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Wood density and microfibril angle from pith to bark of a sugi cultivar (*Cryptomeria japonica*, Japanese cedar, Tosaaka) grown in a Nelder plot

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Abstract

The current stumpage price in harvesting of a 50-year-old sugi (Japanese cedar, *Cryptomeria japonica*) plantation is not enough to motivate forest owners to reforest. Therefore, it is vital to reduce the cost for reforestation after logging as well as for logging and wood transportation of the preceding stands. Low initial tree density has been emphasized as one of the important basic measures to reduce directly the reforestation costs. In this study, for better understanding of the mechanical properties and dimensional stability of lumbers from sugi trees grown in low initial tree density, we examined the effects of initial tree density on wood density and microfibril angle of the S_2 layer in the secondary wall of tracheids in rings from pith to bark in a sugi cultivar (Tosaaka) grown in a Nelder plot (initial density zoning symbols; D (4823 trees/ha), E (3349 trees/ha), G (1615 trees/ha), H (1122 trees/ha) and J (541 trees/ha)). Ring width and latewood percentage in J tree (541 trees/ha) were significantly larger and smaller than those of other initial tree density zoning symbol trees, respectively. Average wood density and earlywood wood density in J tree (541 trees/ha) were significantly smaller than those of other initial tree density zoning symbol trees (Tukey's honestly significant difference test (Tukey HSD), $p < 0.05$). However, effects of initial tree density on the microfibril angles (earlywood and latewood) in rings from pith to bark and the transition patterns of microfibril angle from earlywood to latewood in an outer ring (ring number 22) were not significant or very small. Heartwood width in H trees (1122 trees/ha) and J trees (541 trees/ha) were significantly larger than those in other initial tree density zoning symbol trees (Tukey HSD, $p < 0.05$). There were no significant differences of ring width, average wood density, earlywood density and microfibril angle between H trees (1122 trees/ha) and E trees (3349 trees/ha) in many rings from pith to bark (Tukey HSD, $p > 0.05$). Based on these results of Tosaaka, a sugi cultivar grown in a Nelder plot, it was assumed that mechanical properties and dimensional stability of lumbers from sugi trees grown in low initial tree density (1000 trees/ha) might be quite similar to those from sugi trees grown in the regular initial tree density (3000 trees/ha), although lumbers from J trees (541 trees/ha) might be different from those of sugi trees grown in the regular initial tree density (3000 trees/ha). By taking the effects of genetic factor (variation of sugi cultivars) and the interaction between genetic and environmental factor (initial tree density) into consideration, the effects of low initial tree density (1000 trees/ha) on mechanical properties and dimensional stability of lumbers from sugi plantations in Japan might be smaller than the effects recognized in Tosaaka in this study. In this study, we did not examine the effects of initial tree density on the

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knots and the cross grain of lumbers. Low initial tree density might increase the negative effects of knots and cross grain on mechanical properties of lumbers. However, recent wood processing technology could minimize these negative effects.

Keywords: Low initial tree density, Japanese cedar, Wood density, Microfibril angle, Nelder plot

Introduction

Sugi (*Cryptomeria japonica*, Japanese cedar) is an important afforestation tree species in Japan, mainly used as a structural component of wooden structures. According to the income from harvesting of a 50-year-old sugi plantation, the current stumpage price is not enough to motivate forest owners to reforest [1]. Therefore, it is vital to reduce the cost for reforestation after logging as well as for logging and wood transportation [1]. Recently, the Forestry Agency promotes to introduce an integrated harvesting and planting system for saving costs for reforestation [1]. In this system, harvesting, land preparation and planting were done simultaneously or sequentially using forestry machine [1]. In addition, low initial tree density has been emphasized as one of important basic measures to reduce directly the reforestation costs [2], and the effects of low initial tree density on the initial silvicultural cost, growth rate and wood properties of trees have been examined. In hybrid larch (*Larix gmelinii* var. japonica × *L. kaempferi*), it was reported that the silvicultural costs for the initial 6 years (costs for site preparation, purchase of seedlings/rooted-cuttings, planting, weeding and application of rodenticide) were reduced by 30–40% in the stands with lower initial tree density (< 1000 trees/ha) as compared with those with regular initial tree density (1900 trees/ha) [3].

Initial tree density may affect not only the reforestation costs but also tree growth and wood properties of logs in the plantations. In 34-year-old hinoki trees (*Chamaecyparis obtusa*, Japanese cypress), the effects of initial tree density on the growth of trees were examined, and it was assumed that the yield rate of clear wood with fewer knots from trees grown at a lower initial tree density (1000 trees/ha) was lower than those in wood from trees grown at higher initial tree densities (1500, 2000 and 3000 trees/ha) [4]. In young Douglas firs (*Pseudotsuga menziesii*) grown at different initial tree densities (initial tree density: 309–18,730 trees/ha), it was shown that the mean and maximum nodal branch diameters increased with decreasing initial tree density [5]. In 48-year-old black spruce trees (*Picea mariana*) grown at four different initial tree densities (1372, 2066, 2500 and 3086 trees/ha), it was reported that wider spacing increased the diameter and number of knots and decreased the bending strength and stiffness [6]. In 24-year-old *Pinus radiata* grown at four final tree densities (200, 350, 500, and 1100 stems/ha), a greater wood density, smaller microfibril angle of the S_2 layer in the secondary wall of tracheids (MFA) and a larger modulus of elasticity (MOE) were observed in trees grown at the highest final tree density compared to those grown at the lowest final tree density [7]. Many studies on sugi trees grown at different tree densities have been performed. In 41-year-old sugi trees (unknown genotype) grown at different initial tree densities (400, 630, 1110, 2500 and 10,000 trees/ha) in Taiwan, it was reported that plantation spacing had positive effects on the percentage of heartwood and had no effect on the ring number of the boundary between juvenile and mature wood [8]. The most densely planted trees had the greatest values for air-dried density and mechanical properties, and the knot diameter ratio had a negative effect on the mechanical properties of the sugi lumber [9, 10]. In 35-year-old sugi trees (unknown genotype) grown at different initial tree densities (1500, 3000, 6000 and 10,000 trees/ha), trees grown at lower initial tree densities had significantly larger ring width and knot diameter than trees grown at higher initial tree densities, although there was no effect of initial tree density on wood density or MFA [11]. In 13-year-old sugi clone trees grown at three different initial spacings (1.8 × 1.8 m, 1.4 × 1.4 m and 1.0 × 1.0 m), densely planted sugi clones had smaller ring widths and larger wood density [12], and in 28-year-old sugi cultivars grown at different initial tree densities (1500, 3000 and 5000 trees/ha), densely planted sugi cultivars had slightly higher mature wood density and mechanical properties [13]. The effect of the difference of initial tree density on wood properties was smaller than that of genetic variation [12, 13]. From these results, it was recognized that lower initial tree density induced a larger knot diameter, smaller wood density, larger ring width and smaller mechanical properties in coniferous plantation species than higher initial tree density, although lower initial tree density is a very effective technique for reducing reforestation costs and accelerating tree growth.

