

The effects of crop tree thinning intensity on the ability of dominant tree species to sequester carbon in a temperate deciduous mixed forest, northeastern China

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Abstract: Forest management is one of the important nature-based solutions for climate mitigation. Thinning can indirectly influence tree physiology by changing the microclimate and directly change the stand biomass, which can impact forest carbon sequestration. However, previous results about how thinning might influence carbon stocks remain inconsistent regarding post-thinning carbon accretion. In this study, crop tree release (CTR) thinning in four intensities (CK: 0% of basal area removal, LT: 17.25%, MT: 34.73%, and HT: 51.87%) were conducted in a temperate deciduous forest in Jiaohe, northeastern China in 2011. Plot inventories in 2011, 2013, 2015, 2018 and 2021 and tree cores collected in 2017 and 2018 offered the opportunity to examine how are the interannual carbon sequestration ability of Korean pine and Manchurian ash responded to CTR thinning in four intensities. We quantify the carbon sequestration ability of trees by calculating individual stem carbon stock and annual carbon stock rate to examine whether the previous inconsistency was attributed to different responses of species, and the ignorance of frozen carbon content. The results show: (1) after thinning, the underestimation of carbon stocks of Manchurian ash decreased with the increasing thinning intensity. The greatest underestimation of Manchurian ash reaches 2922kg ha⁻¹, while that of Korean pine only reaches 283kg ha⁻¹. Compared with Manchurian ash, the conventional carbon fraction of 0.5 for Korean pine is more appropriate, and the misestimation of Korean pine didn't show an obvious pattern with the intensity of thinning. (2) Under light thinning, both species maintained a stable carbon stock growth, and the frozen carbon content of Korean pine was significantly increased. During the 10 years after light thinning, the individual stem carbon of Korean pine increased from 57 kg to 81 kg, and Manchurian ash increased from 201 kg to 268 kg. The average rate of increase of individual stem carbon is positively related to tree size. Removing such large-diameter trees from the stand is likely to decrease carbon stock rate. Therefore, it is essential to design carbon-friendly silviculture prescriptions worldwide under the consideration of species, sizes, and intensities.

Key words: crop tree release thinning; thinning intensity; frozen carbon content; carbon stocks; carbon sequestration ability; carbon capture

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1 Introduction

Global warming is receiving unprecedented concern, and the huge potential of forests in climate change mitigation has been widely discussed (Canadell and Raupach, 2008; Pan et al., 2011; Sitch et al., 2015): forests constitute an important global carbon sink with an estimated 296 Gt of carbon in both above- and below- ground biomass (FAO, 2018). Improved forest management is perceived as the third largest natural pathway for climate mitigation and plays a pivotal role in limiting global warming to below 2 °C (Griscom et al., 2017). Compared with forest regeneration and reforestation, forest management is more cost-effective and can be implemented rapidly since it doesn't involve land-use change or tenure change (Griscom et al., 2017).

Thinning is one of the most important silvicultural operations in a managed forest. Through intentionally removing trees to regulate competition, thinning reallocates the growing space, improve growing conditions (ex: light, temperature, water, nutrients) for residual trees, support vigorous tree growth, minimize tree mortality and thus increase forest growth, timber productivity and economic value (Eriksson, 2006; Geng et al., 2021; Saarinen et al., 2020). Many studies have examined the effects of thinning on carbon stocks (del Río et al., 2017; Lin et al., 2018; Schaedel et al., 2017; Shuyong et al., 2017), but previous results remain inconsistent. The rapid regeneration of understory vegetation and the fast growth of post-thinning survivors could lead to greater carbon sequestration rates (Briceño et al., 2006; Hoover and Stout, 2007; López et al., 2003; Schilling et al., 1999; Zheng et al., 2019). Longer rotation may also enhance carbon sequestration, because it allows the biomass production to recover to avoid making the forest a net carbon source (Kaipainen et al., 2004; Nepstad et al., 1999). However, precommercial thinning and thinning in young forests were found to have no influence on aboveground carbon stocks (Lin et al., 2018; Ruiz-Benito et al., 2014). The carbon storage may even be reduced when forests are managed for maximum biomass yield (Cooper, 1983). The inconsistencies may be attributed to different thinning types, intensities, and timing involved in studies (Eriksson, 2006)). Different thinning plans have been designed to achieve various management goals (Ashton and Kelty, 2018; Saarinen et al., 2020). As a special type of thinning, crop tree release (CTR) thinning intends to reduce competition around selected trees so that they can improve in vigor, remain competitive in the stand, and provide desired future benefits (Miller et al., 2007). In a natural mixed forest, not only the stand density and tree size distribution are influenced, but also the composition of tree species (Ameha et al., 2016; Zhang et al., 2014). If the selected trees are light and drought-tolerant species, the reduced stand density and changed canopy complexity can benefit the carbon sequestration because of higher transmitted solar radiation (Hardiman et al., 2011; F. Wang et al., 2020). Since various species and trees in different sizes may have inconsistent responses to thinning, the effect of CTR thinning on forest carbon stocks was case-dependent. However, few studies paid attention to how might CTR thinning impact forest carbon stocks (Li et al., 2021).

When estimating carbon stocks on a large scale, more attention has been paid to biomass estimation. However, the carbon stock is determined by multiplying the biomass with a carbon fraction value (Guerra-Santos et al., 2014; Nizami, 2012; Pan et al., 2011; Tang et al., 2018), which has long been assumed to be 50% (Fang et al., 2007; Zhang et al., 2015; Zhu et al., 2015, 2017). Under such simplicity, many large-scale studies have estimated the national and global vegetation carbon stock (Fang et al., 2007; Pan et al., 2011), but the result varies. The estimates of the carbon sink of Chinese terrestrial ecosystem varies from 0.19 - 0.26 PgC year⁻¹ (Fang et al., 2018; Piao et al., 2009) to 1.11 PgC year⁻¹ (Wang et al., 2020). The under representation of survey data and the difference of methodology used could partially explaining this variation (Li et al., 2021). Besides, frozen carbon content observed in early 2000s (Lamlom and Savidge, 2003; Thomas and Malczewski, 2007) was also considered to be a non-negligible part of carbon content (Alvarez et al., 2014; Martin and Thomas, 2011; Thomas and Malczewski, 2007). Traditional carbon content is measured after the sample is oven dried. The high

temperature could result in loss of volatile carbon, which is constituted by certain low molecular weight compounds, such as alcohols, phenols, terpenoids and aldehydes (Lamloom and Savidge, 2003). Frozen carbon content, however, is the carbon content measured after frozen-drying method. It is believed that freezing can maintain the volatile carbon content in wood sample, thereby avoiding misestimation. Frozen carbon content may vary substantially among tree species and increase the uncertainties in calculating carbon stock (Bert and Danjon, 2006; Lamloom and Savidge, 2003). Nevertheless, the variation of carbon content after thinning has rarely been examined and few studies take frozen carbon into account.

