

# Termite resistance and color of heartwood of hinoki (*Chamaecyparis obtusa*) trees in 5 half-sib families in a progeny test stand in Kyushu, Japan

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**Abstract** Hinoki (*Chamaecyparis obtusa*) wood has been used as a structural material in Japan because of its superior mechanical properties, its excellent durability and the beautiful color of its heartwood. Variations of termite resistance and compositions of extractives among hinoki trees have been reported. However, genetic variation of termite resistance and the effect of heartwood color on termite resistance remain unknown. In this study, we report the characteristics of termite resistance and color indexes ( $L^*$ ,  $a^*$  and  $b^*$ ) of heartwood of hinoki half-sib families in a progeny test stand. The survival days of termites and the mass loss of samples of hinoki heartwood differed significantly among hinoki families. Families with red-color heartwood had larger termite resistance than families with yellow-color heartwood. The termite resistances of individual samples from two families with yellow-color heartwood were as small as that of *Pinus densiflora*. Larger  $a^*$  and smaller  $b^*$  induced larger termite resistance of heartwood. The effect of DBH (diameter at breast height)

of hinoki trees on termite resistance of hinoki families was small.

**Keywords** Hinoki plus tree · Termite resistance · Color of heartwood · Growth rate

## Introduction

Hinoki (*Chamaecyparis obtusa*) is one of the important plantation species in Japan. Hinoki wood has been used as a structural material in Japan because of its superior mechanical properties, its excellent durability and the beautiful color of its heartwood. The MAFF (Ministry of Agriculture, Forestry and Fisheries) of Japan aims to achieve  $\geq 50\%$  wood self-sufficiency by developing a reliable domestic wood supply/use system [1]. Therefore, improvements are sought in the reliability and wood qualities of hinoki wood. To obtain information about the genetic variation of the wood properties of hinoki, we examined the microfibril angle (MFA) and wood densities of hinoki families grown in a progeny test stand [2]. Hinoki heartwood is believed to have excellent durability; however, a house builder in Miyazaki prefecture shared with us his experience that hinoki heartwood is not always consistent on this excellent durability (private communication). Therefore, it is also important to obtain information about genetic variations in the durability of hinoki heartwood. Although preservatives can raise the durability of wood, the current desire is for superior-durability lumber that does not add an extra impact to the environment. In this study, we focused on the variation of the natural durability of hinoki heartwood.

Many studies have examined the chemical components that provide fungal and termite resistance to hinoki

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heartwood. The antifungal components in hinoki heartwood extractives have been identified as sesquiterpene alcohols, mainly cadinols [3]. The termite resistance of hinoki heartwood is attributable to both stable components with low activity and unstable components with high activity [4]. The stable components with small termite resistance include non-volatile cadinols [5] and  $\alpha$ -terpineol [6]. A major unstable component with large termite resistance was the volatile component  $\alpha$ -terpineol acetate [6].

Variations of termite resistance and amounts of extractives among hinoki trees have been reported. Decay and termite resistance, and amounts of extractives of hinoki heartwood from different area in Japan showed variations with no apparent effect of density or ring width on decay and termite resistance [7]. Hinoki wood from a natural forest had higher termiticidal activity than that from a plantation forest, and the higher termiticidal activity was explained by the larger volatile extractive contents and smaller average ring width [8]. Volatile monoterpenes in needle samples of 50 hinoki trees from five different plantations had considerable compositional variation, especially in sabinene composition, and there was no relation between tree size and volatile monoterpene composition [9]. From these studies, it was assumed that the termite resistance of hinoki varied among trees, and that it was so difficult to predict the variations of termite resistance of hinoki heartwood, genetic backgrounds of which were unknown. In western red cedar (*Thuja plicata*) and Alaska cedar (*Chamaecyparis nootkatensis*), methanol-soluble heartwood extractives are an important factor for the natural durability of the heartwood and there was considerable variability of natural durability among and even within trees [10].