However, to obtain more accurate information on sugi trees grown at different initial tree densities, experimental plots with smaller effects of other factors on anatomical wood properties should be used. Nelder plots were developed to analyze the impact of a continuous range of densities on the yield of agricultural crops over a smaller area than traditional designs to minimize the differences in yield related to variability within a site [14]. Researchers have utilized this design in forestry, and this plot consists of a series of concentric circles that represent systematic changes in tree density at a constant proportional rate, with trees planted at the intersection of an arc and a spoke and in roughly square geometry [5]. Therefore, growing a sugi cultivar in Nelder plots can be assumed to minimize the effects of the other factors (genetic variation, difference of environmental condition) that have possible effects on anatomical wood properties. In previous study on a sugi cultivar planted in Nelder plots (376, 541, 779, 1122, 1615, 2326, 3349, 4823 and 6944 trees/ha), we reported that the height-to-diameter ratio (*H*/*D* ratio) and stem stiffness increased with the increase of the initial tree density [15]. In 35-year-old sugi trees, based on an analysis of wood volume density (m^3/ha) and the wood quality of trees grown at different initial tree densities (376–10,000 trees/ha) in a Nelder plot with a sugi cultivar, it was concluded that moderate density (2000–2800 trees/ha) is the appropriate initial tree density for low-cost forestry [16]. The conclusions in these studies were obtained based on the stem stiffness measured by the tree-bending method [15] and stress wave propagation velocity measured on the stem surface [16]. These indices, especially stress wave propagation velocity, related to the stiffness of the outermost layer of the stem [17, 18]. The boxed-heart square timbers without knots on the surface had been used for conventional Japanese constructions. Recently, laminated timbers for wooden constructions would be preferred to boxed-heart square timbers in Japan, because mechanical properties and dimensional stability became important in comparison with the beauty of wood surface in timbers. Wood density and MFA had significant effects on mechanical properties and dimensional stability of sugi lumbers [19, 20]. In addition, the knots in lumbers were removed in the manufacturing process of laminated timbers. Therefore, for efficient use of logs from sugi plantations, variations of wood density and MFA from pith to bark would be very important to understand the mechanical properties and dimensional stability of the end products, instead of the number and diameter of knots.

In this study, we report the effects of initial tree density on the anatomical wood properties of a sugi cultivar planted in a Nelder plot. The objective of this study was to examine (1) growth traits, (2) radial variation of ring width, wood density and MFA, and (3) the heartwood width and ratio at 1.5 m above the ground in the trunks of a sugi cultivar (Tosaaka) grown at different initial tree densities. The current regular initial tree density for sugi plantations is 3000 trees/ha in Japan. Based on the wood properties, we discussed on the possibility of low initial tree density (1000 trees/ha) for reducing reforestation costs in comparison with the regular initial tree density (3000 trees/ha) for sugi plantations in Japan.

Materials and methods

Sample trees and core samples for measurements of wood properties

A 38-year-old sugi cultivar (Tosaaka) planted in a variable initial tree density Nelder plot was used. The initial tree density ranged from 376 to 10,000 trees/ha. The experimental plot used to analyze initial tree density was established in 1974 on a hillslope site within a stand in the national forest in Nichinan city, Miyazaki prefecture, Japan. No silvicultural practices (thinning, pruning) had been carried out for the plot. The plot used for this study was located in the southern part of Miyazaki Prefecture, and the altitude of the plot used for this study was 520 m. The average annual temperature and precipitation (1974–2011) in Miyazaki city (located just north of Nichinan city) were 17.5 °C and 2479.7 mm, respectively.

In this study, as shown in Table 1, we selected 45 trees (five initial tree density zoning (J: 541, H: 1122, G: 1615, E: 3349 and D: 4823 trees/ha) × nine trees in each initial tree density zoning = 45) as sample trees. We did not examine the survival rate of planted trees and the current stocking levels in this study. Because we just focused on the relationship between the wood properties of lumbers and initial tree density related to the initial cost of sugi plantations. The Nelder plot used in this study was different from the plot used in our previous study [15], but the same plot used in another our previous study on annual ring formation and plant hormones [21]. The sample trees used in this study were different from the sample trees used in our previous study [21]. As shown in Fig. 1, two pith-to-bark core samples were obtained at 1.5 m above the ground in each sample tree in 2011 (45 trees × two core samples = Total ninety core samples). One and another core sample from each sample tree was used for wood density measurement (X-ray densitometry) and MFA measurement, respectively.