The natural coniferous and broad-leaved mixed forest in northeastern China account for about one-third of the total national carbon stock and are crucial for China's climatic system (State Forestry and Grassland Administration, 2019; Wang, 2006): the net CO₂ sink during the 2007 growing season was 247 g C m⁻² (Wang et al., 2010). It is characterized by distinctive species composition and high biodiversity (Qian et al., 2019). Korean pine (*Pinus koraiensis* Siebold & Zucc) and Manchurian ash (*Fraxinus mandshurica* Rupr.) are dominant tree species in this forest. They are also important components of the carbon stocks in northeastern China, because under continuing climate warming, the dominance of Korean pine was predicted to decrease and that of Manchurian ash might increase (Dai et al., 2013). Moreover, they are important timber and economic species, having optimal wood quality and huge economic value. Additionally, Korean pine is a conifer, and Manchurian ash is an angiosperm (Zhang, 2008), so they may have different responses to thinning disturbance. Meanwhile, research into 14 native tree species in northeast China found their volatile carbon value, which was calculated as the difference between frozen carbon content and oven-dried carbon content, was on average 2.2% (Thomas and Malczewski, 2007). Neglecting it would cause incorrect estimates of C stocks by approximately 4~6% (Thomas and Malczewski, 2007). Thus, understanding how the carbon sequestration ability of the two species is affected by thinning with frozen carbon content considered is essential for evaluating how temperate mixed deciduous and evergreen forest will respond to forest management.

The objective of this study is to investigate how the frozen carbon stocks of Korean pine and Manchurian ash would respond to CTR thinning in different intensities. The dominant species in temperate deciduous mixed forest, Korean pine and Manchurian ash, were studied to provide insight into the following questions: (1) how much carbon stock will be overestimated or underestimated if frozen carbon content is not taken into consideration before and after thinning? (2) Does an individual tree's ability to sequester carbon vary under different thinning intensities (light, medium, heavy, and no thinning), species, and tree size? Here, we take the frozen carbon content into consideration by measuring the frozen carbon content, and then quantify the carbon sequestration ability of trees by calculating individual stem carbon stock and annual carbon stock rate. Based on previous studies on thinning, forest carbon stocks and frozen carbon content, we hypothesize: (1) carbon stocks will be underestimated using conventional carbon content; (2) under light thinning the individual timber carbon stocks and its rate would be highest.

2 Methods

2.1 Study area description

The study area is located in the Jiaohe Management Bureau of the Jilin Province Forest Experimental Zone, northeastern China (43°57'~43°58'N, 127°43'~127°44'E, Figure 1). It is a temperate continental monsoon climate area. The mean annual temperature is 3.8°C; the average monthly temperature ranges from -18.6°C in January to 21.7°C in July. Mean annual precipitation is approximately 700~800mm. The forest soil is dark brown, around 20~100cm deep (He et al., 2018). The vegetation type is coniferous and broad-leaved mixed forest. The dominant species are *Pinus koraiensis*, *Fraxinus mandshurica*, *Juglans mandshurica* Maxim., *Acer mono* Maxim., *Tilia*

amurensis Rupr., *Quercus mongolica* Fisch. ex Turcz. and *Carpinus cordata* Blume. According to the historical records, the last recorded timber harvesting activities took place during the 1960s when approximately 50% of the stock was extracted (Hao et al., 2018). The forest had been strictly protected from human activities since the launch of China's Natural Forest Protection Project in 1998. Commercial logging, hunting, and road construction are not allowed to limit human disturbance to a minimum (Geng et al., 2021).

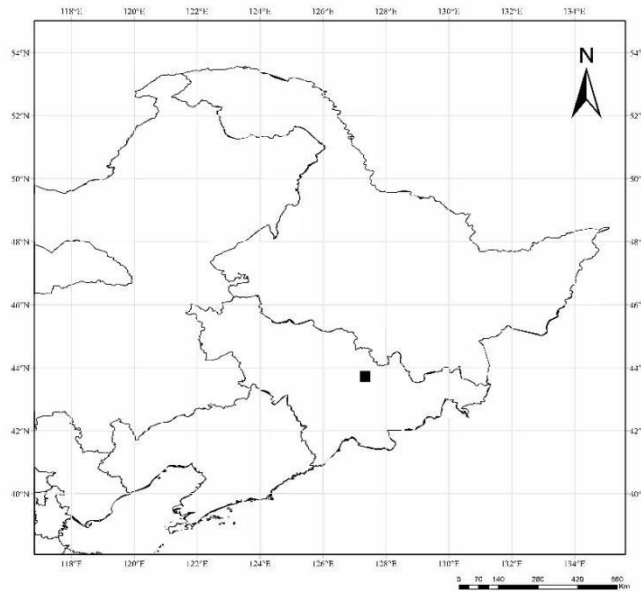


Figure 1 The location of the study area

2.2 Thinning treatments and plot inventory

In July 2011, four permanent plots with a size of 1 ha (100 m × 100 m) were established for thinning. Sites with the same topographical conditions and similar community structures were chosen to establish plots to avoid uncertainty caused by the structural differences. The plots were distributed in field shape and were 100 m apart. Before thinning, a pre-thinning inventory of trees >1 cm in diameter at breast height was carried out to investigate the forest condition. For the convenience of field inventory, each plot was divided into 25 continuous smaller quadrats (20 m × 20 m) using a total station. The species names of woody plants, diameter at breast height (DBH), tree height (H), crown width, and spatial coordinates of all trees were recorded. All these trees were numbered and tagged for long-term observation.

In December 2011, the four plots were designed to have CTR thinning in four intensities respectively: CK (no thinning, 0% of basal area removal), LT (light thinning, 15%), MT (medium thinning, 30%), and HT (heavy thinning, 50%). The smallest thinning DBH is 10 cm. The thinning aimed to maintain crop trees by removing the nearest competing trees or unhealthy trees. Competition among canopies was released. Dominant species like Korean pine and Manchurian ash are mostly retained considering their high economic value. After thinning, the chopped trees were left in the plot to imitate the process of self-thinning, during which the dead trees or fallen trees would stay in the forests, becoming coarse wood debris and providing habitats and protection for various organisms such as insects and fungi (Bunnell and Houde, 2010; Khan et al., 2021). The tree codes of fallen trees were recorded to calculate the actual thinning intensities, which are 0% (CK), 17.25% (LT), 34.73% (MT) and 51.87% (HT) (Table 1). Four repeated plot inventories were done in July 2013, 2015, 2018 and June 2021, respectively.

Table 1 The characteristics of four plots before and after thinning

Site	Stem-number	Total Basal Area	Average Individual	Thinned	Thinned Total	Thinned Individual
	(ha ⁻¹)	(m ² ha ⁻¹)	Basal Area	stem-number	Basal Area	Basal Area
	Pre- / post-thinning	Pre- / post-thinning	(m ² ha ⁻¹ tree ⁻¹)	(ha ⁻¹)	(m ² ha ⁻¹)	(m ² ha ⁻¹ tree ⁻¹)
C	1106 / 1106	30.07 / 30.07	0.0272 / 0.0272	—	—	—
K						
LT	1044 / 958	29.47 / 23.43	0.0282 / 0.0245	86	6.04	0.0703
M						
T	1004 / 823	30.38 / 18.07	0.0303 / 0.022	181	12.31	0.0680
H						
T	1331 / 901	30.64 / 12.26	0.023 / 0.0136	430	18.38	0.0428

2.3 Samples for annual growth measurement

In 2017, tree cores are collected from four plots. Trees with crooked stems, substantial heart-rot, stem abrasion, fungal infections were not sampled. The increment core borer with a bit diameter of 5.15 mm was used to collect the tree cores. All the cores were sealed in plastic straws and taken back to the lab. Cores were mounted in wooden frames using water-soluble glue with the transverse surface facing up. The surface was sanded and polished using successively finer grades of sandpaper (100–1,000 grit size) until optimal surface resolution allowed annual rings visible. Tree-ring widths, which indicates the annual growth, were measured to within 0.001 mm, using the TSAP-Win program (version 0.59 Rinntech) and LINTAB TM6 measuring device (Rinntech, Heidelberg, Germany). All cores were cross-dated by matching patterns of relatively wide and narrow rings to account for the possibility of ring-growth anomalies (e.g.: missing or false rings) or measurement error (Fritts, 1976). The accuracy of assigned dates was further checked by the computer program COFECHA (Holmes, 1983). Tree-ring series poorly correlated with the master series or cannot show clear rings were removed from the final dataset. After removal, 114 cores of Korean pine (DBH: 27.52 ± 1.66 cm; mean \pm SE) and 164 cores of Manchurian ash (DBH: 29.84 ± 0.88 cm) are qualified for future analysis.