In hinoki clones selected as cold and wind-resistant trees, sabinene composition in needles is constitutively steady in most clones [11]. In the heartwood of sugi (*Cryptomeria japonica*), contents and composition of extractives differ among cultivars [12–14]. In the heartwood of sugi plus tree (selected as trees with various superior traits, e.g., fast growth and trunk straightness) families, the contents and composition of extractives were shown to differ among families, and the effects of plantation area on the contents and composition of extractives were observed [15–19]. The termite resistance of sugi heartwood has been shown to differ among cultivars [20]. From these studies, also in hinoki, the genetic effect on termite resistance of heartwood may be significant. Hinoki plus trees could contribute to a reliable domestic wood supply. However, the termite resistance of heartwood of hinoki plus trees remains unknown.

The color of hinoki heartwood is a very important factor for the end user purchasing hinoki wood in Japan. The local cultivar of hinoki in northeastern Fukuoka prefecture has

been established as a brand (Keichiku Hinoki), and a questionnaire survey established that the branded hinoki was characterized by the red-tinged color of its heartwood [21]. Therefore, heartwood color-based selection of hinoki families is assumed to be effective at enticing end users to purchase hinoki wood. In hinoki trees from eight regions in Japan and Tono hinoki (one of brand hinoki trees), it was reported that four norlignans were colored extractives; hinokiresinol, 3-methoxyhinokiresinol and cryptoresinol were pale pink and isocryptoresinol was pale yellow, and the content of hinokiresinol was reflected in the pink color of the heartwood [22]. From these results, it was assumed that heartwood color of hinoki trees came from heartwood extractives and varied from red to yellow. As previously described, many researches reported that heartwood extractives contributed durability of heartwood. Therefore, we guess that there was some relationship between heartwood color and the durability of heartwood; although extractives contributed to durability might have no color. Heartwood color-based selection of hinoki families may affect the termite resistance of hinoki heartwood. However, the relationship between color and the termite resistance of hinoki heartwood remains unknown.

The objective of this study was to examine: (1) the differences between hinoki families in color indexes of heartwood, (2) the differences between hinoki families in termite resistance, (3) the relation between color of heartwood and termite resistance of hinoki families and (4) the relation between DBH (diameter at breast height) and termite resistance in hinoki families. Differences among hinoki families in termite resistance are discussed based on data obtained at 1.2 m above ground. Therefore, the difference between hinoki families at other heights in the trunks remains unclear.

## Materials and methods

### Sample trees

Hinoki half-sib families were planted in a progeny test stand in Oita prefecture, Japan in 1974 by Forest Tree Breeding Center (FTBC), Forestry and Forest Products Research Institute (FFPRI). Hinoki trees (31 families  $\times$  2 replicates  $\times$  49 trees) were planted at 1.8-m spacing, and the stands were not managed by silvicultural practice. Sample trees (31 families  $\times$  2 replicates  $\times$  3 trees) were harvested after 29 years of growth. Eighty hinoki trees (15 half-sib families  $\times$  5 or 6 trees) were selected as sample trees for this study from the harvested trees (Table 1). These 15 half-sib families were the same families discussed in a previous study [2]. The local families in this progeny test stand were the families that had been selected

