Effects of Stand Density on Soil Respiration and Soil Labile Organic Carbon and Their Influence Mechanism in Larix Principis-Rupprechtii Plantations

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Research

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Abstract

Background

Forest soil carbon pool plays a vital role in the global carbon sequestration and carbon emission. Forest management can regulate the sequestration and output of forest soil carbon pool to a certain extent, but mechanism of forest density effects on soil carbon pool still needs to be further researched.

Methods

We established sample plots with density gradients in three-age stands of Larix principis-rupprechtii plantation and measured soil respiration (RS), soil organic carbon (SOC), soil dissolved organic carbon (DOC), and microbial biomass carbon (MBC), light fraction organic carbon (LFOC), and easily oxidizable organic carbon (ROC).

Results and Conclusions

The results showed that, among the forest stands of three ages, RS, heterotrophic respiration (RH), MBC, LFOC, ROC of different stand density levels were significantly different. Moderate density promotes RS rate and RH rate and the sequestration of MBC and LFOC and inhibits ROC sequestration. With the increase of forest stand density, RS, RH, LFOC, and MBC first increased and then decreased, and ROC first decreased and then increased, the quadratic function could fit these changing trends. The RS, RH, and autotrophic respiration (RA) rates of older forest stands were relatively fast, and contents of SOC, MBC, LFOC, DOC, and ROC were higher, and they were more sensitive to changes in stand density. SOC, LFOC, MBC, DOC, and ROC explained 56.05% variations of RS, Rh, and RA. MBC, LFOC, and ROC in soil labile organic carbon were closely related to RS and Rh, but not SOC. Among them, LFOC and MBC played the role of "warehouse" and "tool" and significantly correlated with RS and Rh. ROC, as "raw material," had a significant negative correlation with RS and RH. When the RS and RH rate were fast, ROC maintained a dynamic and stable state of low soil content. Stand density could regulate RH by affecting soil labile organic carbon, an essential path for stand density to regulate soil respiration. Given soil carbon pool significance in forest ecosystems, Continuous research on soil respiration and stand density is suggested to bridge the gaps in our comprehension of the Regulation of Forest Management on forest soil carbon pool.

1. Introduction

Forest is the largest ecosystem which has tremendous amount of carbon sequestrated by plants (Franklin et al., 2009; Poorter et al., 2016; Khan et al., 2018). The soil carbon pool, which is the second largest carbon pool after aboveground forest carbon in the world, cannot be ignored (Sedjo, 1993; Lal, 2005). The carbon exchange between the soil and the atmosphere largely affects the global carbon cycle and climate change all the time (Dib et al., 2014; Tian et al., 2016; Gabriel et al., 2018). The soil respiration is the gas exchange between soil and atmosphere (Wei et al., 2010; Goldberg et al., 2017) and at the same time, soil organic carbon is also stored in forest soil in various forms and plays different roles (Tian et al., 2016; Jílková, 2020). Thus, understanding the characteristics of forest soil respiration and carbon sequestration is important for managing forest ecosystem. Through the proper management of forests, processes of greenhouse gas emission and soil organic carbon sequestration may be controlled to a certain extent.

The stand density, which is usually regulated by thinning, is considered as an indispensible influence on the forest production. The principle of thinning maybe forming the local climax plant community as the basis of forest management research (Jens et al., 2000; Ming et al., 2018), which may be also based on the integrity principle of the forest ecosystem. Therefore, the effect of stand density is not only limited to the change of canopy structure and vertical forest structure (Jack and Long, 1991; Liu et al., 2019), but also to the change of microclimate, interspecific competition, and so on (Shao and Shugart, 1997; Ali et al., 2019; Bello et al., 2019; Eldegard et al., 2019; Liu et al., 2019).

In the previous studies, it was considered that soil respiration (RS) is composed of autotrophic respiration (RA) and heterotrophic respiration (RH) whereby decomposition of microorganisms and the turnover of roots in the soil are the main forms of heterotrophic respiration and autotrophic respiration, respectively (Baggs, 2006; Hopkins et al., 2013; Xu and Shang, 2016). This process provides better understanding on the mechanism of soil respiration and the influence of various factors on soil respiration. These studies, which were carried out on the forest soil within two years of thinning of the Carpinus betulus plantation, have reported significantly higher soil microbial respiration of the thinned plots compared to the control plots, and no significant difference was reported.
between soil respiration among the three thinning intensities (Akburak and Makineci, 2016). In the study of mature Masson pine forests, Leilei reported that thinning could effectively increase the rate of soil respiration in a short period (Lei et al., 2018). When studying the soil respiration of Shanxi *Pinus tabulaeformis* young forests that have been thinned, it is reported that moderate thinning positively affected soil respiration by changing soil temperature and humidity (Cheng et al., 2015). Similar results are also reported elsewhere (Zhang et al., 2018). Many researchers found that moderate thinning could make the stand microclimate changed, thereby optimizing the living environment of soil microorganisms, and reserve the sufficient soil substrates and soil organic carbon for respiration. Few researchers have also reported different results, for example, thinning could cause the death of plant roots and reduce the autotrophic respiration of the soil and much sparse stands would reduce the activities of soil microorganisms(Park et al., 2009; Mosca et al., 2017).

Soil organic carbon (SOC) is the C contained in soil organic matter (SOM), which is an important indicator of measuring soil carbon sequestration (Lull et al., 2020), and this substantially affected by land use changes, forest management, natural and human interference, and so on. Forest soil organic Carbon is attributed to strong dynamic changes (Cambardella and Elliott, 1992; Liang et al., 1997). However, due to the complexity of the composition, structure, and existence of soil organic matter, the performance of a certain functional characteristic of the soil is often the result of a chemical mixture’s simultaneous action with similar chemical components, structural characteristics, and functional groups. It is not the total amount of soil organic carbon that characterizes the soil carbon pool activity, but the soil labile organic carbon. The degree of activity is not only the total amount of soil organic carbon, but also the soil labile organic carbon. The active component of soil organic carbon is the most active and unstable C in the soil, which has the characteristics of availability, easy oxidation, and solubility. Generally, soil carbon may be grouped based on stability of SOC and measure soil dissolved organic carbon (DOC), such as microbial biomass carbon (MBC), light fraction organic carbon (LFOC), and easily oxidizable organic carbon (ROC). This is considered as the most quantitative expression of soil labile organic carbon (LOC) (Hu et al., 2010).