2.4 Samples for carbon measurement

Cores used for measuring annual frozen carbon content were collected in the summer of 2018. They were kept with ice bags to minimize the loss of volatile carbon during transportation from field to laboratory (Gao et al., 2016). Since the plots don't allow large quantities of tree core collection after 2017 for the purpose of maintaining forest health, we only selected trees with $DBH \geq 30$ cm, considering the large-diameter trees contribute more to the total carbon stocks. After cross-dating, 12 Korean pine cores and 14 Manchurian Ash cores were selected to measure frozen carbon content. For each core, annual tree segments from 1987-2016 (30 years) were separately excised with clean sharp razor blade under a stereo microscope. The oxidized tissue was removed considering it may have lost volatile carbon or been contaminated.

2.5 Chemical analysis

All segments were placed in open containers and dried by a vacuum freeze dryer (BiLon FD-1A-50) for more than 48 hours. The segments from the same year, same plot, and same species were mixed and kept in one centrifuge tube (2 mL) for the next step. They were ground into a homogenous fine powder using a Retsch MM20 (Germany) grinding machine with 2~4 steel balls (diameter=4 mm). Then, 2~3 mg sample powder from each year was transferred into a clean, dry tin container (a 5×9 mm cup, CHNOS) and weighted using a balance with 1/100,000 precision. Frozen carbon content (C, %) was measured by PE2400 SERIESII (Maryland, United States). Three replications were conducted for each sample and once outliers occurred, additional repetitions were tested to ensure accuracy.

2.6 Statistical analysis

2.6.1 Stand condition

The mean basal area at breast-height (BA [m²]) for Korean pine and Manchurian ash were calculated by Equation 1:

$$\overline{BA} = \frac{\sum_{i=1}^n \frac{\pi D_i^2}{40000}}{n} \quad (1)$$

where D_i [cm] is the diameter at breast height of individual tree i and n is the number of trees. Then, Equation 2 was used to calculate the-mean diameter at breast height (D_g [cm]) for the two species:

$$D_g = \sqrt{\frac{4}{\pi} \overline{BA}} \times 100 \quad (2)$$

The proportion of one species in the plot is defined as BA of one species divided by BA of the whole plot. These values are used to estimate the relative dominance of two species in the plots before and after thinning.

2.6.2 Stem biomass allometric equations

Here, we used the species-specific stem biomass (SB) allometric equations to estimate stem biomass of Korean pine and Manchurian ash (Equation 3 and Equation 4; Wang, 2006).

$$\log_{10}(SB) = 1.908 + 2.258 \log_{10}(D) \quad (3)$$

$$\log_{10}(SB) = 2.116 + 2.316 \log_{10}(D) \quad (4)$$

where D is diameter at breast height (cm). Using these equations, we estimated the individual stem biomass for 2011, 2013, 2015, 2018 and 2021 based on their diameter measured in plot inventory.

2.6.3 Individual stem carbon calculation

Individual stem carbon of 2011, 2013, 2015, 2018 and 2021 was calculated by multiplying the biomass with frozen carbon content of that year measured in section 2.5 (Equation 5). Since the frozen core were collected in 2018, the frozen carbon value of 2018 and 2021 used for calculation is the average of frozen carbon value of 2011, 2013, and 2015.

$$S_{ai} = C_a \times SB_{ai} \quad (5)$$

where S_{ai} is individual stem carbon of tree i of year a , C_a (%) is frozen carbon content of year a , SB_{ai} is stem biomass of individual tree i of year a and a can be 2011, 2013, 2015, 2018 or 2021. Then, conventional carbon stocks of 2011, 2013, 2015, 2018 and 2021 was calculated by multiplying the biomass with 0.5 (Korean pine) or 0.48 (Manchurian ash) as the carbon fraction. 0.5 has been widely used as the conversion coefficient of biomass and carbon stocks (Fang et al., 2007, 2001; Murillo, 1997). However, the carbon content of broad-leaved trees is generally smaller than that of conifers, thus in this study, we used 0.48, a value suggested by IPCC, to be the conventional carbon fraction of Manchurian ash (IPCC,2006).

2.6.4 Annual carbon stocks rate from 1987 to 2016

Based on the DBH of 2015, we reconstructed historical tree diameters from 1987 to 2016. Also, the frozen carbon content of this 30 years was measured year by year according to section 2.5. Then, we estimated the individual stem carbon stock of this 30 years using Equation 3, 4, 5. The difference of the carbon stocks of adjacent years of each tree was considered as annual carbon stocks rate [kg tree⁻¹ year⁻¹]. For each year we take the average value of all cores in that plot to be the carbon stocks rate for that year.

2.6.5 Average annual carbon increase calculation

Individual stem carbon stocks for each tree of 2011, 2013, 2015, 2018 and 2021 can be calculated by Equation 5. The individual stem carbon value of 2011 and 2021 was used to calculate average annual carbon increase during the 10 years after thinning according to:

$$\bar{V} = \frac{SB_{2021} - SB_{2011}}{2021 - 2011} \quad (6)$$

where SB_{2021} and SB_{2011} are stem biomass of 2021 and 2011, respectively.

All analyses were conducted using R software version 3.4.5 (R Core Team, 2020) and figures were plotted using Sigmaplot 14.0.

3 Results

3.1 The influence of CTR thinning on the stand structure

After the stand structure adjusted by CTR thinning, Korean pine and Manchurian ash were at a more dominant position. Both Korean pine and Manchurian ash have a higher proportion of the basal area in the plot after thinning than before thinning (Table 2). They together compose more than 48% of the forest in the plot. Before thinning, in the LT, MT, and HT plots, the proportions of the basal area of Korean pine were less than 20%, but after thinning, the proportion reaches 20.3%, 26.1%, and 23.0% respectively. As for Manchurian ash, its proportion of the basal area increased to 34.3%, 28.0%, and 25.2% in the LT, MT, and HT plots.

Under four treatments, no Korean pine was cut, so its mean DBH didn't change after thinning (Table 3). The mean DBH of Korean pine in MT (33.12 cm) is larger than that in the other three plots (CK: 15.30 cm, LT: 22.49 cm; HT: 16.64 cm; Table 2). Manchurian ash was cut only in MT and HT (Table 2). Before thinning, the mean DBH of Manchurian ash is similar in CK, LT, and HT. After thinning, the mean DBH in MT and HT are decreased to 24.47cm and 26.08cm and are lower than CK and LT. In both MT and HT, the removed Manchurian ash belong to large diameter class (DBH > 30 cm). The detailed diameter class distributions of four plots are shown in Figure 2.