**Table 1** Sample trees from 15 half-sib families

Family number	Family name	Prefecture	n	DBH (cm)	E <sub>d</sub> (GPa)	Color index of heartwood				Termite test
						L*	a*	b*	Hue	
1	Oita	Oita	5	17.7 (14.6)	9.9 (9.4)	63.1 (5.9)	12.6 (18.6)	30.2 (7.1)	1.18 (6.7)	
2	Kanzaki 5	Saga	5	20.0 (8.0)	9.9 (5.6)	65.9 (1.6)	10.8 (32.9)	28.4 (11.7)	1.21 (10.1)	
3	Takedasho 2	Oita	5	17.2 (19.5)	10.1 (10.4)	66.5 (1.5)	10.7 (30.5)	27.0 (8.8)	1.19 (8.6)	
4	Saeki 5	Oita	5	19.5 (18.4)	9.0 (8.9)	66.1 (1.4)	9.0 (49.2)	27.7 (15.7)	1.26 (12.5)	
5	Onga 1	Fukuoka	5	18.5 (15.9)	10.5 (12.6)	66.4 (1.8)	9.0 (36.6)	29.7 (9.3)	1.28 (8.3)	
6	Kikuchi 1	Kumamoto	6	20.0 (15.3)	9.1 (8.8)	77.4 (2.7)	8.7 (19.1)	25.0 (6.1)	1.24 (4.3)	○
7	Nakatsu 1	Oita	6	17.3 (11.2)	10.1 (9.6)	78.2 (4.0)	8.6 (14.8)	24.9 (5.5)	1.24 (4.6)	○
8	Ukiha 13	Fukuoka	6	18.7 (7.4)	9.5 (14.7)	78.3 (2.4)	8.4 (25.3)	24.9 (4.1)	1.24 (6.9)	○
9	Aira 2	Kagoshima	5	17.5 (19.8)	8.2 (5.1)	67.3 (2.1)	8.3 (29.8)	28.8 (8.7)	1.29 (7.4)	
10	Nankourai 2	Nagasaki	5	18.4 (8.7)	9.2 (15.7)	66.3 (2.5)	7.9 (51.2)	28.0 (7.6)	1.30 (9.4)	
11	Yamada 2	Fukuoka	5	19.8 (11.0)	8.7 (15.6)	65.8 (3.5)	7.3 (47.5)	28.2 (11.6)	1.32 (8.6)	
12	Isa 1	Kagoshima	6	19.0 (20.1)	9.4 (7.5)	78.2 (2.6)	6.1 (25.9)	26.7 (4.9)	1.35 (3.6)	○
13	Kitamorokata 2	Miyazaki	6	19.2 (18.7)	8.8 (21.9)	77.9 (1.9)	5.9 (29.8)	25.8 (3.7)	1.35 (4.6)	○
14	Kusu 6	Oita	5	18.2 (11.1)	10.8 (14.0)	66.5 (1.9)	5.8 (37.7)	28.5 (11.5)	1.37 (6.6)	
15	Nakatsu 3	Oita	5	19.7 (9.5)	10.2 (7.1)	66.7 (2.6)	5.6 (49.3)	29.5 (3.3)	1.38 (6.6)	

Oita, Nakatsu 1 and Nakatsu 3 are local families in Oita prefecture. Other families are plus tree families. The values of DBH, E<sub>d</sub> and Color index represent the averages of sample trees and the values of parentheses represent the coefficients of variation (%). Prefecture is the prefecture where the trees are selected

n number of samples, DBH diameter at breast height, E<sub>d</sub> dynamic modulus of elasticity, L\* brightness, a\* redness, b\* yellowness, Hue tan<sup>-1</sup>b\*/a\* (JIS Z8729)

and bred as families with superior traits before the project of plus tree selection started and plus tree seedlings were able to be utilized. Wood materials for evaluation of heartwood color and termite test were obtained from a 20-cm length of trunk at 1.2 m above the ground.

Measurements of color of heartwood

An air-dried edge grain board with the pith in the center was cut from a 20-cm length of trunk at 1.2 m above ground. After the edge grain surface was made smooth with a planer, the color of the heartwood in the edge grain surface was evaluated by colorimeter (Tokyo Denshoku, TC-PIII) and with an L\*a\*b\* color system. Color index values indicating the brightness (L\*), redness (a\*), and yellowness (b\*) were obtained from the L\*a\*b\* system based on JIS Z8729. The spot size of color index measurement was 20 mm. For color index measurement, 3 different points with no defects on the edge grain surface of heartwood were randomly selected. A color index of each sample was obtained from an average of 3 replicates of evaluation.

