

Antifungal Effects of Staining Process on Wood: Hardness, Gloss, and Color Change

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This study determined the effects of wood staining on wood-destroying fungi. To achieve this goal, different types of wood samples were used, including Scotch pine (*Pinus sylvestris* L.), Eastern beech (*Fagus orientalis* Lipsky), sessile oak (*Quercus petraea* Liebl.), and mahogany (*Entandrophragma cylindricum*). Aniline (C₆H₂NH₂), chemical (tannin (C₁₄H₁₀O₉) + potassium dichromate (K₂Cr₂O₇)), and Van Dyke brown stains (Fe₂O₃MnO₂ + K₂Cr₂O₇ + H₂O) were applied to the samples, because a walnut color (brown) is preferred by customers. The stained samples were exposed to *Fomitopsis palustris* and *Coriolus versicolor*, and mycelium growing on wood was observed for 3 months. Hardness, gloss, and total color change tests were applied to the samples to determine the antifungal effects. The results showed that staining increased the total color change values of the wood, while decreasing in the gloss and hardness values. The chemical stain showed antifungal effects against both fungi.

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INTRODUCTION

Wood is a strong and versatile material that can be used in many different ways. Due to the increase in human population and the growing use of wood in construction, the demand for wood has increased. Global forest assets are being threatened, and there is a need for more efficient use of this limited resource (Karal 2017). Wood in its natural state can be destroyed by fungi and insects, and this causes great financial losses every year (Broda 2020). It is estimated that millions of dollars of damage are caused every year by rot fungi, especially brown and white rots (Schmidt 2006; Broda 2020).

The damage caused by fungi is often irreversible. Most fungi first appear in wood with color changes and a dull appearance. This spoils the aesthetic appearance of the material. The discolored part often cannot be repaired and must be replaced completely. Some fungi cause severe tissue damage in later stages by destroying the chemical structure of the wood. Among the most important fungi that destroy wood at the point of use are brown rot fungi (destruction rot fungi), which belong to the Basidiomycetes class. This fungus rapidly reduces the strength properties of wood and causes it to turn dark brown (Griffin 1977; Goodell *et al.* 2008). Wood color changes primarily due to the degrading of the cellulose content of wood by these fungi; naturally dark-colored lignin, extractive substances, and tannins are left behind. Degradation due to these fungi results in transverse cracks, dimensional changes, and cell wall collapse in the final stages (Schmidt 2006).

White rot (corrosion rot) fungi, another species belonging to the Basidiomycetes class, destroy the lignin content of the wood and leave a white fibrous cellulose structure, unlike brown rot fungi (Gilbertson 1980; Blanchette 1991). A wood material that is suffering from white rot becomes cottony, soft, and lighter in color. Hardwood tree species are more susceptible to these fungi than softwood tree species. Wood that is affected by white rot fungus exhibits abnormal dimensions shrinkage, transverse cracks, and collapses, much like the wood that is affected by brown rot fungus (Boyle 1995; Schmidt 2006; Broda 2020).

There are many methods and chemicals for protecting wood, extending its service life, and preserving it from these fungi (Reinprecht 2016). The partial charring of wood and the use of animal, vegetable, and mineral oils were the earliest examples of wood preservation (Richardson 1993). Extracts and tannins obtained from the root, stem, bark, leaves, and fruits of the plants are used as natural preservatives (Broda 2020). The preservative chemicals industry, which has made remarkable progress, has introduced many alternative chemical products. However, a number of these products have been banned due to their adverse effects on the environment and human health (Humar *et al.* 2005). When used indoors, these chemicals threaten the health of humans; they were developed primarily for use outdoors (Taşçıoğlu *et al.* 2013). A substance that is used in wood preservation must exhibit abiotic properties, particularly against wood-destroying fungi and insects (Taşçıoğlu *et al.* 2013; Broda 2020). While impregnation materials and pesticides are examined for this purpose, environmentally friendly alternatives are still being explored.