Measurement of wood density

Wood density was measured using X-ray densitometry, according to the previous studies [22, 23]. From each sample tree, an increment cores were cut into 2 mm-thick strips, as shown in Fig. 1. Removal of the extractives in heartwood was not done in this study. The grayscale image of the strips was obtained using soft X-ray apparatus (SOFTEX Co. Ltd., Japan), digital X-ray senser NAOMI NX-04H (RF Co. Ltd., Japan, software (NAOMI)) with calibration wedge. The soft X-ray radiation was done at 30 kV and 15 mA for 0.1 s. The

Table 1 Initial tree densities and growth traits of sample trees

Symbol	Density (trees/ha)	Age	*n*	*H* (m)	DBH (cm)	HCB (m)	*H/D* ratio
D	4823	38	9	17.4 (1.2)^{a}	22.7 (3.5)^{a}	10.8 (1.1)^{a}	77.6 (8.5)^{a}
E	3349	38	9	18.3 (2.4)^{a}	28.5 (4.7)^{ab}	11.5 (1.0)^{a}	65.3 (9.2)^{b}
G	1615	38	9	18.3 (1.3)^{a}	27.7 (3.8) ^{ab}	11.0 (1.4)^{a}	66.9 (7.2)^{b}
H	1122	38	9	19.4 (2.6)^{a}	34.9 (7.7)^{b}	9.2 (1.9)^{a}	56.8 (7.3)^{b}
J	541	38	9	19.7 (2.3)^{a}	43.1 (8.7)^{c}	5.9 (2.5)^{b}	46.5 (4.7)^{c}

The values represent averages in three trees; values in parentheses indicate standard deviations. Different characters show significant differences among density-zoning symbols (Tukey HSD), $p < 0.05$)

H/D ratio was calculated by the following formula. H/D ratio = tree height (m)/diameter at breast height (cm) × 100

Symbol initial tree density zoning symbol, Density initial tree density, *n* number of sample trees, *H* tree height, DBH diameter at breast height, HCB height at crown base, H/D ratio height-to-diameter ratio

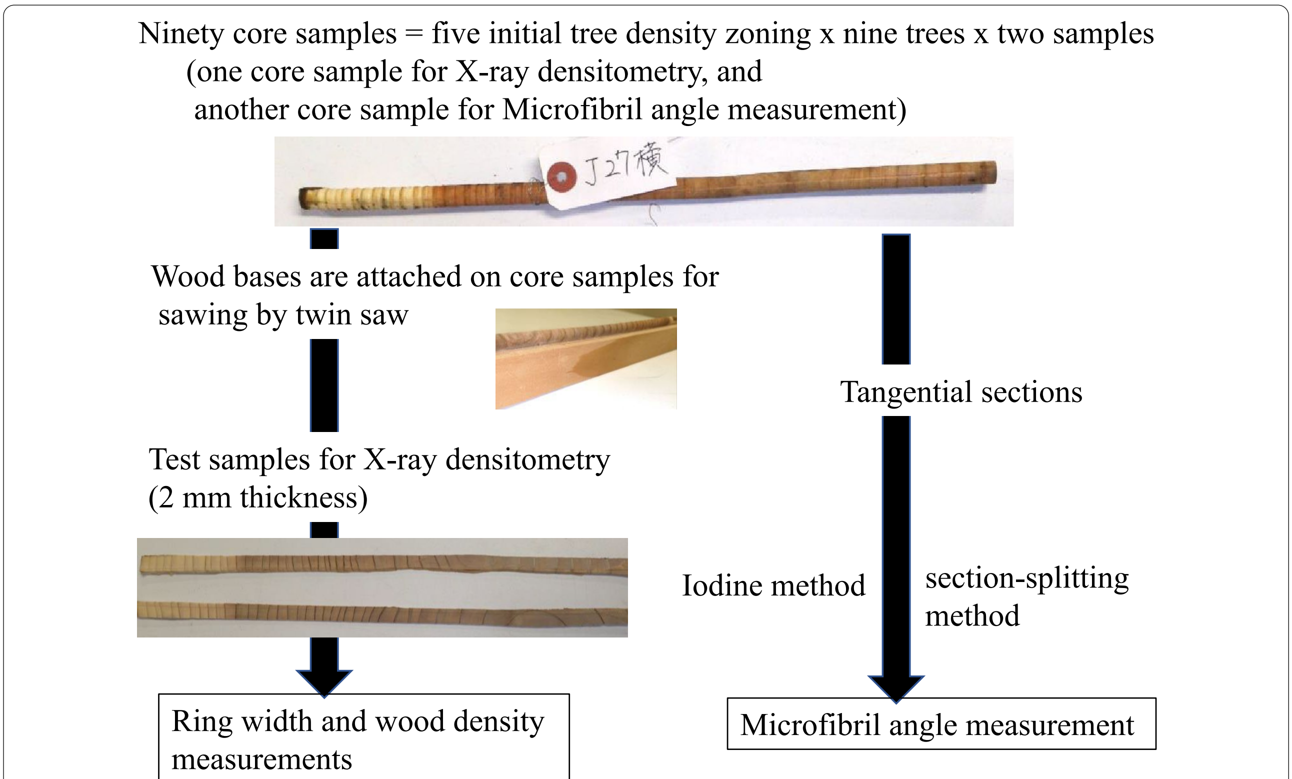

Fig. 1 Samples for X-ray densitometry and Microfibril angle measurement

resolution of the grayscale image (750 dpi) obtained by the digital X-ray senser was smaller than those in the previous study using films (2400 dpi) [23]. The grayscale images of calibration wedge and wood samples with different densities (*Talia japonica,* samples ranged from one to five layers) were also obtained and then calibration curve was obtained using image analysis software (Image J [24]). Wood density and ring width of the strips was measured using the image analysis software. Demarcation between earlywood and latewood in each ring was 550 kg/m^3 [25]. We conducted X-ray densitometry analysis on ring number from 7 to 36. The rings near the pith in some core samples include the knots, and the knots affect the accuracy of wood density measurements. Therefore, we did not measure wood densities on rings near the pith. As previously described, core samples were obtained at 1.5 m above the ground in 38-year-old sugi trees. Some core samples lack the ring number 37 and

38 because of variation of height growth at younger age among sample trees and the damage on core samples during sampling. We decided the ring number 36 as the outermost ring for X-ray densitometry analysis. As previously reported [4–6], core samples from low initial tree density trees had the large number and size of knots. Number of trees for wood density in each initial tree density zoning reduced, because we removed some 2 mm-thick strips with large knots from the investigation and the numbers ranges from five to nine.