No-choice termite tests

For the termite test, five families were selected from 15 hinoki half-sib families (Table 1). For selection of families

for the termite test, a\* and hue (tan<sup>-1</sup>b\*/a\*) were used (JIS Z8729). The families with larger coefficients of variation (%) of a\* and hue (Table 1) were avoided for the termite test, because the average value of color index with a larger coefficient of variation could not show the characteristic of each family. In addition, to examine families from wide area, families from different prefectures were selected. Therefore, Kikuchi 1 (Kumamoto prefecture), Nakatsu 1 (Oita prefecture), Ukiha 13 (Fukuoka prefecture), Isa 1 (Kagoshima prefecture) and Kitamorokata 2 (Miyazaki prefecture) were used for termite test. Although Oita (Oita prefecture) had larger a\* and smaller hue, the coefficients of variation (%) of a\* and hue were larger than those of Nakatsu 1 (Oita prefecture). Therefore, Nakatsu 1 (Oita prefecture) was used for termite test instead of Oita (Oita prefecture).

Two test blocks (2.0(L) × 2.0(R) × 2.0(T) (cm)) were cut from the middle or outer heartwood of each sample tree (Table 1), except near the pith. The total samples numbered 60 blocks (5 families × 6 trees × 2 replicates). Test blocks were air-dried at 20 °C, 60 % RH, and the air-dried weight was measured before and after the termite test. In Japan, *Coptotermes formosanus*, *Reticulitermes speratus* and recently *Incisitermes minor* mainly attacked wooden constructions. For termite test to evaluate the performance of preservatives, *C. formosanus*

was decided as the test insect in JIS K 1571. However, the habitation region of *C. formosanus* was limited to the western part of Japan. In this study, *R. speratus* was used as a test insect, because *R. speratus* inhabited all over Japan and the aim of this study was the comparison of the natural termite resistance among hinoki families. A test block was placed in cylinder (10 cm in diameter and 6 cm in height) with sea sand at the bottom, together with 95 workers and 5 soldiers of *R. speratus*. The test cylinders were placed in an incubator (28 °C, 80–90 % RH). The test period was 54 days. Eight ml distilled water was initially added to the sea sand in the test cylinder, and 1–2 ml distilled water was added every day during the test period. Termite mortality (%) during the test period and the mass loss of the samples (%) after the test period were measured. We used heartwood of *Pinus densiflora* as control.

### Statistical analysis

The number of families and replications used for this study was small.

Therefore, data from two replicates for each family were combined, and analyzed by one-way analysis of variance or non-parametric test (Kruskal–Wallis test) and multiple comparisons (Tukey HSD test and Bonferroni test or Games Howell test) (statistical analysis software, SPSS ver.16 with Regression and Advanced Models). When equality of variances between data of families was not recognized, Kruskal–Wallis test and Games Howell test were used.

## Results and discussion

### Genetic variation of color of heartwood

As previously described, the color of hinoki heartwood may be a more attractive factor for end users than its mechanical properties and durability. Therefore, color index values (brightness ( $L^*$ ), redness ( $a^*$ ) and yellowness ( $b^*$ )) of 15 hinoki half-sib families were examined. As shown in Table 1, the  $a^*$  values of the families ranged from 5.6 to 12.6, and there was significant difference of  $a^*$  among families ( $p < 0.05$ ), although there was no significant difference of  $L^*$  and  $b^*$  among families. The coefficients of variation (%) of  $a^*$  of each family were larger than those of other color indexes of each family. These results are similar to those of previous reports [23, 24]. As shown in Fig. 1, we also examined the variation of color indexes at 3 different points in individual hinoki trees. It was recognized that coefficient of variation of  $a^*$  in

individual trees was larger and had wider range than those of  $L^*$  and  $b^*$ .