It has been reported that the decrease of stand density increases soil temperature, humidity, soil respiration, soil SOC, N, P, K, etc. (Wic Baena et al., 2013; Zhang et al., 2018). However, many studies have found that the changes of soil temperature, humidity, and soil respiration by thinning could become stable after certain period of thinning, and even return to the original level (Fernandez et al., 2012; Olajuyigbe et al., 2012; Bai et al., 2016). These studies also reported that the root system or the plant remaining in the stand after thinning acted as the exogenous carbon, which led to the change of soil rather than the effect of stand density. Most of the existing studies focus on the short-term “stress response” of forest soil carbon pool after thinning (Ryu et al., 2009; Bolat, 2013; Zhao et al., 2019). Although thinning directly changes the stand density, the impact of stand density on the soil should be the steady state achieved by biochemical action under the influence of different stand densities. The effect of stand density on the soil carbon pool is caused by the interaction of litter return, root growth and respiration, microbial activity, and mineral turnover after thinning. Our study intends to explore more on the effect of different stand density on the soil respiration and soil labile organic carbon and their mechanism.

This is not clear whether the soil carbon pool of different age plantations would have different responses to different stand densities. This study considers paying enough attention to the factors of forest development and growth in carrying out more effective human intervention and forest management on the forests of all ages. As the substrate of soil respiration, soil organic carbon cannot not be discussed in isolation, the hypothesis presented in this study was that both stand density and stand age are essential factors influencing soil respiration, soil organic C and soil labile organic C, and a close correlation between soil labile organic C and soil respiration. Therefore, objectives of this study were to: 1) Examine whether RS, RH, RA and SOC, MBC, DOC, LFOC, ROC in mineral soil would be significantly affected by stand density and stand age during the studied period (Five years after thinning); 2) Find out the trend of the variables mentioned above with the stand density, and compare the trends for different stand ages; 3) Identify the crucial factors shaping the RS, RH and RA in soil organic carbon pools.

### 2. Materials And Methods

#### 2.1 Study area

The study area is located in Yeshagou (37 ° 44 ′ N, 111 ° 30 ′ E) of Xiaowenshan forest farm in Pangguangou Nature Reserve, Lvliang City, Shanxi Province, China, with an altitude of 1760–2210 M (Fig. 1). The climate of this area is temperate continental monsoon climate, cold and dry in winter and hot and humid in summer. The annual average temperature is 4.2 °C, the average yearly
precipitation is 822.6 mm, and the annual average relative humidity is 70.9%. The soil is leached cinnamon soil with a humus layer thickness of 3–7 cm. In the study area, the forest types were mainly pure *Larix principis-rupprechtii* forest and *Pinus tabulaeformis* plantation, occasionally accompanied by a few *Betula platyphylla* and *Quercus liaotungensis*; the main shrub species are *Spiraea salifolia*, *Rosa xanthina*, and *Lespedeza bicolor*.

Toward the end of April 2020, we selected sub-compartments with similar site conditions, slopes, and slope positions of different ages, with an area of more than 2 ha, according to the average height of dominant trees (*Larix principis-rupprechtii*), and the age of sub-compartments were 27-(27a), 36- (36a), and 48-year-old (48a), respectively. In order to prevent the human interference and short-term effect after thinning, the latest thinning operation was conducted sub-compartment on 2015 and another thinning on the same area on 2020. Based on the stand density difference formed by thinning, nine standard sample plots (20 m x 20 m) with different stand densities were laid out in the stands with different ages (stand density calculated according to the actual density on the sample plot), and a 5 m wide buffer zone was set around the sample plots. A total of 27 sample plots were laid out with inter-plot distance limited to 40 m. We set three sample plots with similar stand densities as replications, dividing the nine sample plots of each age into three density levels, namely high density (HD), medium density (MD), and low density (LD). We measured the trees with DBH greater than 5 cm in each plot, and record their DBH, height, and other parameters.

### 2.2 Field survey and soil sampling

When laying out the sample plot, we randomly placed six polyvinyl chloride (PVC) material rings with inner diameters of 20 cm and heights of 10 cm within the sample plot, and driven them into the soil by about 6 cm with a hammer. Three of them were treated with root removal (trench method) to distinguish the autotrophic breathing from the heterotrophic breathing. The root removal method involved selecting an area of size 0.5 m × 0.5 m and digging trenches vertically to a depth of 0.6–0.8 m around this area with a shovel (up to the depth of the root system accessed), cutting off the roots (but not removing) and inserting the nylon net with 100 mesh for preventing root growth (Kuzyakov, 2006). The vegetation of the sample plot was clipped and all the trench-digging sample plots were regularly cleaned for surface vegetation and litter to distinguish autotrophic respiration and heterotrophic respiration.

In order to avoid the influence of soil disturbance on the soil respiration, soil respiration was measured twice in mid-July 2020. Soil carbon flux automatic measurement system (Li-cor 8100a, Li-COR, Inc., Lincoln, Nebraska) was applied to measure soil respiration three times on each respiratory ring. The measurement was made between 9:00 am to 15:00 whereby the soil respiration of the same age and different density was also measured in the same period. A total 972 measurements of 162 respiratory rings were made. The soil was sampled at the depth of 0–10 cm, 10–20 cm, and 20–30 cm by using a 5 cm circular soil auger. Three samples from each soil layer were collected, which resulted in 9 soil samples per sample plot, and 243 soil samples in total. The collected soil samples were put into the numbered aseptic soil bags and stored at low temperature, and brought back to the laboratory. The fine roots and stones were removed from the soil samples, sieved through 2 mm holes, and divided into two parts. One sample was fresh and stored in a 4 °C refrigerator to determine MBC and DOC; other samples collected were dried in the laboratory to determine soil physical and chemical properties, and other labile organic carbon components (SOC, ROC, LFOC). The summary characteristics of the sample plots are presented in Table 1.