Measurement of MFA

The sections for MFA measurement were taken from the different core samples from the core samples for wood density measurement. The MFA of the tangential wall was measured from 24-µm-thick tangential sections cut from the earlywood and latewood of ring numbers 5, 10, 15, 20, 25 and 30 of each core sample. MFA was measured by the iodine-staining method [26]. I_2 crystallizes in gaps between microfibrils in sections were observed with a light microscope. In light microscopy, MFA was measured using image analysis software (Image J [24]). The MFA of each ring was obtained by averaging the MFA of 30 tracheids.

The transition from earlywood to latewood in MFA in an annual ring was also examined. MFA decrease gradually from earlywood to latewood. However, initial tree density might affect the pattern in transition from earlywood to latewood in MFA in an annual ring. In this study, section-splitting method [27] was used to examine the transition from earlywood to latewood in MFA in an annual ring. We want to measure MFAs in four relatively equal position in an annual ring. Therefore, we selected this method, because we could measure MFA of 30 tracheids in one section by this method. Tangential sections were sandwiched with water between two slide glasses, frozen in liquid nitrogen, and split with the glass slides, instead of using epoxy resins. We can observe many striations along microfibrils occurred during preparation of samples. To examine the accuracy of the section-splitting method, we cut a tangential section in half in tangential direction, and measured MFAs of the two sections by iodine-staining method and section-splitting method, respectively. As shown in Fig. 2, it was assumed that there was no difference between MFA values measured by the both methods.

Measurement of heartwood width and ratio

We measured the heartwood width and ratio using the 2 mm-thick strips from core samples shown in Fig. 1. The boundary between heartwood and sapwood was decided based on the change of color in the 2 mm-thick strips.

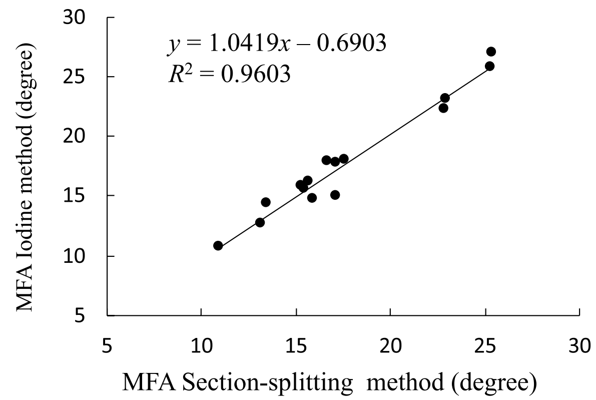

Fig. 2 Relationship between the MFAs measured by different methods. Sections were obtained from earlywood, transition wood from early to latewood and latewood in the ring numbers 3, 7, 11, 15, 19 and 23 of a sugi cultivar (Tosaaka)

Results

Growth traits of sample trees with different initial tree densities

As shown in Table 1, there were significant differences in diameter at breast height (DBH), H/D ratio and height at crown base (HCB) among initial tree densities (Tukey HSD, $p < 0.05$). Tree height decreased with increasing initial tree densities, although there was no significant difference in tree height. These results agreed with those of previous studies [4, 7, 8] and our previous study [15]. DBH, HCB and H/D ratio in J trees (541 trees/ha) were significantly different from those in the other initial density zoning symbol trees (Tukey HSD, $p < 0.05$). There was no significant difference in growth traits between H trees (1122 trees/ha) and E trees (3349 trees/ha), although lower initial tree density induced larger average value of DBH and H, and smaller average value of HCB and *H*/*D* ratio.

From these results, it was shown that initial tree density had larger effects on diameter growth than those on height growth. It was assumed that there was no significant difference of growth traits between in low initial tree density (1000 trees/ha) and in the regular initial tree density (3000 trees/ha) in this sugi cultivar (Tosaaka).

Ring width and latewood percentage of sugi trees with different initial tree densities

As shown in Fig. 3, lower initial tree density induced larger ring width (RW), especially in initial growth. As shown in Table 2, ring width of both H trees (1122 trees/ha) and J trees (541 trees/ha) were significantly larger than those of other initial tree density zoning symbol

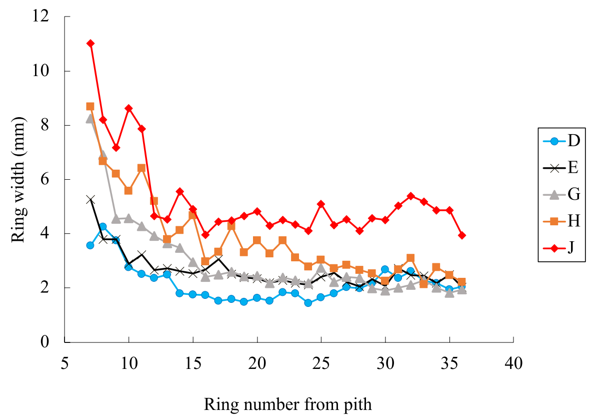

Fig. 3 Effects of initial tree density on ring width. Each plot shows an average value in a ring number among sample trees of each tree density zoning symbol. Number of sample trees of each initial tree density zoning symbol range from five to nine. Initial tree density zoning symbols were the same, as shown in Table 1

trees until ring number 22, although RW of only J trees (541 trees/ha) were significantly larger than those of other initial tree density zoning symbol trees from ring number 25 to the outer most ring (Tukey HSD, $p < 0.05$). Significant difference between H trees (1122 trees/ha) and E trees (3349 trees/ha) was recognized only in ring number ten, eleven and fifteen (Tukey HSD, $p < 0.05$). Average values of latewood percentage (LP) in lower initial tree density trees were smaller than those in higher initial tree density trees (Fig. 4), although there was no significant difference in many rings (Table 2). Significant difference of LP between H trees (1122 trees/ha) and E trees (3349 trees/ha) was recognized only in ring number 25 (Tukey HSD, $p < 0.05$).