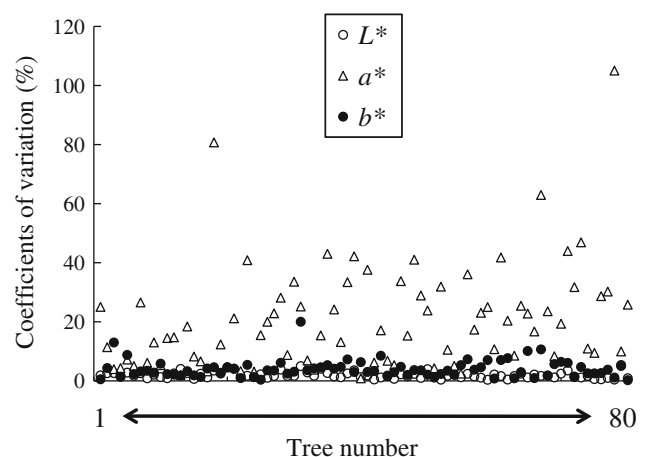
From these results, selection of families by  $a^*$  was assumed to be an efficient method for enhancing a reliable hinoki wood supply with attractive heartwood color. However, the large coefficient of variation (%) of  $a^*$  within not only each family but also individual trees should be considered.

From measurement of color indexes, it was recognized that Kikuchi 1, Nakatsu 1 and Ukiha 13 were the families with red-color heartwood, and Isa 1 and Kitamorokata 2 were the families with yellow-color heartwood.

### Genetic variation of termite resistance of heartwood

Average number of survival days of termites of each family ranged from 21 to 34 days (Fig. 2). Survival days of termites were smallest in Ukiha 13 and largest in Kitamorokata 2. The survival days of termites of hinoki heartwood significantly differed among families (one-way ANOVA,  $p < 0.05$  and Kruskal–Wallis test,  $p < 0.01$ ). By multiple comparisons, it was recognized that Ukiha 13 induced statistically fewer survival days of termites than Kitamorokata 2 ( $p < 0.05$ ). In the heartwood of *P. densiflora* (used as control), mortality did not reach 100 % during the test period (54 days). Mortality of most hinoki heartwood except for 2 wood blocks (Kitamorokata 2 and Isa 1) reached 100 % during the test period.

Average mass loss of samples of each family ranged from 1.05 to 2.47 % after a 54-day test period (Fig. 2). The average mass loss of samples was smallest in Kikuchi 1 and largest in Isa 1. The mass loss of samples of hinoki heartwood significantly differed among families (Kruskal–Wallis test,  $p < 0.01$ ). Equality of variance of mass

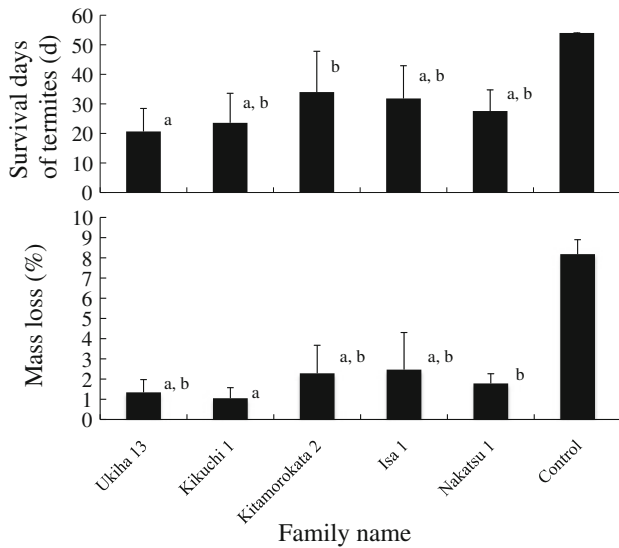


**Fig. 1** Coefficients of variation of color indexes in individual hinoki trees. Coefficients of variation were calculated from color indexes at 3 different points in individual hinoki trees. Tree number (1–80) was given to 80 hinoki trees as in Table 1

loss between families did not recognized. Therefore, significant difference among families was analyzed by non-parametric test (Kruskal–Wallis test). By multiple comparisons (Games Howell test), it was recognized that Kikuchi 1 had a statistically smaller mass loss of sample than Nakatsu 1 ( $p < 0.05$ ). Although Isa 1 and Kitamorokata 2 had larger