Table 2 The proportion of the basal area of Korean pine and Manchurian ash (%) before and after thinning

Site Species		CK	LT	MT	HT
Korean pine	Before	3.1	15.5	18.2	9.2
	After	3.1	20.3	26.1	23.0
Manchurian ash	Before	20.1	26.1	22.7	17.0
	After	20.1	34.3	28.0	25.2

Table 3 Characteristics of Korean pine and Manchurian ash in four plots before and after thinning

Site	Species	Stem number	Mean	Mean	Basal area	Basal area
		Pre- / post- thinning (ha ⁻¹)	diameter before thinning (cm) (±SE)	diameter after thinning (cm) (±SE)	before thinning (m ² ha ⁻¹)	after thinning (m ² ha ⁻¹)
CK		51 / 51	15.30(±1.03)	15.30(±1.03)	0.9377	0.9377
LT	Korean pine	98 / 98	22.49(±1.38)	22.49(±1.38)	3.8920	3.8920
MT		64 / 64	33.12(±2.23)	33.12(±2.23)	5.5123	5.5123
HT		130 / 130	16.64(±0.84)	16.64(±0.84)	2.8272	2.8272
CK	Manchurian ash	79 / 79	31.22(±1.40)	31.22(±1.40)	6.0458	6.0458
LT		88 / 88	30.81(±1.34)	30.81(±1.34)	6.5602	6.5602

MT	132 / 126	25.80(±0.95)	24.47(±0.88)	6.9033	5.9245
HT	75 / 58	29.80(±1.22)	26.08(±1.15)	5.2296	3.0975

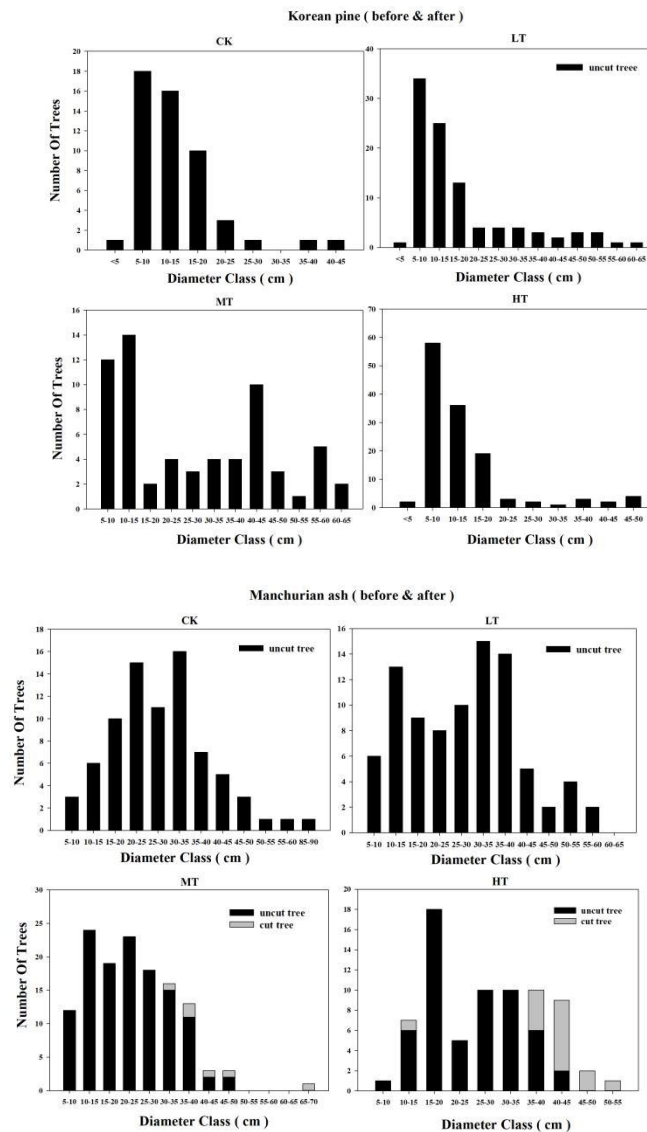


Figure 2 The diameter class distribution of Korean pine and Manchurian ash before and after thinning

3.2 The effects of different thinning intensities on carbon stocks

3.2.1 Frozen carbon stocks estimation

Our study shows that using 0.5 as the carbon fraction of Korean pine will lead to underestimation of 282.8 kg ha⁻¹. Using 0.48 as the carbon fraction of Manchurian ash will lead to underestimation of 2921.9 kg ha⁻¹.

Before thinning, the underestimations of the carbon stocks of Manchurian ash in the four plots were similar, ranging from 949.8 kg ha⁻¹ in CK to 1171.8 kg ha⁻¹ in LT. After thinning, the underestimation of carbon stocks decreased with greater thinning intensities (Figure 3: LT > MT > HT). After light and medium thinning, the underestimation of the carbon stocks of Manchurian ash was increased, while after heavy thinning, the underestimation decreased. In CK, the underestimation of the carbon stocks of Manchurian ash even doubled from 2011 to 2013 (Figure 3, 2011: 1902.2 kg ha⁻¹, 2013: 3913.9 kg ha⁻¹).

Compared with Manchurian ash, the conventional carbon fraction of 0.5 for Korean pine is more appropriate. In MT, using conventional carbon fraction underestimated the carbon stocks of Korean pine in 2011 by 282.8 kg ha⁻¹ but overestimated its carbon stocks in 2013 by 11.6 kg ha⁻¹. Also, the misestimation of Korean pine didn't show an obvious pattern with the intensity of thinning. However, after thinning, especially after light thinning and during 2015-2018, the underestimation is enlarged (LT: 2015: 154.6 kg ha⁻¹; 2021: 199.6 kg ha⁻¹; MT: 2015: 66.6 kg ha⁻¹; 2021: 109.6 kg ha⁻¹; HT: 2015: 41.2 kg ha⁻¹; 2021: 141.6 kg ha⁻¹), so taking frozen content into carbon estimation may be necessary when there is a thinning disturbance.