From these results, it was recognized that RW and LP in J tree (541 trees/ha) were significantly larger and smaller than those of other initial tree density zoning symbol trees, respectively. However, it was assumed that there was very small difference of ring width and latewood percentage in this sugi cultivar (Tosaaka) between in low initial tree density (1000 trees/ha) and in the regular initial tree density (3000 trees/ha).

Wood density of sugi trees with different initial tree densities

The wood density of sugi showed its highest value near the pith then decreased and became stable toward outer rings [12]. As shown in Fig. 5, this inherent characteristic of sugi in radial variation of average wood density (AWD) were also recognized in trees grown in different initial tree densities. AWD of J trees (541 trees/ha) were significantly smaller than those of other initial tree density zoning symbol trees in almost rings (Fig. 5; Table 2, Tukey HSD, $p < 0.05$). Significant difference between H trees (1122 trees/ha) and E trees (3349 trees/ha) was recognized only in ring number 25 and 31 (Tukey HSD, $p < 0.05$). As shown in Fig. 6, earlywood density (EWD) near the pith was also large and EWD decreased with ring number. EWD of J trees (541 trees/ha) were significantly smaller than those of other initial tree density zoning symbol trees in many rings (Fig. 6; Table 2, Tukey HSD, $p < 0.05$). Significant difference of EWD between H trees (1122 trees/ha) and E trees (3349 trees/ha) was not recognized in any rings. As shown in Fig. 7, latewood density (LWD) had large variation with ring number in each initial tree density zoning symbol tree. Significant differences of LWD were recognized only eight rings (Table 2, Tukey HSD, $p < 0.05$). Significant difference of LWD between H trees (1122 trees/ha) and E trees (3349 trees/ha) was recognized in ring number 25, 26 and 36 (Table 2, Tukey HSD, $p < 0.05$).

From these results, it was recognized that AWD and EWD in J tree (541 trees/ha) were significantly smaller than those of other initial tree density zoning symbol trees. However, it was assumed that there was only small difference of AWD, EWD and LWD in this sugi cultivar (Tosaaka) between in low initial tree density (1000 trees/ha) and in the regular initial tree density (3000 trees/ha).

Microfibril angle of sugi trees with different initial tree densities

As shown in Fig. 8 and Table 2, MFA of both earlywood and latewood in the rings near the pith (ring number five) in J trees (541 trees/ha) were significantly larger than those of other initial tree density zoning symbol trees (Tukey HSD, $p < 0.05$). However, effects of low initial tree density on MFA were unclear in the other rings. Significant difference of MFA between H trees (1122 trees/ha) and E trees (3349 trees/ha) was not recognized in any rings. MFAs in earlywood were larger than those in latewood (Fig. 8). Therefore, if the initial tree density affects the pattern of transition in MFA in an annual ring, the pattern of transition from earlywood to latewood in MFA in an annual ring also could affect the mechanical properties of lumbers. As shown in Fig. 9, there was no significant difference in the pattern of transition from earlywood to latewood in MFA in ring number 22 among initial tree density zoning symbol trees. Based on Fig. 8, MFAs in ring number twenty in each initial tree density zoning symbol trees showed almost 30°. Therefore, we considered the tracheids with MFAs $\geq$ 30 and 35° as the tracheids with unusually large MFAs. As shown in Table 3, there was no significant difference in the numbers of tracheids with larger MFAs $\geq$ 30 and 35° in ring number 22 among initial tree density zoning symbol trees.

Table 2 Significant difference of anatomical wood properties among initial tree density zoning symbols

RN	RW	LP	AWD	EWD	LWD	MFA(E)	MFA(L)
5						DH-J	D–J
7	D-GHJ, E–J	*ns*	*ns*	*ns*	*ns*		
8	*ns*	*ns*	*ns*	*ns*	*ns*		
9	DE-J	*ns*	DG-J	*ns*	*ns*		
10	DE-H, DEGH-J	*ns*	DE-J	*ns*	*ns*	*ns*	*ns*
11	DEG-H, DEG-J	*ns*	DG-J	D–J	*ns*		
12	D-HJ	*ns*	*ns*	DG-J	*ns*		
13	DE-J	*ns*	*ns*	*ns*	*ns*		
14	DE-J	*ns*	D–J	D–J	D-GH		
15	DEG-H, DEG-J	D-EHJ	D–H, DG-J	*ns*	*ns*	*ns*	*ns*
16	DG-J	*ns*	D–J	D–J	*ns*		
17	D-EGHJ, EGH-J	*ns*	DG-J	*ns*	*ns*		
18	D–H, DEG-J	D-EGHJ	D-GHJ	*ns*	*ns*		
19	D–H, DEG-J	*ns*	D–J	*ns*	D-EH		
20	D–H, DEG-J	*ns*	*ns*	*ns*	*ns*	*ns*	*ns*
21	D–H, DEG-J	D-EGHJ	D-GHJ	*ns*	*ns*		
22	D–H, DEG-J	D-HJ, E–J	D–H, DEG-J	*ns*	*ns*		
23	DEG-J	*ns*	DE-J	DEGH-J	*ns*		
24	D–H, DEGH-J	*ns*	*ns*	*ns*	*ns*		
25	D–J	E-HJ	DE-H, DEG-J	DE-J	E-HJ	*ns*	*ns*
26	DEGH-J	G–H, DG-J	DEG-J	DG-J	E-DGHJ		
27	DEGH-J	*ns*	EG-J	DEGH-J	*ns*		
28	DEG-J	D-GHJ	D-GH, DEG-J	DE-J	D–H		
29	DEGH-J	D-HJ, E–J	D–H, DEG-J	*ns*	D-GHJ		
30	DEGH-J	*ns*	DEG-J	DEGH-J	*ns*	D–E	D–E
31	DEGH-J	*ns*	E–H, EG-J	EG-J	E–G		
32	DEG-J	E–J	DEG-J	DEG-J	*ns*		
33	DEGH-J	E–J	DEG-J	E–J	*ns*		
34	DEGH-J	*ns*	EG-J	EGH-J	*ns*		
35	DEGH-J	*ns*	EG-J	EG-J	*ns*		
36	EGH-J	E–J	E–J	E–J	E-DGHJ		