average mass losses of samples, their mass losses were not significantly different from that of Kikuchi 1 because of the large variations among their samples. In the heartwood of *P. densiflora* (used as control), the average mass loss was 8.2 %. Although the mass losses of most hinoki heartwood were obviously smaller than that of the heartwood of *P. densiflora*, the mass loss of 2 samples of Kitamorokata 2 and Isa 1 was 5.8 and 7.0 %, respectively. Termite attack was limited to the surface of hinoki heartwood samples while termite attack on the heartwood of *P. densiflora* penetrated the sample from one surface to the surface of the opposite side.

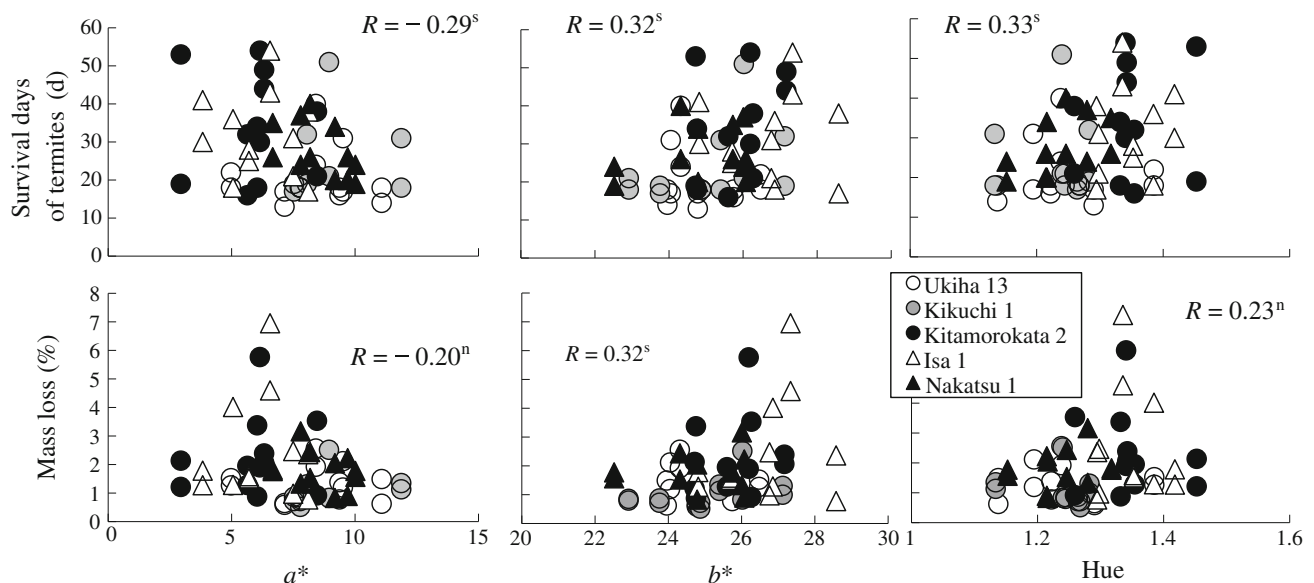
It has been believed throughout Japan that hinoki heartwood has large termite resistance. However, it was shown here that termite resistance differed among families and that there are families with less termite resistance. Especially, the termite resistances of individual samples of Kitamorokata 2 and Isa 1 were as low as that of *P. densiflora*. It was suggested that for a reliable hinoki wood supply, more information about the termite resistance of hinoki families should be obtained.