### Table 1

**The summary characteristics of the sample plots.** SWC, soil moisture content; BD, soil bulk density; TN, soil total nitrogen content; TP, soil total phosphorus content. Values are as the mean ± standard deviation (SD). Different lowercase letters (a, b, and c) indicate significant differences (p < 0.05) among different thinning treatments. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.
| Stand Age (year) | Elevation (m) | Stand Density Level | Stand Density (trees/ha) | Aspect | Slope (°) | DBH (cm) | Height (m) | SWC (%) | BD (g·cm$^{-3}$) | pH | TN (g/kg) | TP (g/kg) |
|-----------------|--------------|---------------------|--------------------------|--------|-----------|---------|-----------|--------|----------------|-----|-----------|-----------|
| 27a             | 1854         | LD                  | 975                      | N10°E  | 13        | 18.79± 3.09 | 14.38± 1.45 | 29.16± 3.83a | 1.04± 0.03a | 7.02± 0.16a | 2.28± 0.10a | 0.79± 0.04a |
| 1860            | 1100         |                     |                          |        | 14        | 18.16± 2.86 | 14.43± 1.32 |          |                   |     |           |           |
| 1865            | 1250         |                     |                          |        | 12        | 18.23± 2.10 | 13.69± 1.09 |          |                   |     |           |           |
| 1860 MD         | 1500         |                     |                          |        | 12        | 18.09± 2.05 | 14.77± 0.87 |          |                   |     |           |           |
| 1864            | 1675         |                     |                          |        | 11        | 17.86± 2.36 | 13.73± 1.99 |          |                   |     |           |           |
| 1870            | 1825         |                     |                          |        | 12        | 18.01± 3.10 | 14.24± 1.84 |          |                   |     |           |           |
| 1868 HD         | 2125         |                     |                          |        | 10        | 17.15± 2.21 | 13.85± 2.01 |          |                   |     |           |           |
| 1880            | 2300         |                     |                          |        | 18        | 15.87± 2.77 | 13.44± 1.38 |          |                   |     |           |           |
| 1885            | 2425         |                     |                          |        | 14        | 16.26± 2.03 | 14.04± 3.32 |          |                   |     |           |           |
| 36a             | 2030         | LD                  | 525                      | N40°E  | 18        | 24.60± 3.65 | 17.15± 2.13 | 31.76± 5.36a | 1.09± 0.03a | 6.98± 0.11a | 3.4± 0.51b | 0.87± 0.06a |
| 2050            | 750          |                     |                          |        | 13        | 22.57± 2.56 | 16.93± 1.34 |          |                   |     |           |           |
| 2054            | 925          |                     |                          |        | 20        | 21.92± 3.14 | 16.62± 1.66 |          |                   |     |           |           |
| 2043 MD         | 1100         |                     |                          |        | 15        | 20.41± 2.42 | 15.98± 2.07 |          |                   |     |           |           |
| 2052            | 1325         |                     |                          |        | 12        | 19.96± 3.77 | 16.49± 1.83 |          |                   |     |           |           |
| 2063            | 1500         |                     |                          |        | 15        | 20.89± 2.75 | 16.03± 1.98 |          |                   |     |           |           |
| 2073 HD         | 1550         |                     |                          |        | 16        | 19.71± 3.01 | 15.76± 2.61 |          |                   |     |           |           |
| 2066            | 1700         |                     |                          |        | 16        | 18.12± 3.16 | 16.08± 2.15 |          |                   |     |           |           |
| 2070            | 2075         |                     |                          |        | 17        | 18.37± 2.98 | 16.36± 1.97 |          |                   |     |           |           |
| 48a             | 2122         | LD                  | 400                      | N20°E  | 13        | 29.41± 3.28 | 19.45± 2.13 | 38.08± 3.47b | 1.13± 0.06a | 6.93± 0.14a | 2.95± 0.48c | 1.10± 0.33b |
| 2128            | 600          |                     |                          |        | 15        | 29.78± 3.43 | 18.93± 1.34 |          |                   |     |           |           |
| 2140            | 725          |                     |                          |        | 26        | 27.52± 3.27 | 18.62± 3.66 |          |                   |     |           |           |
| 2160 MD         | 875          |                     |                          |        | 18        | 28.06± 2.54 | 18.58± 2.07 |          |                   |     |           |           |
| 2162            | 975          |                     |                          |        | 12        | 27.16± 3.00 | 18.49± 2.83 |          |                   |     |           |           |
### 2.3 Soil sample analysis

The physical and chemical properties, such as soil moisture content, bulk density, and pH were measured. TN was measured by the Kjeldahl method, and TP was measured by the sulfuric acid digestion-colorimetric method (Qiu et al., 2019).

The SOC was measured by potassium permanganate external heating method; ROC was measured by potassium permanganate oxidation colorimetric method, soil dissolved organic carbon and soil microbial carbon were measured by chloroform fumigation and K$_2$SO$_4$ extraction method. Then, the TOC/TN analyzer (multi N/C 2100, Jena) was used to measure, LFOC was separated by the NAI solution with a specific gravity of 1.7 kg/L and measured by the potassium permanganate external heating method (Hu et al., 2010).

### 2.4 Data processing

We calculated RA as below:

$$R_A = R_S - RH \quad (1)$$

where RA, autotrophic respiration; RS, soil respiration; RH, heterotrophic respiration (obtained by trench method).

We calculated MBC as below:

$$MBC = C_U - C_{uf} / K \quad (2)$$

where CU and NU are the carbon and nitrogen contents of fumigated soil extract, K is the conversion coefficient of fumigated extraction method (0.45) (Brookes et al., 1985)

Based on the testing normality and consistency of the data, the statistical analysis was carried out the multi-factor analysis of variance, linear and quadratic regressions, and structural equation modeling, correlation analysis, and redundancy analysis using R4.0.3 (R Development Core Group), and all the illustrations in the article were made using this software version.

### 3. Results

#### 3.1 Factors affecting SOC, soil labile organic carbon, and soil respiration

We used the multi-factor analysis of variance to test the factors that may affect RS, SOC, and labile organic carbon. Stand density significantly affected RS, RH, and RA (Table 2). Stand age had no significant effect on soil respiration and its components. The interaction between stand density and age had no significant effect. The SOC and labile organic carbon were significantly affected by both the stand density and stand age. Except for DOC and LFOC, other organic carbon measures were not much sensitive to the interaction effects of the stand density and stand age, meaning that regulation effect of stand density on the soil respiration was significant, and the sequestration state of SOC and labile organic carbon components were significantly affected by stand density and stand age.

Table 2.
Analysis results of factors affecting RS, RH, RA and soil labile organic carbon. In the table * represents statistical significance, in which, P < 0.1; ** P < 0.05; *** P < 0.01; **** P < 0.001. RS, soil respiration; RH, heterotrophic respiration; RA, autotrophic respiration.

| Factor | Stand density | Stand age | Stand age × Stand density |
|--------|---------------|-----------|---------------------------|
|        | F-value | P-value | F-value | P-value | F-value | P-value |
| RS     | 8.002   | 0.0101 * | 2.488   | 0.1072 | 1.323  | 0.2876 |
| RH     | 12.987  | 0.00167 ** | 2.577   | 0.09978 | 0.691  | 0.51231 |
| RA     | 0.038   | 0.847 | 2.411   | 0.114 | 0.958  | 0.4 |
| SOC    | 151.631 | 4.52e-11 *** | 72.849  | 3.57e-10 *** | 0.722  | 0.498 |
| ROC    | 3.174   | 0.08928 . | 9.225   | 0.00133 ** | 0.24  | 0.78852 |
| MBC    | 18.382  | 0.000327 *** | 12.163  | 0.000310 *** | 0.433  | 0.654519 |
| DOC    | 364.062 | 9.59e-15 *** | 469.313 | <2e-16 *** | 3.501  | 0.0487 * |
| LFOC   | 67.413  | 5.42e-08 *** | 24.824  | 2.94e-06 *** | 4.059  | 0.0323 * |

3.2 Effects of stand density on soil respiration

We analyzed the differences of RS, RH, RA between different density levels. We use the advantages of density sequence diagrams to discover the trend of these indicators with stand density and try to find the mechanism of stand density on soil respiration.