Significant difference of anatomical wood properties among initial tree density zoning symbols were examined (Tukey HSD)

ns no significant difference ($p \geq 0.05$), DE-J in the row of RW means that ring width in J trees were significantly different from those in D tress and E trees ($p < 0.05$). RN ring number, RW ring width, LP latewood percentage, AWD average wood density, EWD earlywood density, LWD latewood density, MFA(E) MFA of earlywood tracheids, MFA(L) MFA of latewood tracheids

From these results, it was recognized that the effects of initial tree density on MFA might be small in comparison with the effects on RW and AWD. Therefore, it was assumed that there was very small difference of MFA in this sugi cultivar (Tosaaka) between in low initial tree density (1000 trees/ha) and in the regular initial tree density (3000 trees/ha).

Heartwood width and heartwood ratio of sugi trees with different initial tree densities

Heartwood width in H trees (1122 trees/ha) and J trees (541 trees/ha) were significantly larger than those in other initial tree density zoning symbol trees (Fig. 10). Heartwood ratio in J trees (541 trees/ha) were significantly larger than those in other initial tree density zoning symbol trees (data not shown). This result was consistent with previous study [8]. Mean of heartwood width in H trees (1122 trees/ha) was larger than that of E trees (3349 trees/ha), although there was no significant difference between both trees. Therefore, it was assumed that trees grown at lower initial tree densities could produce many lumbers with heartwood only, and these lumbers were expected to have superior natural durability because of the absence of sapwood. From these results, it was assumed that lower initial tree density induced a larger volume of heartwood per tree.

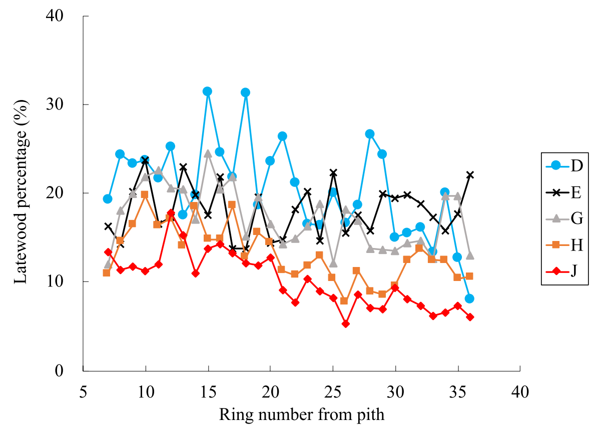

Fig. 4 Effects of initial tree density on latewood percentage. Each plot shows an average value in a ring number among sample trees of each tree density zoning symbol. Number of sample trees of each initial tree density zoning symbol range from five to nine. Initial tree density zoning symbols were the same, as shown in Table 1.

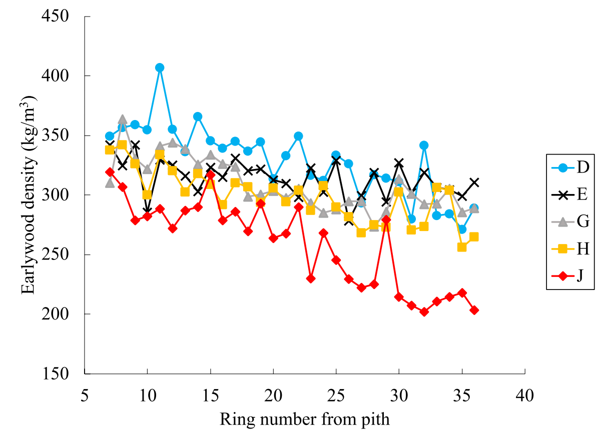

Fig. 6 Effects of initial tree density on earlywood density. Each plot shows an average value in a ring number among sample trees of each tree density zoning symbol. Number of sample trees of each initial tree density zoning symbol range from five to nine. Initial tree density zoning symbols were the same, as shown in Table 1

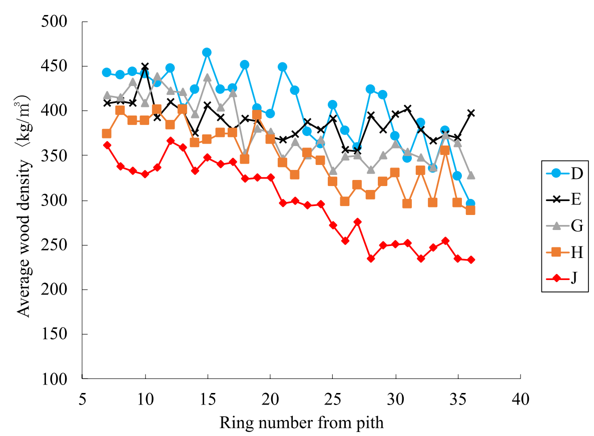

Fig. 5 Effects of initial tree density on average wood density. Each plot shows an average value in a ring number among sample trees of each tree density zoning symbol. Number of sample trees of each initial tree density zoning symbol range from five to nine. Initial tree density zoning symbols were the same, as shown in Table 1.