**Fig. 2** Termite resistance of hinoki trees from 5 half-sib families. Survival days of termites are incubation time when mortality reached 100 %. Survival days of termites of wood blocks that termites survive after 54-day test period are shown as 54 days. Mass loss (%) was mass loss of wood block after test periods. Survival days of termites and mass loss (%) were average of 6 trees (2 replicates per tree) of each family. Means with different letters are significantly different ( $p < 0.05$ ). Error bar standard deviation. Control *Pinus densiflora*

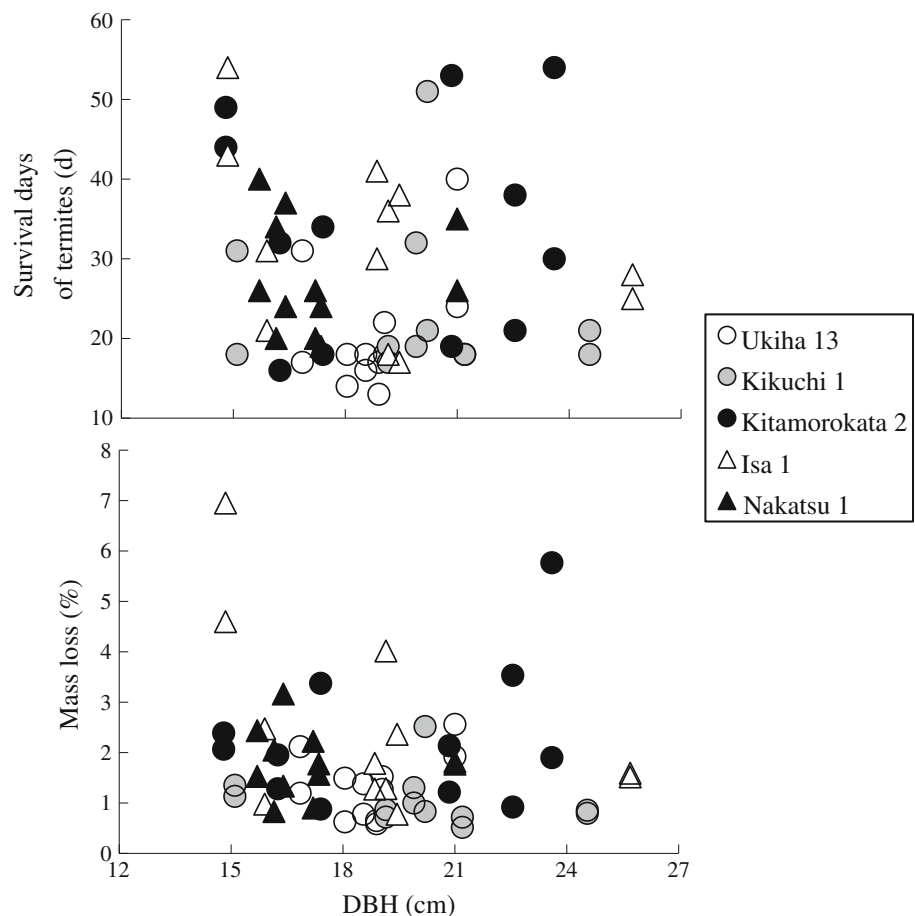
Effects of color of heartwood on termite resistance

As previously described, Kikuchi 1, Nakatsu 1 and Ukiha 13 were the families with red-color heartwood, and Isa 1 and Kitamorokata 2 were the families with yellow-color heartwood. From the results of the termite test, the families with red-color heartwood (Kikuchi 1 and Ukiha 13) had termite resistance superior to that of the families with yellow-color heartwood (Isa 1 and Kitamorokata 2).



**Fig. 3** Effect of color indexes on termite resistance of hinoki heartwood. Hue  $\tan^{-1}b^*/a^*$  (JIS Z8729), n no significant correlation, s significant correlation ( $p < 0.05$ )