3.2.1 Difference of RS, RH, RA of different stand density levels

For the 27a stand, the RH of MD was significantly higher than that of HD and slightly higher than LD; the RS of MD was slightly higher than that of LD and HD; the RA of HD was the highest, followed by MD and LD was the lowest (Fig. 2A). For the 36a stand, the RH of MD was significantly higher than that of HD and slightly higher than LD; the RS of MD was significantly higher than the other two grades; there was no significant difference in RA (Fig. 2B). For the 48a stand, the RS of MD was significantly higher than HD and LD; RH was slightly higher than LD and higher than HD, RA of MD was significantly higher than LD and HD (Fig. 2C).

3.2.1 Variation of RS, RH, RA for stands with different ages with stand density levels

We attempted to characterize the variations of RS, RH, and RA for the stands with stand density and ages using regression analyses (Fig. 3). The quadratic function was used to fit the changing trend of RS, RH, and RA at different ages, and this function fitted the data well, describing stand density variations for RS and RH by more than 50% (R^2 > 0.5). The older the stand age, the higher was the RS and RH rate. However, when the stand density was too large, the RS and RH of the three age stands were the same. The model curve of the quadratic function seems to be the highest at the same time, maintaining higher RS and RH levels for the 48a stand, followed by the 36a stand. This indicated that the older stand was more sensitive to stand density than what? In terms of autotrophic respiration, except for stand with 48 years, the autotrophic respiration of 36a and 27a stands showed a clear upward trend with an increase of stand density.

We tried to explore more about the mechanism of stand density effects on RS after the effect of stand density on different ages of *Larix principis-rupprechtii* plantation was clarified. Therefore, we used R 4.0.3 Lavan package to establish the structural equation model (SEM) for the four observed variables of three different aged-stands. It was found that Lavaan normally ended after 60 iterations, and the model's p-value (p = 0.653, χ^2 = 1.628,CFI = 1.000) was greater than 0.05, so the structural equation model was acceptable (Table 3). We found that the influence of forest density of *Larix principis-rupprechtii* plantation on soil respiration at different ages mainly came from the regulation of heterotrophic respiration, and a small part came from the regulation of autotrophic respiration. The direct effect of heterotrophic respiration on soil respiration was 0.84, autotrophic respiration was 0.64.

Table 3. Structural equation model parameter estimates. SD, stand density; SD2, stand density square.

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3.3 Effects of stand density on SOC and labile organic carbon

We separately analyzed the differences of SOC, MBC, DOC, LFOC, ROC between different density levels. We used the advantages of the density sequence plots to find the trend of these indicators with stand density.

3.3.1 SOC, MBC, DOC, LFOC, ROC of different stand ages and stand density levels

No significant difference was found in the SOC of the three age stands among different stand density levels (Table 4). For the 27a stand, MBC of MD was slightly higher than that of LD and significantly higher than HD; DOC had no significant difference; LFOC of MD and LD was significantly higher than HD; ROC had no significant difference, but ROC of MD was lower than LD and HD. In the 36a stand, the MBC of MD was significantly higher than that of LD and HD; there was no significant difference in DOC; the LFOC of LD and MD was significantly higher than that of HD; there was no significant difference in ROC, but the ROC of MD was lower than that of LD and HD. For the 48a stand, the MBC of MD was significantly higher than that of HD and slightly higher than that of LD; the DOC of LD was slightly higher than that of MD and significantly higher than HD.

| Stand age | Stand density level | SOC (g/kg) | MBC (mg/kg) | DOC (mg/kg) | LFOC (mg/kg) | ROC (mg/kg) |
|-----------|---------------------|------------|-------------|-------------|--------------|-------------|
| 27a       | LD                  | 14.19 ± 1.56a | 286.37 ± 18.89ab | 196.42 ± 15.17a | 381.53 ± 9.89a | 1328.85 ± 100.73a |
|           | MD                  | 14.21 ± 0.41a | 308.94 ± 4.17a  | 206.48 ± 5.71a  | 379.92 ± 14.00a | 1298.91 ± 240.39a |
|           | HD                  | 12.03 ± 1.77a | 263.93 ± 21.01b | 191.90 ± 2.69a  | 351.99 ± 6.51b  | 1503.01 ± 73.66a |
| 36a       | LD                  | 19.83 ± 1.62a | 402.62 ± 29.44a | 257.88 ± 9.94a  | 419.48 ± 5.43a  | 1651 ± 72.05a   |
|           | MD                  | 19.33 ± 0.79a | 455.49 ± 3.51ab | 258.21 ± 13.72a | 426.35 ± 10.62a | 1321.13 ± 281.17a |
|           | HD                  | 17.88 ± 0.14a | 384.30 ± 33.61a | 260.66 ± 6.52a  | 403.21 ± 4.67b  | 1636.35 ± 82.91a |
| 48a       | LD                  | 24.96 ± 1.58a | 441.18 ± 71.05ab| 364.68 ± 4.20a  | 501.76 ± 13.60a | 2302.76 ± 83.38a |
|           | MD                  | 22.67 ± 2.32a | 533.82 ± 81.96a | 355.39 ± 14.31ab| 523.81 ± 16.39a | 1548.48 ± 286.68b |
|           | HD                  | 22.16 ± 0.55a | 381.57 ± 14.75b | 345.63 ± 2.64b  | 419.88 ± 5.36b  | 2200.04 ± 343.99a |
3.3.2 The variation of SOC and labile organic carbon with stand density at different stand ages

The SOC of different ages showed a downward trend with stand density changes and could be adequately described by linear regression. Although the SOC of each age stand with the increase of stand density, SOC remained high for 48 years, followed by 36 years and the lowest at 27 years (Fig. 5A). The DOC did not show an obvious trend with stand density (Fig. 5B). The LFOC of three age stands first increased and then decreased with the increase of stand density, and the trend of the 48a stand was the most obvious, and the changes for s36a and 27a stands were not significant (Fig. 5C). The trend of soil ROC changes was opposite to that of LFOC, and the trend of soil ROC of three aged-stands showed the first decreasing trend and then increasing trend (Fig. 5D). Soil MBC first increased and then decreased with an increase of the stand density. The MBC content of 48a stands reduced to 400 mg/kg when the density was higher than 1300 trees/ha, which was lower than 36a stand with similar density (Fig. 5E).

Based on these trends, we could conclude that although the general trend of SOC and soil labile organic carbon affected by density was the same in different age stands, there were some differences in their sensitivity to the density changes. For the 48a stand, density change response is the strongest, as this was especially reflected in the LFOC, ROC, and MBC. For the 27a stand, response was weaker than that for 36a and 48a, and the range of variation with stand density was rather small.