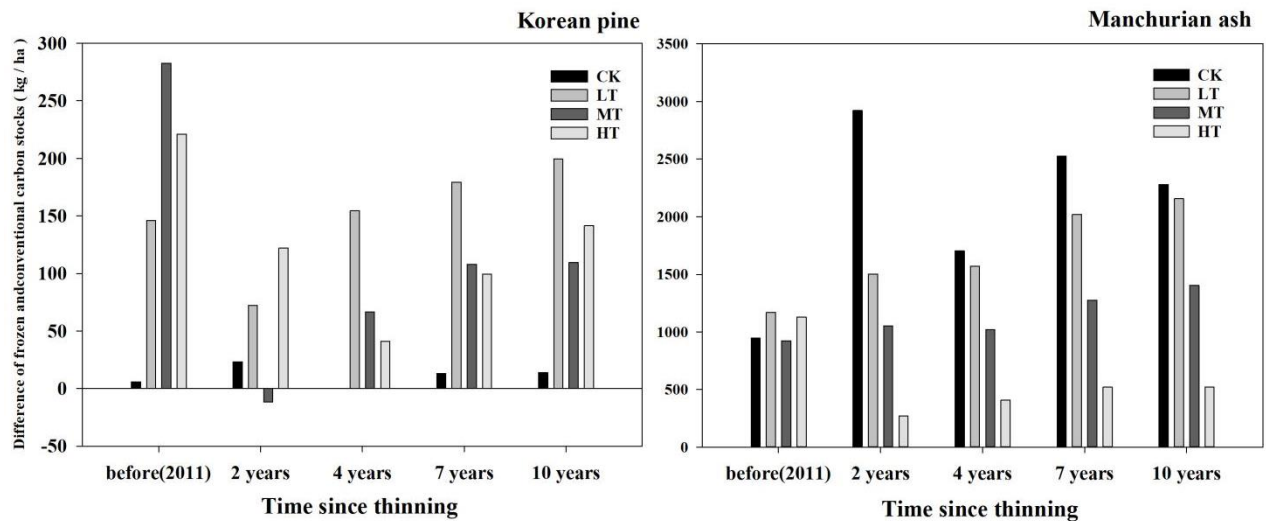


Figure 3 The difference between frozen carbon stocks and conventional carbon stocks in four plots within 10 years after thinning

3.2.2 Individual stem carbon stocks

Thinning can influence carbon stocks at the individual level, and the effect varies with thinning intensities and species. In CK, without thinning disturbance, the individual stem carbon of Korean pine in CK doesn't increase much, from 21.4 kg in 2011 to 25.5 kg in 2021. With light thinning, the individual stem carbon of Korean pine increases steadily during the 10 years after thinning, from 56.9 kg in 2011 to 80.8 kg in 2021 (Figure 4). In MT, the carbon stock decreased in 2013-2015, then increased during 2015-2021, reaching 143.1 kg. Under heavy thinning, Korean pine shows a similar pattern as CK in 2011-2018 and doesn't show an increasing trend until 2018-2021 (Figure 4).

The individual stem carbon of Manchurian ash steadily increases from 2011 to 2018 in the CK plot without thinning disturbance. Light thinning also leads to a steady increase of individual carbon stocks, from 201.1 kg in 2011 to 268.4 kg in 2021. The average increase is 6.7kg year⁻¹. In both MT and HT, the individual stem carbon decreased a little in 2013, which can be attributed to the fact that large trees were removed in these two plots (Figure 2). Nevertheless, in MT, the individual stem carbon soon made up the deficits and got the surplus in 2015, while in HT, it never recovered to the pre-thinning level (Figure 4).

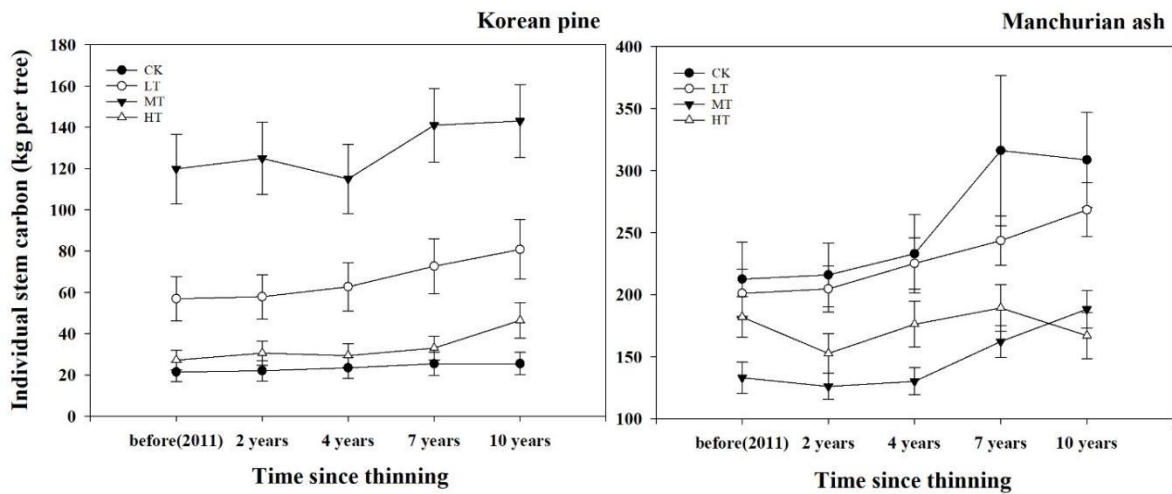


Figure 4 The change of individual stem carbon stocks from 2011 to 2021 in the four plots. Mean \pm SE (error bar) is given.

3.2.3 The trend of annual carbon stocks rate from 1987-2016

To further analyze how the annual carbon stocks rate changes with time and the effects of thinning, we reconstructed the historic diameters through tree ring width. Then, we multiplied annual biomass growth, which was calculated by allometric equations (Wang, 2016), with annual carbon content to get annual carbon stocks. The difference between two adjacent years is identified as annual carbon stock rate.

Usually, the annual carbon stock rate presents periodic variation. The annual carbon stock rate of Korean pine shows a continuous increasing trend during the five years after thinning. After light thinning, the annual carbon stock rate steadily increased from 1.8 kg year⁻¹ in 2012 to 5.2 kg year⁻¹ in 2016. In the MT plot, it increased from -1.3 kg year⁻¹ in 2012 to 3.8 kg year⁻¹ in 2016. In the HT plot, the annual carbon stock rate firstly decreased to -0.2 kg year⁻¹ in 2013 and then increased to 1.6 kg year⁻¹ in 2016. Before thinning (1987-2011), the annual carbon stock rate didn't show an obvious trend (Figure 5).

Manchurian ash shows a different pattern. Before thinning, the annual carbon stock rate of Manchurian ash presented a decreasing trend in the three thinning plots. Even though the trend in the CK plot is a slight increase, the slope is about 0. Thinning didn't change the decreasing trend except in the MT plot, and Manchurian ash in the LT and HT plot even showed a smaller slope after thinning (LT: before: -0.183, after: -0.79; HT: before: -0.266, after: -1.378). With the annual carbon stock rate rises undulating, Manchurian ash in the MT plot transitioned to have a positive slope after thinning (before: -0.116, after: 0.614). However, even if Manchurian ash presents a decreasing trend in annual carbon stock rate (Figure 5), it holds a higher amount of carbon stock than Korean pine, which shows its carbon sequestration value in the stand.