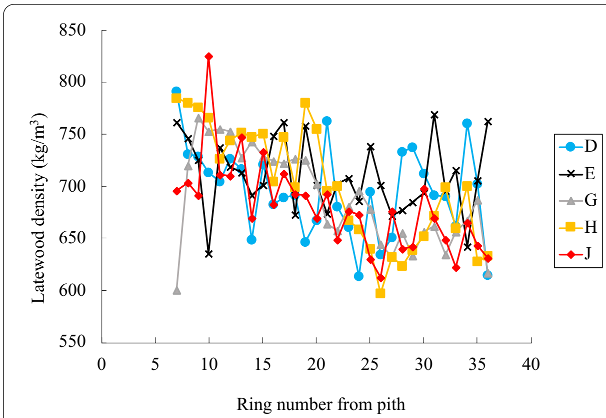

Fig. 7 Effects of initial tree density on latewood density. Each plot shows an average value in a ring number among sample trees of each tree density zoning symbol. Number of sample trees of each initial tree density zoning symbol range from five to nine. Initial tree density zoning symbols were the same, as shown in Table 1

Discussion

Effects of species-specific characteristic in annual ring formation on wood density

As previously described, low initial tree density increased diameter growth (Table 1). We reported the seasonal variation of tracheids formation and the amounts of plant hormones in cambial region tissues in a sugi cultivar grown in the same Nelder plot used in this study [21]. The amount of auxin (IAA) was positively correlated with the number of tracheids formed in early and mid-season, but not in late season, and had no relation to tracheid differentiation. H/D ratio was negatively correlated with IAA amounts in early and mid-season, but not in late season. Therefore, low initial tree density sugi trees had small H/D ratios, large amounts of IAA and then formed large number of tracheids. In addition, we previously reported the species-specific annual ring formation characteristics of three conifers (slash pine (*Pinus elliottii*), hinoki and sugi) grown in the same stand over 2 years [28], and the seasonal variations of the plant hormones in cambial-region tissues and their effects on annual ring formation in three conifers (slash pine, hinoki, and sugi) with inherently different wood densities [29]. Sugi (small

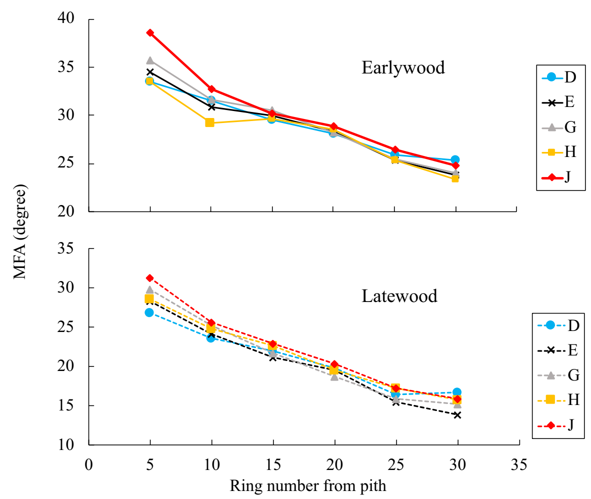

Fig. 8 Effects of initial tree density on MFA of earlywood and latewood in each ring Each plot shows an average value in a ring among five sample trees of each tree density zoning symbol. Initial tree density zoning symbols were the same, as shown in Table 1

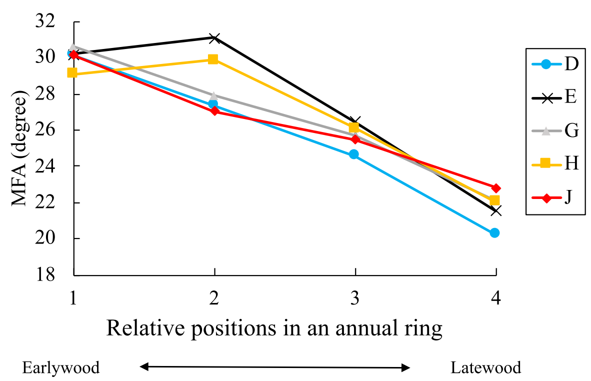

Fig. 9 Effects of initial tree density on the transition from earlywood to latewood in MFA in the ring number 22. Each plot was mean of five sample trees in each tree-density-zoning symbol. Initial tree density zoning symbols were the same, as shown in Table 1

Table 3 Ratio of tracheids with unusually large MFA in the ring number 22 among tree-density-zoning symbols

Symbol	Density (trees/ha)	Age	*n*	≥ 30° (%)	≥ 35°
D	4823	38	5	24 (8)[a]	10 (6)[a]
E	3349	38	5	38 (11)[a]	15 (7)[a]
G	1615	38	5	33 (14)[a]	12 (10)[a]
H	1122	38	5	33 (14)[a]	11 (4)[a]
J	541	38	5	31 (12)[a]	11 (5)[a]

The values represent averages in four trees; values in parentheses indicate standard deviations. Total number of measured tracheids were one hundred twenty in each tree (Fig. 9). Different characters show significant differences among density-zoning symbols ($p < 0.05$)

Symbol initial tree density zoning symbol, Density initial tree density, *n* number of sample trees