3.4 The correlations between soil organic carbon and labile organic carbon and soil respiration

We further explored whether there was a certain correlation between soil respiration and soil labile organic carbon under the background of stand density regulation (Fig. 6). The correlation analysis showed that RS was significantly negatively correlated with ROC, significantly positively correlated with the DOC, LFOC, and MBC, RH was significantly negatively correlated with ROC, SOC, and MBC. For the 36a stand, RS and Rh showed a negative correlation with ROC, strong positive correlation with MBC, strong positive correlation with LFOC, and significant positive correlation between RA and DOC. For the 48a stand, RS was significantly positively correlated with MBC, LFOC, DOC, but significantly negatively correlated with ROC, RA and SOC, MBC, MBC, and RH Doc and LFOC were significantly positively correlated, but negatively correlated with ROC. RA was significantly positively correlated with MBC and LFOC and significantly negatively correlated with ROC.

3.5 Explanation of soil organic carbon pool to the soil respiration variability

After understanding the correlations between SOC, soil labile organic carbon and soil respiration, we carried out the redundant analyses of soil respiration and soil organic carbon using SOC and soil labile organic carbon to explain the variation of soil respiration rate. The axes 1 and 2 described 99.99% variations of the variable of interest, indicating well representation of the original explanatory variable. The RDA1 and RDA2 contained almost all the information of the axes and jointly explained 56.05% of the variance of RS, Rh, and RA (Fig. 7). See Table 5 for detailed parameter estimates.

Table 5.
Redundancy analysis results.

| Parameters                                      | RDA1    | RDA2    | RDA3    |
|------------------------------------------------|---------|---------|---------|
| Eigenvalues                                     | 0.0133  | 0.0025  | 0.0170  |
| Soil respiration cumulative percentage variance | 47.2490 | 56.0529 | 56.0530 |
| Soil organic carbon factor - Soil respiration cumulative percentage variance | 84.2910 | 99.9971 | 1.0000  |
| Sum of all canonical eigenvalues                 | 56.053  |         |         |
| Monte Carlo permutation test                    |         |         |         |
| Significance of first canonical axis            | P < 0.001 |         |         |
| Significance of all canonical axis              | P < 0.001 |         |         |
4. Discussion

4.1 The effect of stand density on soil respiration of different aged-stands

The RS and RH in the different density levels show significant differences (Fig. 2). The RS and RH of MD of the three aged-stands were higher than LD and HD, and the RA of the 48a stand was significantly higher than LD and HD, but there was no significant difference in RA of the other two age stands. We consider that for the three stand ages, moderate stand density promotes RS and RH, combine the trends of RS, RH, and RA to give a reasonable explanation.

The RS and RH first increase and then decrease with an increase of the stand density. This trend can be well fitted by the quadratic function (Figure 7), which was also reported in the previous studies, and coincided with the idea of Intermediate-Disturbance Hypothesis (Connell, 1979). However, it seems that most of the research conclusions are limited to that thinning can significantly improve forest soil respiration, and different thinning intensities (retention density) have different enhancement, that is, appropriate reduction of the stand density would increase RS and RH (Shao et al., 2017; Lei et al., 2018; Zhang et al., 2018). This is similar to what we found in our study. The only difference is that RS and RH first increase and then decrease with the stand density, and they could not keep a high level when the density was too low. We assume that there may be two reasons, such as under the effect of long-term low density, the soil microenvironment created by low stand density and upper shading could not be suitable for soil microbial activities (Yang et al., 2017). A litter with low stand density may not be suitable for soil microbial activities, and the amount returned was less than that of high density, which resulted in the limitation of organic carbon components as respiratory substrates in some soil carbon pools. RA’s trend for 48a stand with stand density is similar to RS and RH, and RA shows an upward trend with stand density for 36a and 27a stands. (Fig. 3C). we consider that there is a threshold for the capacity of the soil to hold plant roots. An increase in the root density within the threshold will cause an increase in the soil autotrophic respiration. This is the reason for the increase in soil autotrophic respiration for 27a and 36a stands, while in 48a plantations, too high forest stand density causes root density to exceed this threshold. Plant roots and root microbes compete fiercely (Grubb, 2000), even leading to reduced root microbial activity, root death, and even plant death (Kuzyakov and Larionova, 2005), so for 48a stand, soil RA will first increases and then decrease with an increasing stand density.

The trends appeared in RS, RH, and RA were significantly age-related (Fig. 3), and was fastest in the 48-year-old stand, followed by the 36-year-old stand, and slowest in the 27-year-old stand. (Gao et al., 2019; Yu et al., 2019; Wu et al., 2020). Some researchers also obtained different results. They found that soil respiration decreased with the growth of stand age, and the decline of soil fine root biomass may be the reason for the decline of soil respiration (Saiz et al., 2006). We found that in the forest with positive succession, with the increase of forest age, due to the accumulation of more organic matter and the improvement of Soil Microenvironment, soil microorganisms can better play their role as decomposers in the soil, and promote the reox process in the soil (Shrestha et al., 2014; You et al., 2014), Therefore, the difference of soil respiration and heterotrophic respiration rate in stand age and the increase with age were caused.

The regulation mechanism of stand density on RS is that the stand density mainly regulates soil respiration by changing RH and then by regulating RA, which is empirically proved by the structural equation model (Figure 7). The change of RA and its effect on soil respiration are weaker than heterotrophic respiration (Bond-Lamberty et al., 2004). The reason may be that soil organic carbon status and Soil Microenvironment of different stand densities are different. In other words, from the perspective of soil respiration mechanism, it is the substrate state, process enzyme activity, temperature, and humidity may have differences, which are also the major factors affecting RH.

