organic carbon under mixed stands may be attributed to the higher annual litterfall in mixed stands. The higher soil OC in the Cinnamomum stand compared to Pinus is likely connected with the litter decomposition rate, as the needle leaves of *Pinus* accumulated on the forest floor with a lower composition rate. The higher litter input may not necessarily increase the soil carbon content (Fontaine et al., 2004), although many previous studies have also shown larger carbon stocks under coniferous species than broad-leaved species (Kasel and Bennett, 2007; Schulp et al., 2008; Wang et al., 2013). With increasing stand age, the gap in soil OC stock between the Cinnamomum and Pinus stands became more pronounced in all soil profiles investigated. Furthermore, when compared with broadleaf tree species, conifer species tended to allocate much more total organic matter production to aboveground growth, which features them as fast-growing tree species and caused a lower direct carbon input to soil (Cuevas et al., 1991). In this study, the highest OC stock presented in 24-year-old stands; in the Cinnamomum stands, though not significant, carbon stock still had an increasing trend from 24- to 45-year-old stands. In mixed stands, though with a relatively smaller portion of Cinnamomum, the trend in soil OC stock is similar to Cinnamomum stands but not the Pinus stands. This confirmed that broadleaf tree species are ahead of conifers in long-term growth.

The mixed stand exhibited the highest soil N concentrations and N stocks, revealing the priority of soil N accumulation in mixed Pinus-Cinnamomum stands compared to corresponding monocultures. Compared to the mean value of the counterpart monoculture at the same age, the soil N stock in mixed stands increased by 8.30, 11.17 and 31.45%, respectively, suggesting that these admixing effects were more pronounced with stand ages along the chronosequence. Whether this enhanced the admixing effect on soil N accumulation will continue needs further investigation. In monocultures, the Pinus stand showed priority in earlier stages, while Cinnamomum showed priority in the later stage in terms of soil N stocks. This is similar to the trend in soil OC stocks but less pronounced. A large amount of soil nitrogen us stored in soil organic matter, and with the decomposition of soil organic matter, nitrogen will be released and able to be taken up by plants or leaching, as reported in our previous study. Soil nitrogen concentration positively correlated to soil organic carbon concentration (Wen et al., 2014), and thereby the soil nitrogen stocks to some degree shared similar patterns of soil OC stocks under the same circumstances. As Hume et al. (2016) discussed, the accumulation of soil nitrogen may be slower than carbon because nitrogen is progressively locked up in live biomass, which is likely the explanation for why the soil nitrogen stock changed less significantly with stand ages and between different stand types compared with soil OC stocks here.

In the uppermost soil layer, soil organic carbon stocks under the three stand types all significantly increased from 10- to 24-year-old stands and then became stable thereafter; those of the *Pinus* stands slightly declined from 24- to 45year-old stands without significant differences. This trend is inconsistent with a previous study that reported a decline in

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soil organic carbon in the topsoil (0-10 cm) within a certain period of time after plantation establishment (Turner and Lambert, 2000). However, it is consistent with the results from a Chinese fir plantation where the soil OC stocks increased with increasing stand ages from about 10 to 20 years and then stabilized (Chen et al., 2013). Most similar research that focuses on the changes in organic carbon stocks along the chronosequence has suggested that soil organic carbon stocks in topsoil reach a stable level after approximately 20 years (Tremblay et al., 2006; Chen et al., 2013). This can be interpreted as the formation of a balance that depends on the carbon input and organic decomposition (Hume et al., 2016). In the initial stage of afforestation, the changes in organic carbon storage differed with increasing stand age. The conversion of plantations into natural native forests will always present a decrease in soil organic carbon in topsoil within 10 years, and then some will rebound and increase until reaching an equilibrium level (Chen et al., 2013); the remaining amount will continue decreasing to the lowest level (Turner and Lambert, 2000). In our results the soil OC and N stocks decreased significantly with increasing soil depth. However, the magnitude of over-performance in the Pinus-Cinnamomum mixed stands over monocultures increased with soil depth (Figs. 3 and 4). Topsoils under forests are always the most susceptible to disturbance, and they can be directly impacted by carbon input through litter and fine roots, which always decline with soil depth (Wang et al., 2016). Our data for the same sites also suggested that fine roots mostly assemble in the upper soil, and fine-root overyielding occurred in the topsoil layer (0-10 cm; see Table S1 in the Supplement). Also, in our previous study (Wen et al., 2014), the ratio of soil microbial biomass to soil organic carbon was the lowest in topsoil, which may suggest a lower mineralization rate in deeper soil and make the accumulation of OC and N stocks in deeper soil along the chronosequence more pronounced over the topsoil layer. Here we only collected the soil down to 30 cm of depth, and the effects of species diversity and species identity on the deeper soil merits further investigation to improve model parameters of soil OC and N processes in the deep soil profile.

4.2 Admixing effect along the chronosequence

In the two topsoil layers (0–10 and 10–20 cm), the admixing effect on soil OC was more pronounced along the chronosequence. This likely accounted for the increasingly intensive interactions between the two species with increasing stand age. In the pure stands, intraspecific competition also become more intensive along the chronosequence, which will block carbon sequestration because of nutrient and water limits and favor the mixed forests where the interspecific competition is relatively less intensive in general (Lei et al., 2012a). Regarding the additive effect shown in the 20–30 cm soil layer under 10-year-old mixed stands, the observations were almost equal to the expected values, which confirmed is the space occupied by belowground and aboveground tree biomass was not big enough to exert significant influence when the stands are relatively young or the period of soil OC accumulation processes is relatively short. Our results also showed, to some degree, higher admixing effects in the deeper soil layer that may contribute to the lower expected values in monocultures in the deeper soil layer, which makes the (observed – expected)/expected values more sensitive to the increment of observed values.

The admixing effects of soil N stocks in the whole profile suggested a consistent increasing trend in the whole soil profile. Fine roots in mixed stands compared with pure stands are always assumed to exploit deeper soil (Cremer et al., 2016), so they may assemble more nitrogen from deeper soil (even deeper than our samples) to upper soil. Also, Lei et al. (2012b) reported higher fine-root turnover caused by higher fine-root production of mixed stands, which potentially increases nitrogen input in mixed stands. In addition, mixed stands are more resistant to environmental disturbances, are more capable of N retention and prevent leaching in soil (Tilman et al., 1996). This merits further studies in more diverse communities in forests to confirm this pattern of higher nitrogen stock under mixed stands over monocultures and these increasing positive effects along the chronosequence.

5 Conclusions

The tree species and composition as well as the stand age significantly affect soil organic carbon and nitrogen stocks. Converting pure stands into mixed stands can significantly enhance soil OC and N stocks. This positive admixing effect becomes more pronounced along the chronosequence for OC only in topsoil, while inconsistent trends present in the deeper soil. However, for soil N stock, it becomes more pronounced along the chronosequence in the whole soil profile. Tree species identification also affects the soil OC sequestration and N stock. In topsoil, *Cinnamonum* stands always contain more soil OC and N stocks than *Pinus* stands due to the different strategies of carbon and nutrient allocation and different rates of organic decomposition, but these differences are less pronounced in deeper soil layers.

Data availability. The fine root biomass data are available in the Supplement, and the full data set for soil carbon and nitrogen that we used is available upon request from the corresponding author (Pifeng Lei, pifeng.lei@outlook.com).

The Supplement related to this article is available online at https://doi.org/10.5194/bg-14-3937-2017-supplement.

Competing interests. The authors declare that they have no conflict of interest.

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