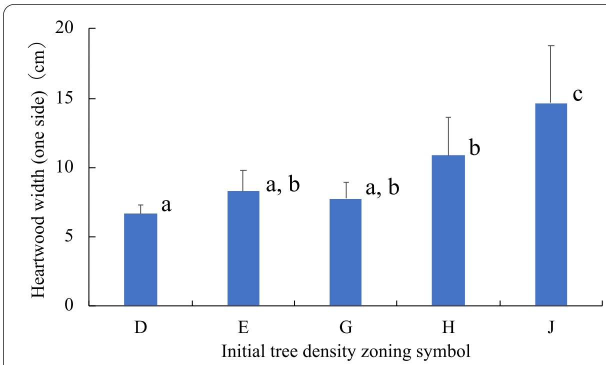

Fig. 10 Effects of initial tree density on heartwood width. Each bar and error bars show a mean and a standard deviation of eight or nine trees of each tree density zoning symbol. Different characters mean significant differences (Tukey HSD, $p < 0.05$). Initial tree density zoning symbols were the same, as shown in Table 1.

wood density) had significantly higher levels of IAA and formed more tracheids in the early season than in the late season, although slash pine (large wood density) had higher levels of IAA and formed significantly more tracheids in the late season than in the early season. Hinoki (intermediate wood density) had constant IAA levels and formed a constant number of tracheids throughout the season. There were significant positive correlations between the levels of IAA in cambial-region tissues and the number of tracheids formed during late season in the two conifer species. A close relationship was observed between the seasonal ratio of the IAA levels (late/early season) and wood density. From these studies, it was assumed that low initial tree density induced large diameter through active earlywood formation induced by large IAA amounts in cambial region tissues in early season as a species-specific characteristic of sugi. Therefore, it was assumed that effects of low initial tree density (J tree (541 trees/ha)) were recognized in LP, AWD and EWD because of the species-specific characteristic of sugi. Significant decrease in EWD of J trees (Fig. 6) was induced by typical earlywood tracheids with large radial diameter and thin cell wall.

Effects of height growth on wood properties in low initial tree density

As previously described, initial tree density had smaller effects on height growth than on diameter growth (Table 1); therefore, small H/D ratio of sugi trees in low initial tree density was induced by mainly variation of DBH in this study. On the other hand, we previously reported the wood properties of the sugi trees grown in a stand with large variation of tree height and small variation of DBH [19]. Therefore, H/D ratios of these sugi trees vary with mainly variation of tree height [19]. Taller trees had large stem stiffness because of smaller MFA and large wood density, while shorter tree had small stem stiffness because of larger MFA and small wood density [19]. We also examined the stem stiffness of sugi trees grown in different three stands with large variation of tree height and small variation of DBH [15]. Stem stiffness of sugi trees in a stand with larger tree height was larger than those of sugi trees in other stands with smaller tree height. We examined wood properties in the rings of hinoki trees grown in a stand, as well as growth traits in the years when the rings at breast height were formed by stem analysis [30]. H/D ratio had a close correlation with stem stiffness of hinoki trees. Tree height in the ring-formed year had a significant negative effect on MFA in juvenile wood, in transition wood between juvenile and mature wood, and in mature wood of hinoki trees. DBH in the ring formed year had a significant negative effect on density in mature wood of hinoki trees. From these studies, it was assumed that larger height growth might induce small MFA in sugi trees. According to our hypothesis on the effects of tree height on MFA, low initial tree density might have very small effects on MFA. Because there was no significant effect of initial tree density on tree height, and the average value of tree height of low initial tree density trees was larger than those of higher initial tree density trees.

Interaction between initial tree density (environmental factor) and sugi cultivar (genetic factor)

Based on the results on wood density and MFA of Tosaaka, a sugi cultivar grown in a Neldar plot, it was assumed that mechanical properties and dimensional stability of lumbers from low initial tree density (1000 trees/ha) would be quite similar to those from the regular initial tree density (3000 trees/ha). However, wood properties vary with the variation of both environmental factor and genetic factor. We have to take the possible interaction between initial tree density (environmental factor) and sugi cultivar (genetic factor) into consideration. H/D ratio had a close positive correlation with stem stiffness in Tosaaka grown in Nelder plots [15]. We also reported the relations between H/D ratio and stem stiffness in the other sugi cultivars [15]. Based on our study, Tosaaka was one of cultivars with a close positive correlation between H/D ratio and stem stiffness, although some cultivars had the significant but weak correlations. The variation of H/D ratio and stem stiffness were relatively constant in some sugi cultivars (Kumotoshi and Arakawa) in comparison with those of other cultivars grown in the same stand. Yabukuguri was a sugi cultivar that had a close negative correlation between H/D ratio and stem stiffness (the opposite trend to other cultivars). Therefore, Yabukuguri with smaller H/D ratio had larger stem stiffness.

The names of sugi cultivars used for reforestation in Japan are usually unknown. Therefore, unknown sugi cultivars are planted in a plantation. By taking the effects of genetic factor into consideration, the effects of low initial tree density on mechanical properties and dimensional stability of lumbers in sugi plantations in Japan might be smaller than the effects recognized in Tosaaka in this study.

Abbreviations

Tukey HSD: Tukey's honestly significant difference test; MFA: Microfibril angle; RW: Ring width; LP: Latewood percentage; AWD: Average wood density; EWD: Earlywood density; LWD: Latewood density; MOE: Modulus of elasticity; DBH: Diameter of the trees at breast height; H: Tree height; H/D ratio: Height-to-diameter ratio; HCB: Height at crown base; IAA: Auxin, indole acetic acid.

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Author contributions

YK designed this study and examined the wood properties of a sugi cultivar grown in a Nelder plot, and was a major contributor in writing the manuscript. TT contributes to discussion on the obtained results. HO contributes to experimental method and build the hypothesis in this study. RH and SI contributes to descriptions on techniques to reduce reforestation costs in forestry in introduction. All authors read and approved the final manuscript.

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Availability of data and materials

Not applicable.

Declarations

Competing interests

The authors declare that they have no competing interests.

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