4.2 The effect of stand density on SOC and labile organic carbon of different age stands

Many studies have confirmed that soil depth substantially affects organic carbon and soil nutrients, so it does not appear as the discussion content of this study (Baldrian et al., 2012). We can see that in the three age stands, the differences in soil labile organic carbon at different stand density levels are mainly reflected in three indicators: MBC, LFOC, and ROC. Among them, the MBC and LFOC of MD are significantly higher than LD and HD. ROC is lower than LD and HD. In other words, moderate stand density promotes the accumulation of MBC and LFOC and inhibits the accumulation of ROC. We know that SOC could increase with the decrease of stand density to a certain extent. Although the reduction of stand density increases the loss of litter (Lim et al., 2012; Lull et al., 2020), the appropriate soil environment increases the biochemical process rate, thus promoting the increase of SOC, which is consistent with
that the enzymes related to the soil C cycle play the role of “controlling the speed,” and then speculate whether the existing RS model respiration LFOC, MBC and ROC in carbon components are responsible for “warehouse” and “tools” and “raw materials.” We speculate that is, stand density soil labile organic carbon soil respiration, which is also an important path of stand density regulating soil hand, it regulates the state and content of labile organic carbon in soil carbon pool and affects soil respiration from another aspect, On the one hand, stand density affects the soil microenvironment, such as temperature and humidity (Table or Figure?). On the other that SOC and soil labile organic carbon have a high degree of explanation (56.05%) for the variance variation of RS, RH, and RA. to link the relationship between labile organic carbon and soil respiration. The results of RDA also proved this point well. We found “tools should support the high rate of soil respiration,” LFOC, MBC and RS, RH show a significant positive correlation. This allows us with RS and RH, which also explains the trend that ROC first decreases and then increases with stand density. Because more powerful “tools should support the high rate of soil respiration,” LFOC, MBC and RS, RH show a significant positive correlation. This allows us to link the relationship between labile organic carbon and soil respiration. The results of RDA also proved this point well. We found that SOC and soil labile organic carbon have a high degree of explanation (56.05%) for the variance variation of RS, RH, and RA. On the one hand, stand density affects the soil microenvironment, such as temperature and humidity (Table or Figure?). On the other hand, it regulates the state and content of labile organic carbon in soil carbon pool and affects soil respiration from another aspect, that is, stand density soil labile organic carbon soil respiration, which is also an important path of stand density regulating soil respiration LFOC, MBC and ROC in carbon components are responsible for “warehouse” and “tools” and “raw materials.” We speculate that the enzymes related to the soil C cycle play the role of “controlling the speed,” and then speculate whether the existing RS model
can be improved by using the characteristics of labile organic carbon (respiration substrate characteristics) and enzyme activity (rate characterization), so as to make the model more ecological and scientific, which will be the focus of our subsequent study.

4.5 Proposal of the optimal density for each age of Larix principis-rupprechtii plantation

Although this is a statement with substantial limitations, it is an essential reference for regional forest management.

Taking afforestation density of 3300 plants/ha as an example, the forest stand for 27 years should maintain about 1650 plants/ha (50%), the forest stand for 36 years should be about 1250 plants/ha (38%), and the forest stand for 48 years should be maintained at 900 plants/ha (27%), so that both the above-ground vegetation carbon pool and the soil carbon storage carbon sequestration can be maintained at a high level, while the soil labile organic carbon content is at a high level and stable, the soil microbial activity is high, and the soil quality is good. This is what a forest manager wants to see.

5. Conclusion

Among the forest stands of three ages, RS and RH of different stand density levels was significantly different, MD was higher than LD and HD. Moderate density promotes RS rate and RH rate; RS and RH of three ages stand to increase with stand density and then decrease. The quadratic function can better fit this trend. Except for 48 years, RA increases gently with the increase of stand density; stand density affects both RH and RA, but it mainly affects RS by regulating RH.

Among the forest stands of three ages, the MBC, LFOC, and ROC (0-30cm) of different density levels are significantly different. The MBC and LFOC of MD are higher than LD and HD, and the ROC of MD is lower than LD and HD; moderate Density promotes the sequestration of MBC and LFOC, and inhibits the sequestration of ROC. With the increase of forest density, LFOC and MBC first increased and then decreased, and ROC first decreased and then increased, the quadratic function can fit these changing trends.

The RS, RH, and RA rates of older forest stands are relatively fast, and the contents of SOC, MBC, LFOC, DOC, and ROC are higher, and they are more sensitive to changes in stand density.

The SOC, LFOC, MBC, DOC, and ROC explained 56.05% variance of RS, Rh, and RA. MBC, LFOC, and ROC in soil labile organic carbon were closely related to RS and Rh, but not SOC. Among them, LFOC and MBC play the role of "warehouse" and "tool" and significantly correlate with RS and Rh. ROC, as "raw material," has a significant negative correlation with RS and Rh. When RH and RS’s rate is fast, the dynamic stability of a low level is maintained. Stand density can regulate RH by affecting soil labile organic carbon, which is an essential path for stand density to regulate soil respiration.

Abbreviations

BD: Bulk density.
SWC: Soil moisture content
TN: Soil total nitrogen content
TP: Soil total phosphorus content
LD: Low density
MD: Medium density
HD: High density
RS: Soil respiration
RH:Heterotrophic respiration
RA: Autotrophic respiration
SOC: Soil organic carbon
MBC: Microbial biomass carbon
LFOC: Light fraction organic carbon
DOC: Dissolved organic carbon
ROC: Easily oxidizeable organic carbon
PVC: Polyvinyl chloride

Declarations

CRediT authorship contribution statement
Tairui Liu: Conceptualization, Formal analysis, Investigation, Data curation, Writing - original draft, Writing - review & editing. Daoli Peng: Conceptualization, Resources, Supervision, Project administration, Funding acquisition. Zhijie Tan: Investigation, Data curation, Validation, Visualization. Jingping Guo: Investigation, Data curation, Resources. Yunxiang Zhang: Resources.

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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References
1. Akburak, S., Makineci, E., 2016. Thinning effects on soil and microbial respiration in a coppice-originated Carpinus betulus L. stand in Turkey. iForest - Biogeosciences and Forestry 9, 783-790.
2. Ali, A., Dai, D., Akhtar, K., Teng, M., Yan, Z., Urbina-Cardona, N., Mullerova, J., Zhou, Z., 2019. Response of understory vegetation, tree regeneration, and soil quality to manipulated stand density in a Pinus massoniana plantation. Global Ecology and Conservation 20.
3. T., Christensen, 2001. Physical fractionation of soil and structural and functional complexity in organic matter turnover. European Journal of Soil Science.
4. Baggs, E.M., 2006. Partitioning the components of soil respiration: a research challenge. Plant and Soil 284, 1-5.
5. Bai, S.H., Dempsey, R., Reverchon, F., Blumfield, T.J., Ryan, S., Cernusak, L.A., 2016. Effects of forest thinning on soil-plant carbon and nitrogen dynamics. Plant and Soil 411, 437-449.

6. Baldrian, P., Kolarik, M., Stursova, M., Kopecky, J., Valaskova, V., Vetrovsky, T., Zifcakova, L., Snajdr, J., Ridl, J., Vlcek, C., Voriskova, J., 2012. Active and total microbial communities in forest soil are largely different and highly stratified during decomposition. ISME J 6, 248-258.

7. Bello, J., Vallet, P., Perot, T., Balandier, P., Seigner, V., Perret, S., Couteau, C., Korboulewsky, N., 2019. How do mixing tree species and stand density affect seasonal radial growth during drought events? Forest Ecology and Management 432, 436-445.

8. Bolan, N.S., Baskaran, S., Thiagarajan, S., 2008. An evaluation of the methods of measurement of dissolved organic carbon in soils, manures, sludges, and stream water. Communications in Soil Science and Plant Analysis 27, 2723-2737.

9. Bolat, İ., 2013. The effect of thinning on microbial biomass C, N and basal respiration in black pine forest soils in Mudurnu, Turkey. European Journal of Forest Research 133, 131-139.