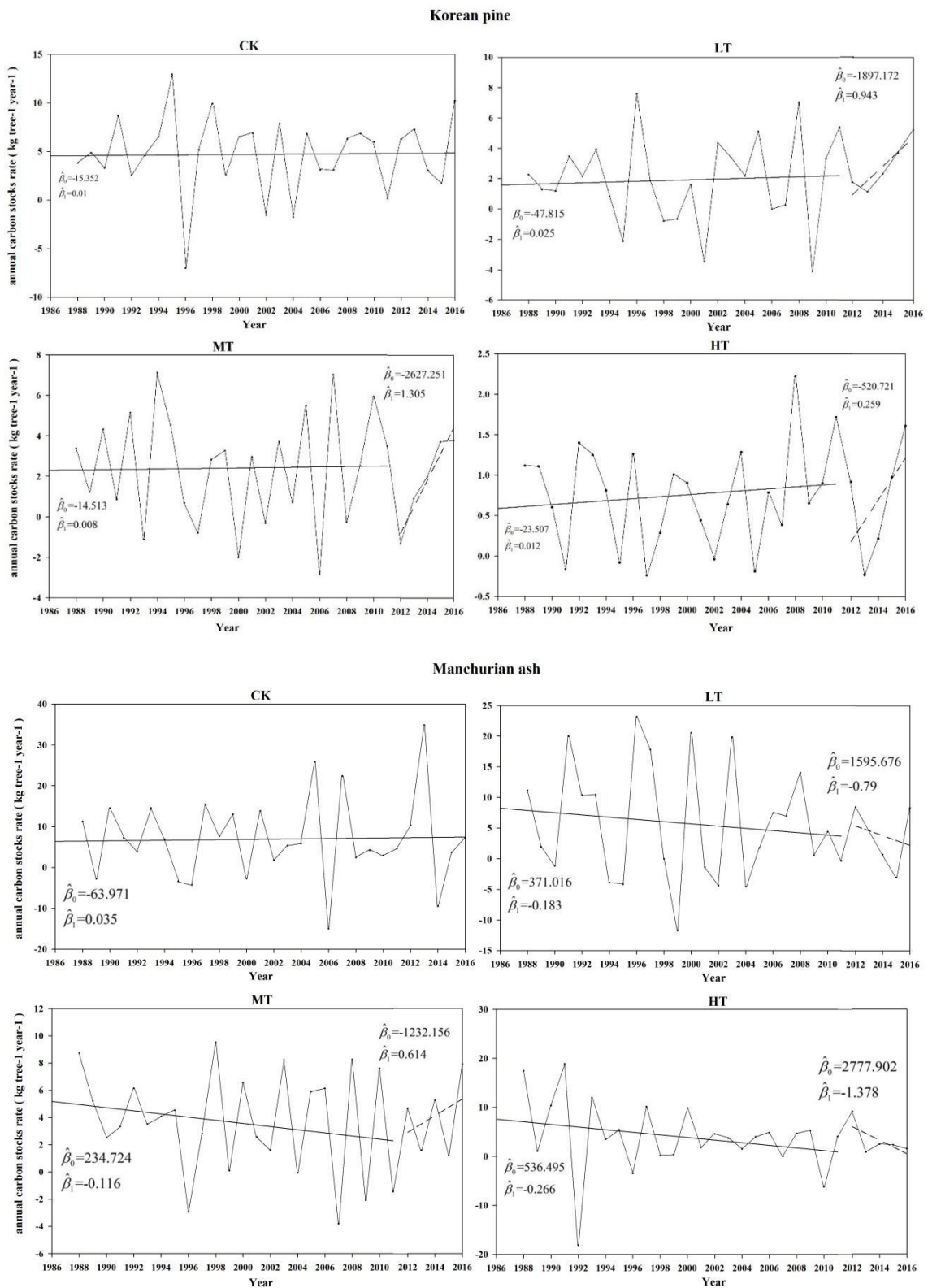
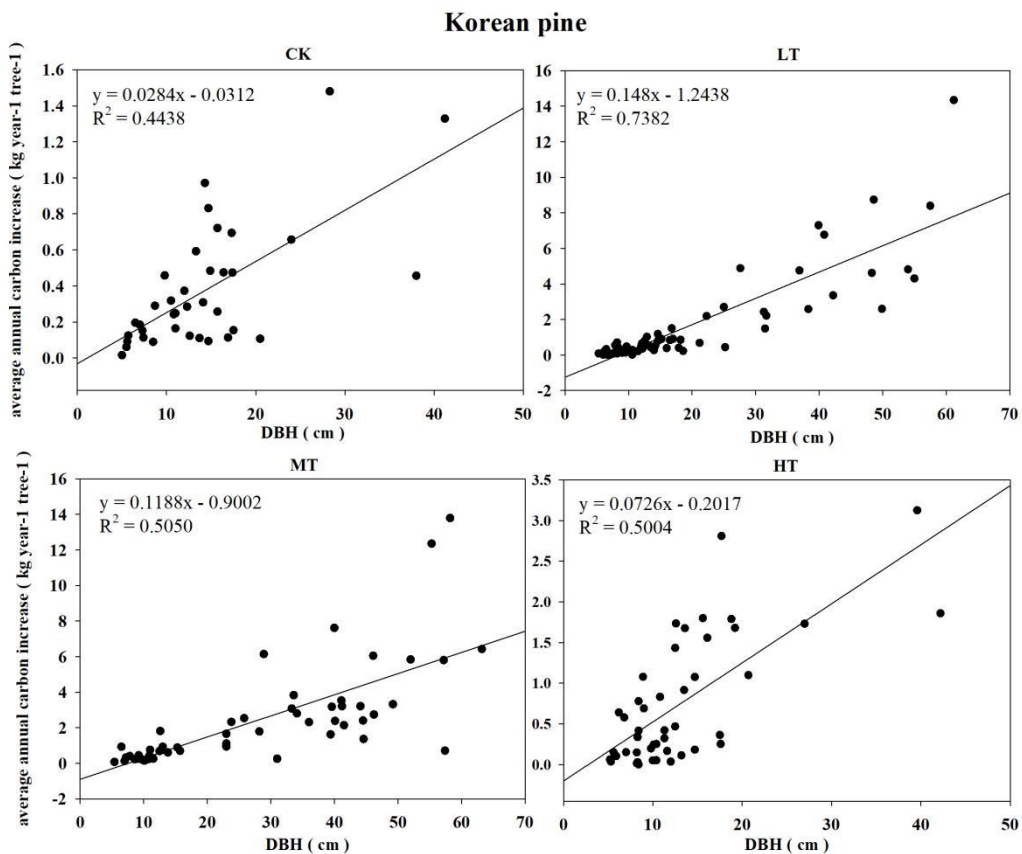


Figure 5 The annual carbon stocks rate from 1987 to 2016 in four plots

Note: β_0 refers to the intercept, β_1 refers to the slope of the fitted simple linear regression. Thinning was conducted in the winter of 2011. The lines of dashes begin from 2012.

3.3 The relationship between average annual carbon increase with diameter class

Korean pine and Manchurian ash in different plots show the same trend with diameter class: the average individual stem carbon increase rate rises with the tree size. This shows that large size trees may have better carbon sequestration ability. Here, the slope of the regression line shows that in the same size class, how much individual stem carbon increase one can get. The slope in LT is highest (slope: LT: 0.148, CK: 0.0284, MT: 0.1188, HT: 0.0726), which indicates that light thinning may be most beneficial for the accumulation of carbon of Korean pine in different diameter classes. Manchurian ash generally has a larger slope than Korean pine. Its largest slope is 0.4749 and occurs in the CK plot. The smallest slope is 0.2387 in HT. One interesting thing is that Manchurian ash remains fast growth speed in CK. This may attribute to the initial large mean diameter in CK, which is very competitive.