**Fig. 4** Effect of DBH on termite resistance of hinoki trees from 5 half-sib families



Therefore, the difference of color indexes of heartwood may be an indicator of termite resistance. As shown in Fig. 3, there was significant correlation between color indexes ( $a^*$  and  $b^*$ ) of heartwood and survival days of termites during the test period in all samples ( $p < 0.05$ ). Larger  $a^*$  and smaller  $b^*$  induced larger termiticidal activity of heartwood of hinoki. Therefore, smaller hue induced larger termiticidal activity of heartwood (Fig. 3). There was also significant correlation between  $b^*$  of heartwood and mass loss of hinoki heartwood (Fig. 3,  $p < 0.05$ ), although there was no significant correlation between  $a^*$  of heartwood and mass loss of hinoki heartwood ( $p = 0.13$ ). Smaller  $b^*$  of heartwood induced smaller mass loss. From these results, it was recognized that larger  $a^*$  and smaller  $b^*$  induced larger termite resistance of heartwood of 5 hinoki families;  $b^*$  was an especially important color index for termite resistance. Therefore, selection of hinoki families with red-color heartwood (smaller hue ( $\tan^{-1}b^*/a^*$ )) is assumed to enhance a reliable hinoki wood supply with not only attractive heartwood color, but also superior termite resistance. As described in the previous section, there was no significant difference of  $b^*$  among hinoki families. Variation of  $b^*$  assumed to

contribute to variation of termite resistance among trees in each hinoki families.

In this study, we selected families with smaller  $a^*$  variation and we identified a significant difference in termite resistance among families. If a termite test had been done on other families with larger  $a^*$  variation, there may not have been a significant difference among them. In the selection of families by heartwood color indexes, smaller variation of color indexes within the family is assumed to be important. As shown in Fig. 1, there were hinoki trees with larger variations of  $a^*$  and  $b^*$ . As described above, larger  $a^*$  and smaller  $b^*$  induced larger termite resistance of heartwood of 5 hinoki families. Therefore, in these trees, it was assumed that termite resistance varied in individual trees by the variations of  $a^*$  and  $b^*$ . For reliable supply of hinoki wood with superior heartwood durability, control of intra- and inter-tree variations of  $a^*$  and  $b^*$  assumed to be important.

To enhance the use of hinoki wood, in hinoki wood supply, reliability of hinoki wood performance should be guaranteed. As previously described in introduction, more information about termite resistance of hinoki families should be obtained. However, termite test is not a non-

destructive method. Therefore an easy, rapid and especially non-destructive method for estimation of termite resistance should be developed. In the decay resistance of Togolose teak (*Tectona grandis*), for example, the relationship of  $L^*$  with mass loss was shown to be highly significant; thus, the use of colorimetry as a rapid and cheap tool to estimate short-term natural durability could be considered [25]. In hinoki families, the relationships between color indexes and termite resistance were significant; however, the correlation coefficients were small (Fig. 3). Further study on more accurate non-destructive estimation of termite resistance of hinoki families (e.g., estimation by near-infrared spectrometry [26]) is needed.

#### Effects of DBH on termite resistance

Fast growth rate is the important characteristic in trees planted for wood production. However, if fast growth reduces the durability of hinoki heartwood, higher growth rates would not be worthwhile. As described in introduction, hinoki wood from a natural forest had higher termiticidal activity than those from a plantation forest, and the higher termiticidal activity was explained by the proportionally larger content of volatile extractives and smaller average ring width [8]. Genetic factors or growth rate or both are assumed to contribute to termiticidal potential. To clarify the effect of the growth rate, the relationship between DBH and termite resistance was examined in each hinoki family. As shown in Fig. 4, there was no significant effect of DBH on survival days of termites and mass loss during the test period. The number of sample trees in each family was small (6 trees in each family). Therefore, all sample trees of 5 families were combined and separated into 3 groups (large DBH, middle DBH and small DBH), and then the differences of termite resistance between groups were examined. The average survival days of the large-DBH group, middle-DBH group and small-DBH group were 29.8, 22.3 and 30.6 days, respectively. By multiple comparisons (Tukey HSD test), it was shown that the middle-DBH group had significantly fewer survival days of termites during test period than the small-DBH group ( $p < 0.05$ ). From these results, it was assumed that the effect of DBH on termite resistance was small and that smaller DBH did not increase the termite resistance of hinoki trees from 5 families. This result does not agree with that of Ohtani et al. [8]; further study on this issue is needed.

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