10. Bond-Lamberty, B., Wang, C., Gower, S.T., 2004. A global relationship between the heterotrophic and autotrophic components of soil respiration? Global Change Biology 10, 1756-1766.

11. Brookes, P.C., Landman, A., Pruden, G., Jenkinson, D.S., 1985. Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil - ScienceDirect. Soil Biology and Biochemistry 17, 837-842.

12. Cambardella, C.A., Elliott, E.T., 1992. Particulate Soil Organic-Matter Changes across a Grassland Cultivation Sequence. Soil Science Society of America Journal 56.

13. Cheng, X., Kang, F., Han, H., Liu, H., Zhang, Y., 2015. Effect of thinning on partitioned soil respiration in a young Pinus tabulaeformis plantation during growing season. Agricultural and Forest Meteorology 214-215, 473-482.

14. Christensen, B.T., 1992. Physical Fractionation of Soil and Organic Matter in Primary Particle Size and Density Separates. Adv Soil 20.

15. Connell, J.H., 1979. Intermediate-Disturbance Hypothesis. Science 204, 1344-1345.

16. Dib, A.E., Johnson, C.E., Driscoll, C.T., Fahey, T.J., Hayhoe, K., 2014. Simulating effects of changing climate and CO(2) emissions on soil carbon pools at the Hubbard Brook experimental forest. Glob Chang Biol 20, 1643-1656.

17. Eldegard, K., Scholten, J., Stokland, J.N., Granhus, A., Lie, M., 2019. The influence of stand density on bilberry (Vaccinium myrtillus L.) cover depends on stand age, solar irradiation, and tree species composition. Forest Ecology and Management 432, 582-590.

18. Fernandez, I., Álvarez-González, J.G., Carrasco, B., Ruiz-González, A.D., Cabaneiro, A., 2012. Post-thinning soil organic matter evolution and soil CO2 effluxes in temperate radiata pine plantations: impacts of moderate thinning regimes on the forest C cycle. Canadian Journal of Forest Research 42, 1953-1964.

19. Franklin, O., Aoki, K., Seidl, R., 2009. A generic model of thinning and stand density effects on forest growth, mortality and net increment. Annals of Forest Science 66.

20. Gabriel, C.E., Kellman, L., Prest, D., 2018. Examining mineral-associated soil organic matter pools through depth in harvested forest soil profiles. PLoS One 13, e0206847.

21. Gao, J., Zhang, Y., Song, Q., Lin, Y., Zhou, R., Dong, Y., Zhou, L., Li, J., Jin, Y., Zhou, W., Liu, Y., Sha, L., Grace, J., Liang, N., 2019. Stand age-related effects on soil respiration in rubber plantations (Hevea brasiliensis) in southwest China. European Journal of Soil Science 70, 1221-1233.

22. Goldberg, S.D., Zhao, Y., Harrison, R.D., Monkai, J., Li, Y., Chau, K., Xu, J., 2017. Soil respiration in sloping rubber plantations and tropical natural forests in Xishuangbanna, China. Agriculture, Ecosystems & Environment 249, 237-246.

23. Grubb, C.P.J., 2000. Impacts of Root Competition in Forests and Woodlands: A Theoretical Framework and Review of Experiments. Ecological Monographs 70, 171-207.

24. Hopkins, F., Gonzalez-Meler, M.A., Flower, C.E., Lynch, D.J., Czimczik, C., Tang, J., Subke, J.A., 2013. Ecosystem-level controls on root-rhizosphere respiration. New Phytol 199, 339-351.

25. Jack, S.B., Long, J.N., 1991. Analysis of stand density effects on canopy structure: a conceptual approach. Trees 5, 44-49.

26. Jens, Emborg, and, Morten, Christensen, and, Jacob, Heilmann-Clausen, 2000. The structural dynamics of Suserup Skov, a near-natural temperate deciduous forest in Denmark. Forest Ecology and Management 126, 173-189.
28. Jílková, V., 2020. Soil respiration in temperate forests is increased by a shift from coniferous to deciduous trees but not by an increase in temperature. Applied Soil Ecology 154.

29. Khan, M.N.I., Shil, M.C., Azad, M.S., Sadath, M.N., Feroz, S.M., Mollick, A.S., 2018. Allometric relationships of stem volume and stand level carbon stocks at varying stand density in Swietenia macrophylla King plantations, Bangladesh. Forest Ecology and Management 430, 639-648.

30. Kuzyakov, Y., 2006. Response to the comments by Peter Högberg, Nina Buchmann and David J. Read on the review ‘Sources of CO2 efflux from soil and review of partitioning methods’ (Soil Biology & Biochemistry 38, 425–448) Object- versus method-oriented terminology. Soil Biology and Biochemistry 38, 2999-3000.

31. Kuzyakov, Y., Larionova, A.A., 2005. Root and rhizomicrobial respiration: A review of approaches to estimate respiration by autotrophic and heterotrophic organisms in soil. Journal of Plant Nutrition and Soil Science 168, 503-520.

32. Lal, R., 2005. Forest soils and carbon sequestration. Forest Ecology and Management 220, 242-258.

33. Lei, L., Xiao, W., Zeng, L., Zhu, J., Huang, Z., Cheng, R., Gao, S., Li, M.H., 2018. Thinning but not understory removal increased heterotrophic respiration and total soil respiration in Pinus massoniana stands. Sci Total Environ 621, 1360-1369.

34. Liang, B.C., Mackenzie, A.F., Schnitzer, M., Monreal, C.M., Voroney, P.R., Béyaeart, R.P., 1997. Management-induced change in labile soil organic matter under continuous corn in eastern Canadian soils. Biology and Fertility of Soils 26, 88-94.

35. Lim, H., Choi, W.-J., Ahn, K., Lee, K.-H., 2012. Ecosystem respiration and tree growth influenced by thinning in a red pine forest in southern Korea. Forest Science and Technology 8, 192-204.

36. Liu, T., Dong, W., Tan, Z., Zhang, Y., Guo, J., University, S.A., University, S.A., 2019. Analysis of characteristics of north China larch plantations’ competitive relationship and its driving factors. Journal of Forest & Environment.

37. Mosca, E., Montecchio, L., Barion, G., Dal Cortivo, C., Vamerali, T., 2017. Combined effects of thinning and decline on fine root dynamics in a Quercus robur L. forest adjoining the Italian Pre-Alps. Ann Bot 119, 1235-1246.

38. Olajuyigbe, S., Tobin, B., Saunders, M., Nieuwenhuis, M., Nieuwenhuis, M., 2012. Forest thinning and soil respiration in a Sitka spruce forest in Ireland. Agricultural and Forest Meteorology 157, 86-95.

39. Poorter, L., Bongers, F., Aide, T.M., Zambrano, A.M.A., 2016. Biomass resilience of Neotropical secondary forests. Nature.