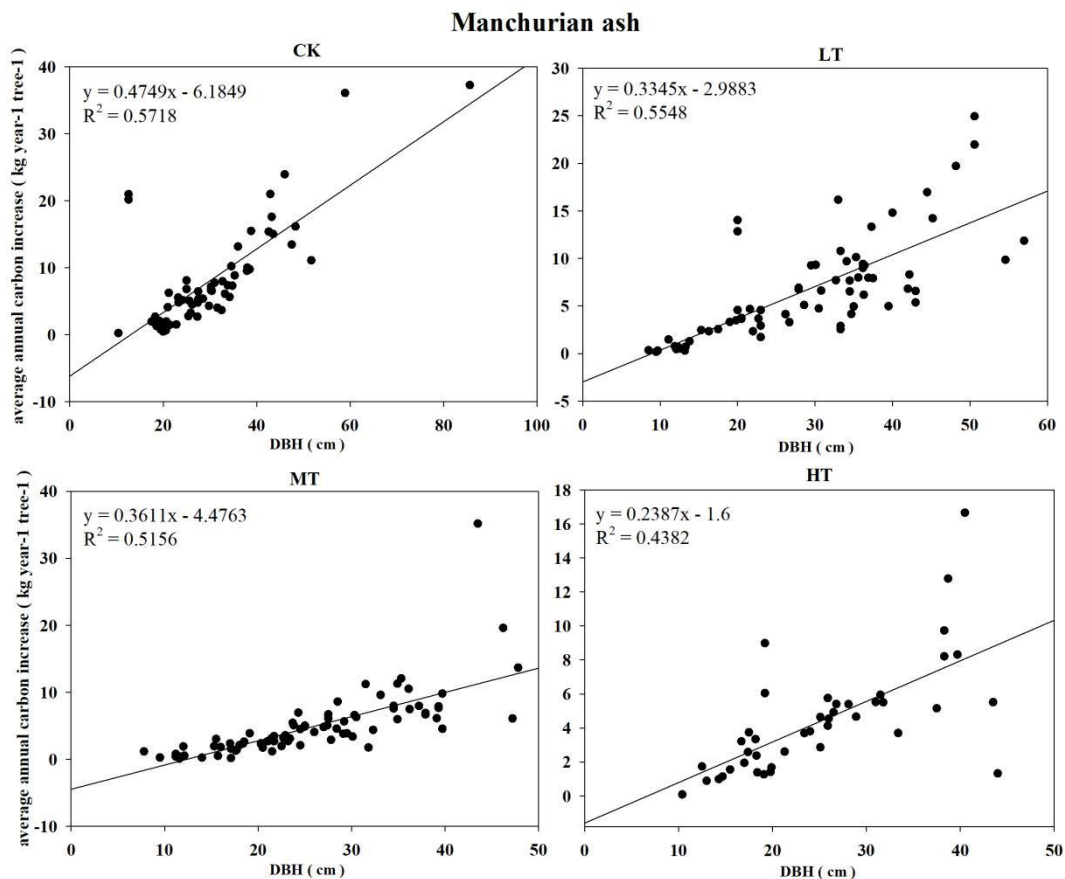


Figure 6 The relationship between average annual carbon increase and diameter class between 2011 and 2021

4 Discussion

The objective of this study was to examine how CTR (crop tree release) thinning in different intensities would influence frozen carbon stocks of Korean pine and Manchurian ash. The results showed that using conventional carbon content will underestimate the carbon stocks of the two species, especially Manchurian ash. In this study, the largest underestimation of Manchurian ash's carbon stock reaches 2921.9 kg ha⁻¹. Also, thinning can impact stem carbon stocks at the individual level. The effect varies with thinning intensities, species, and tree diameter. Finally, thinning may have long-term positive effects on the carbon stocks of Korean pine but have negative effects on Manchurian ash by influencing its average carbon stocks rate.

Variation in carbon content can lead to inconsistency in carbon stocks estimation because carbon stock is determined by multiplying the biomass with the carbon content value (Guerra-Santos et al., 2014; Ma et al., 2018; Nizami, 2012; Wang et al., 2020). The conventional carbon fraction for Korean pine and Manchurian ash are considered as 0.5 and 0.48 respectively. However, this study found that using 0.48 as the carbon fraction of Manchurian ash will lead to the underestimation of carbon stocks reaching 2921.9 kg ha⁻¹ and using 0.5 as the carbon fraction of Korean pine will lead to underestimation of carbon stocks reaching 282.8 kg ha⁻¹. Based on the inventory data of 2018, if one were to use conventional carbon content to estimate the carbon stocks of coniferous and broad-leaved mixed forest in northeastern China, this will lead to underestimation of 6.9 billion kg when no thinning occurs. When thinning in different intensities occurs, this will lead to underestimation of 7.1 billion (LT), 4.6 billion (MT), 2.0 billion (HT) kg respectively.

Thinning can influence the carbon sequestration ability of trees at the individual level, and the effect varies with thinning intensities and species (Figure 4). Light thinning was found to help both species maintain a stable individual stem carbon growth, yet Korean pine and Manchurian ash still have different responses to other thinning intensities. Thinning may promote individual stem carbon stock of Korean pine. During the 10 years after thinning, Korean pine showed increases in individual stem carbon stock in different degrees, while in the CK plot, Korean pine didn't show an obvious trend (Figure 4). When examining the annual carbon stock rate from 1987 to 2016, one will find that Korean pine shows a continuous increasing trend during the five years after thinning (Figure 5). As for Manchurian ash, its individual stem carbon kept a steady rise after light and medium thinning but declined sharply at 10 years from its value at 7 years (Figure 4). Only in the MT plot, the annual carbon stock rate of Manchurian ash transferred from decreasing to increasing (Figure 5). The differential response of Korean pine and Manchurian ash to thinning offers some explanation to previous inconsistent results about thinning's impacts on forest carbon stocks (del Río et al., 2017; Lin et al., 2018; Schaedel et al., 2017; Shuyong et al., 2017).

Since climate is also a potential influencing factor to carbon stock, the correlations between climatic factors and the annual carbon stock rate were examined. The result shows that carbon stock rate rarely has significant correlation with climatic factors at year level, such as annual average temperature, annual average precipitation, SPEI, extreme temperature, annual range of temperature etc. Only the annual maximum and minimum vapor pressure deficit (VPD) have significantly positive effects on carbon stocks. This is because low atmospheric VPD can promote stomatal aperture and stomatal conductance, thus increasing leaf and canopy photosynthetic rates (Wang et al., 2021; Yuan et al., 2019). The annual carbon stock rate have some significant correlations with climatic factors at monthly level, which is consistent with previous research (Yu et al., 2013). Generally, precipitation during the growing season has a positive effect on Korean pine. The annual carbon stock rate of Korean pine is positively correlated to monthly maximum precipitation (CK: April, October; LT: June). Manchuria ash, conversely, is more related to temperature. The monthly lowest temperature in winter has significantly negative effects on carbon stock rate (MT, LT: February; HT: September), while in spring and summer, the monthly annual temperature tends to have positive effect (HT: April, May; MT: May). Moreover, inconsistent correlation between climatic factors and annual carbon stocks rate in each plot indicates that other than climatic factors, the changes of microclimate or competition caused by thinning play much more important role in influencing carbon stocks.

Thinning not only influences forest carbon stocks through impacting tree growth and changing stand biomass but also by influencing carbon content. With the microclimate changed after thinning, the efficiency of carbon sequestration may change (Wang et al., 2020), resulting in variation in annual carbon content. This study found that thinning can increase the water-use efficiency at the tree-level and stand-level especially in a drought year (Wang et al., 2020). Using analysis of variance (ANOVA) to compare the carbon content of five years before thinning (2007-2011) and five years after thinning (2012-2016), we found that light thinning significantly increased the frozen carbon content of both species ($P < 0.05$). The 5-year average frozen carbon content of Korean pine increased from 50.62% to 51.43%, and that of Manchurian ash increased from 52.30% to 53.08%. Since light thinning is widely conducted in forest management (Geng et al., 2021), it is necessary to take frozen carbon content into account when thinning occurs. Interestingly, in CK without disturbance, the change of frozen carbon content resulted in the underestimation of the carbon stocks of Manchurian ash almost doubled (Figure 4, 2011: 1902.2 kg ha⁻¹, 2013: 3913.9 kg ha⁻¹). This further emphasizes that frozen carbon content should be considered in a wider range of scenarios, because natural factors like topography and the initial diameter class of tree can also influence the carbon fraction (Tang et al., 2018; Zhu et al., 2019).

Initial diameter class can influence trees' response to thinning too. For example, Manchurian ash showed even faster individual stem carbon increase when there is no thinning disturbance (Figure 3). This may be attributed to the initial larger diameter class. Through analyzing the relationship between DBH and individual stem carbon increase rate, we found that Korean pine and Manchurian ash in different plots show the same trend: the individual stem carbon increase rate rises with the diameter class (Figure 6), which means large size tree could have a better carbon sequestration ability. This corresponds with previous research: Stephenson et al, 2014 found that the aboveground tree mass growth rate (or, rate of carbon gain) is positively related to tree size ($\log(\text{mass})$) (Stephenson et al., 2014). Compared with Korean pine, Manchurian ash in this study generally has a larger diameter class and benefited less from thinning. Indeed, Manchurian ash had a decreasing trend in its annual carbon stock rate after thinning (Figure 5). This indicates that when managing natural forests, we must consider the development stage of trees. For large-size mature or old trees, thinning may not have a large influence or may even have negative effects. The positive effects of thinning on tree growth and carbon sequestration may only occur at a certain stage or under certain weather (ex: drought) (Wang et al., 2020).

Forest management is considered to be a cost-effective measure for climate mitigation (Griscom et al., 2017). As a management tool, thinning influences stand structure, and the degree of misestimating also depends on the thinning method. In this study, we conducted CTR thinning to adjust density structure, release competition and provide a better environment to the large crop trees. Even the tree number is decreased, the large crop trees have the chance to achieve a faster carbon sequestration rate. Under such kind of thinning, the dominant species: Korean pine and Manchurian ash are almost left untouched, and their carbon sequestration ability was increased. This provides double benefits: the crop trees can provide high-quality wood products, which can store the carbon for 70-100 years and can offset the carbon emissions by fossil fuels (Lindroth et al., 2018; Finkral and Evans, 2008). Moreover, in this treatment, the fallen trees are left in the forests to imitate the natural self-thinning process and become coarse wood debris. Despite the small carbon efflux through the respiration of coarse wood debris, the complete degradation of coarse wood debris takes years to centuries. Even after degrading completely, certain amount of the carbon from wood debris would enter the soil, enhancing the soil carbon content. Thus, leaving the dead wood or fallen wood in the forests could contribute to the forest carbon sequestration (Gough et al., 2007; Magnússon et al., 2016). This indicates that forest management can benefit and augment forest carbon storage. Some have argued that proper thinning and harvests may also bring climate benefits and that carbon-efficient uses of wood should be encouraged (Bellassen and Luysaert, 2014; Churkina et al., 2020).

In this study, even though the annual carbon stock rate of Manchurian ash decreased in some years, it still holds a higher amount of carbon stock than Korean pine. This emphasizes large trees contribute more to the stand carbon stocks due to their fast growth and overall higher annual carbon stocks rate. Also, recent research found that in temporal forest, large trees drive aboveground biomass and its gain and loss better than species diversity and trait composition (Yuan et al., 2021). However, it's crucial to notice here that large tree is not synonym for old tree. "Large tree" or "big-sized tree" can be defined as the largest 1% of trees ≥ 1 cm diameter at breast height (DBH) or all trees ≥ 60 cm DBH depending on the diameter structure of the forest (Ali et al., 2019; Lutz et al., 2018; Yuan et al., 2021). Since one of the aim of CTR thinning is to keep forest healthy and achieve sustainable use, large trees that are no longer in good condition, or trees that have become "over-mature", showing a slowing or stopping growth rate are necessary to be removed. Forest managers are encouraged to maintain those healthy large trees in the stand instead of cutting them down too early. Therefore, it is essential to design silviculture plans under the consideration of species, age, climate, and management goals. Indeed, in both mono-species and

multi-species forests, there occur more and more silvicultural prescriptions aiming at sustainable use of forests (Dore et al., 2012; Pretzsch and Zenner, 2017). Exploring the carbon-friendly thinning strategies and the appropriate rotation is important for the carbon budget of forests in northeastern China.

With the world asking for natural climate solutions and the carbon markets gradually being accepted, higher accuracy in carbon estimation is required. Frozen carbon content should be considered for estimating forest carbon stocks. Our hypothesis about using conventional carbon content may lead to misestimating and light thinning might promote carbon stocks is supported by the data from this study. Korean pine and Manchurian ash show different responses to thinning, which may help explain the inconsistent previous research results. In the future, with more data, we can continue to analyze how thinning would influence the carbon content of all the species in temperate deciduous mixed forest, to get a better idea of the carbon sequestration ability of this kind of forest.

5 Conclusion

This article investigates how the carbon sequestration ability of Korean pine and Manchurian ash would respond to CTR thinning in different intensities and considers the contribution of frozen carbon content. To quantify the individual carbon sequestration ability of trees, we calculate the individual stem carbon stock and annual carbon stock rate by combining plot inventory data and dendroecological methods. The results show that ignoring frozen carbon content may lead to underestimation of the carbon stocks of Manchurian ash by 2921.9 kg ha⁻¹. Using 0.5 as carbon content for Korean Pine may be more appropriate, but still can lead to underestimation of 282.8 kg ha⁻¹. The present findings confirm that using a uniform value may be an oversimplification and frozen carbon is an indispensable part of large-scale carbon stock estimation. Manchurian ash and Korean pine have different response patterns to CTR thinning. Light thinning was found to promote the carbon sequestration of both species and can even significantly increase the frozen carbon content of Korean pine. Despite the intensity, the initial tree diameter also matters. Large trees tend to have a higher individual stem carbon increase rate and contributes more to the stand carbon stocks. This study adds to a growing corpus of research showing that thinning could have the chance to promote forest sequestration if species, tree size, and intensity are all considered.

Future studies should consider the potential effects of frozen carbon content more carefully, and more experiments can be done to see what kind of thinning designs are beneficial to carbon sequestration. Small-scale carbon research focused on several species or locations can benefit large-scale estimation by offering field-based data and giving reference to ecosystem modeling. Accurate forest carbon measurement method can not only help elucidate the global carbon cycle under climate change, but also provide suggestions for sustainable forest management and ecological conservation. Forest conservation may not be the only allowed way in natural forest, proper management may also promote carbon stocks. It is essential to design silviculture plans under the consideration of species, size, climate, and management goals. Although this study is limited to the carbon stocks of two dominant species in temperate deciduous mixed forest in northeast China, the research methods of this paper expanded the scope of future research and will be helpful to accurately evaluate the dynamics of forest carbon reserves.

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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