40. Qiu, X., Peng, D., Wang, H., Wang, Z., Cheng, S., 2019. Minimum data set for evaluation of stand density effects on soil quality in Larix principis-rupprechtii plantations in North China. Ecological Indicators 103, 236-247.

41. Ryu, S.-R., Concilio, A., Chen, J., North, M., Ma, S., 2009. Prescribed burning and mechanical thinning effects on belowground conditions and soil respiration in a mixed-conifer forest, California. Forest Ecology and Management 257, 1324-1332.

42. Shao, G., Shugart, H.H., 1997. Notes: A Compatible Growth-Density Stand Model Derived from a Distance-Dependent Individual Tree Model. Forest Science 43, 443-446.

43. Shao, G., Tian, S.Y., Liu, Y.K., Li, Y.H., Sun, Z.H., 2017. Effects of density control on soil respiration in Larix olgensis plantation. Journal of Beijing Forestry University 39, 51-59.

44. Shrestha, R.K., Strahm, B.D., Sucre, E.B., Holub, S.M., Meehan, N., 2014. Fertilizer Management, Parent Material, and Stand Age Influence Forest Soil Greenhouse Gas Fluxes. Soil Science Society of America Journal 78, 2041.

45. Tian, Q., He, H., Cheng, W., Bai, Z., Wang, Y., Zhang, X., 2016. Factors controlling soil organic carbon stability along a temperate forest altitudinal gradient. Sci Rep 6, 18783.
51. Wander, M.M., Traina, S.J., Stinner, B.R., Peters, S.E., 1994. Organic and Conventional Management Effects on Biologically Active Soil Organic Matter Pools. Soil Sci. Soc. Am. J. 58, 1130-1139.

52. Wei, W., Weile, C., Shaopeng, W., 2010. Forest soil respiration and its heterotrophic and autotrophic components: Global patterns and responses to temperature and precipitation. Soil Biology and Biochemistry 42, 1236-1244.

53. Vic Baena, C., Andrés-Abellán, M., Lucas-Borja, M.E., Martínez-García, E., García-Morote, F.A., Rubio, E., López-Serrano, F.R., 2013. Thinning and recovery effects on soil properties in two sites of a Mediterranean forest, in Cuenca Mountain (South-eastern of Spain). Forest Ecology and Management 308, 223-230.

54. Wu, T., Schoenau, J.J., Li, F., Qian, P., Malhi, S.S., Shi, Y., 2003. Effect of tillage and rotation on organic carbon forms of chernozemic soils in Saskatchewan. Journal of Plant Nutrition and Soil Science 166.

55. Wu, X., Xu, H., Tuo, D., Wang, C., Fu, B., Lv, Y., Liu, G., 2020. Land use change and stand age regulate soil respiration by influencing soil substrate supply and microbial community. Geoderma 359.

56. Xu, M., Shang, H., 2016. Contribution of soil respiration to the global carbon equation. J Plant Physiol 203, 16-28.

57. Yang, Y., Geng, Y., Zhou, H., Zhao, G., Wang, L., 2017. Effects of gaps in the forest canopy on soil microbial communities and enzyme activity in a Chinese pine forest. Pedobiologia 61, 51-60.

58. You, Y., Wang, J., Huang, X., Tang, Z., Liu, S., Sun, O.J., 2014. Relating microbial community structure to functioning in forest soil organic carbon transformation and turnover. Ecol Evol 4, 633-647.

59. Yu, K., Yao, X., Deng, Y., Lai, Z., Lin, L., Liu, J., 2019. Effects of stand age on soil respiration in Pinus massoniana plantations in the hilly red soil region of Southern China. Catena 178, 313-321.

60. Zhang, X., Guan, D., Li, W., Sun, D., Jin, C., Yuan, F., Wang, A., Wu, J., 2018. The effects of forest thinning on soil carbon stocks and dynamics: A meta-analysis. Forest Ecology and Management 429, 36-43.

61. Zhao, B., Cao, J., Geng, Y., Zhao, X., von Gadow, K., 2019. Inconsistent responses of soil respiration and its components to thinning intensity in a Pinus tabuliformis plantation in northern China. Agricultural and Forest Meteorology 265, 370-380.

62. Hu, H., Ma, H., Luo, C., Hu, T., 2010. Classification and determination of forest soil organic carbon. Soil bulletin, 1018-1024.

63. Liu, T., Dong, W., Tan, Z., Zhang, Y., Guo, J., 2019. Effects of different thinning intensities on competition of Larix principis ruprechtii plantation. Acta forest and environment 39, 44-49.

**Figures**
Figure 1

Study area. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand. Note: The designations employed and the presentation of the material on this map do not imply the expression of any opinion whatsoever on the part of Research Square concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. This map has been provided by the authors.
Figure 2

RS, Rh, and RA for the stands with different ages and stand density levels. RS, soil respiration; RH, heterotrophic respiration; RA, autotrophic respiration. Values in the black box are as the mean ± standard deviation (SD). Different lowercase letters (a, b, and c) indicate significant differences (p < 0.05) among different stand density levels. RS, soil respiration; RH, and heterotrophic respiration; RA, autotrophic respiration. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.
Figure 3

Variation of RS, Rh, and RA of the stands with different stand density and stand ages. Error bars show the standard deviation (SD). The gray part represents the confidence interval. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.
Figure 4

Structural equation model of the influence mechanism of stand density on RH, RA, and RS. Stand density 2 is the square of stand density; all lines are direct effects, the red line is a negative effect, and the green line is a positive effect.
Figure 5

Variation of SOC and soil labile organic carbon with stand density in different aged-stands. Error bars show the standard deviation (SD). The gray part represents the confidence interval. SOC, soil organic carbon; DOC, soil dissolved organic carbon; MBC, microbial biomass carbon; LFOC, light fraction organic carbon; ROC, easily oxidizable organic carbon. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.
Figure 6

Correlations between RS, RH, RA and SOC, soil labile organic carbon at different age-stands. SOC: soil organic carbon; DOC, soil dissolved organic carbon; MBC, microbial biomass carbon; LFOC, light fraction organic carbon; ROC, easily oxidizable organic carbon; RS, soil respiration; RH, and heterotrophic respiration; RA, autotrophic respiration. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.
Figure 7

Redundancy analysis (RDA) to determine the effects of the selected Soil organic carbon factor (blue arrow) on soil respiration (red arrow). The SOC: soil organic carbon; DOC, soil dissolved organic carbon; MBC, microbial biomass carbon; LFOC, light fraction organic carbon; ROC, easily oxidizeable organic carbon; RS, soil respiration; RH, and heterotrophic respiration; RA, autotrophic respiration. 27a, 27-year-old stand; 36a, 36-year-old stand; 48a, 48-year-old stand.