To obtain colors other than the natural color of wood, staining is necessary when manufacturing wooden furniture and decoration elements. Color harmony and product diversity are taken into account during this process (Siva 2007; Tolvaj *et al.* 2019). These stains, which are typically used indoors, are not adequately tested for their ability to prevent fungus and insects. Although there are studies to eliminate the effects caused by fungi, there is a need for studies investigating the effects of these stains against fungi (Gorbushina *et al.* 1993; Okino *et al.* 2015).

The aims of the study were to examine the antifungal properties of some stains and to evaluate their suitability for protecting wood materials. Four different types of wood materials were stained using different stains. They were exposed to brown rot (*Fomitopsis palustris*) and white rot (*Coriulus versicolor*) fungi, and the samples were tested for their hardness, gloss, and color change.

EXPERIMENTAL

Preparation of the Wooden Materials

Scotch pine (*Pinus sylvestris* L.), Eastern beech (*Fagus orientalis* Lipsky), sessile oak (*Quercus petraea* Liebl.), and mahogany (*Entandrophragma cylindricum*) were evaluated. A random selection of timbers was made from timber suppliers in the northwest Turkish province of Düzce. These woods were selected because they are commonly used in furniture and decoration in Türkiye, and they have different anatomical features. The specimens were inspected to ensure that they did not have rot, knots, cracks, or density differences. Samples were cut in draft dimensions of 400 × 22 × 12 mm from sapwood parts with annual rings parallel to the surface. To test the effect of each factor (2 fungi, 4 woods, 4 stains), 6 samples were prepared ($2 \times 4 \times 4 \times 6 = 192$ in total). The samples were

sanded and calibrated using 80 and 100 grit sandpapers. The calibrated samples were cut to their final dimensions of $20 \times 20 \times 10$ mm. The samples were kept at 20°C and 65% relative humidity until they reached a constant weight (Fig. 1). Density values at 12% moisture content were 0.49 g/cm^3 for Scotch pine, 0.53 g/cm^3 for beech, 0.59 g/cm^3 for oak and 0.63 g/cm^3 for mahogany. A control group and a treatment group were formed after the conditioning process.

Staining Process

Walnut brown was preferred as the color, and aniline ($\text{C}_6\text{H}_2\text{NH}_2$), chemical (tannin ($\text{C}_{14}\text{H}_{10}\text{O}_9$) + potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$)), and Van Dyke brown ($\text{Fe}_2\text{O}_3\text{MnO}_2 + \text{K}_2\text{Cr}_2\text{O}_7 + \text{H}_2\text{O}$) were used as stains. The pH values of the prepared stains were measured using a pH-meter (EcoScan pH5, Eutech Instruments PTE LTD, Singapore), using the mean value obtained from three measurements. Table 1 shows the mixing ratios and pH values of the stains.



Fig. 1. Preparation and conditioning of samples

Table 1. Mixing Ratios and pH Values of the Stains

Stain Type	Mixing Ratio	pH
Aniline	$\text{C}_6\text{H}_2\text{NH}_2$ - 3%	5.80
Chemical	$\text{C}_{14}\text{H}_{10}\text{O}_9$ - 5% (First Step)	3.82
	$\text{K}_2\text{Cr}_2\text{O}_7$ - 5% (Second Step)	4.45
Van Dyke Brown	6 Unit - Fe_2O_3 , MnO_2 -10%	6.60
	3 Unit - $\text{K}_2\text{Cr}_2\text{O}_7$ - 5%	4.45
	1 Unit - H_2O - Distilled	7.18

To increase the stain penetration depth, a 2-min dipping method was utilized (Fig. 2). The chemical stain application of Eastern beech, sessile oak, and mahogany woods was conducted with only 5% solution of potassium dichromate due to their tannin content. Because Scotch pine does not contain tannin, chemical stain was applied in two stages. The samples were treated with 5% tannin solution and kept at room temperature (20°C) for 24 h; then, surfaces were stained with 5% potassium dichromate solution.

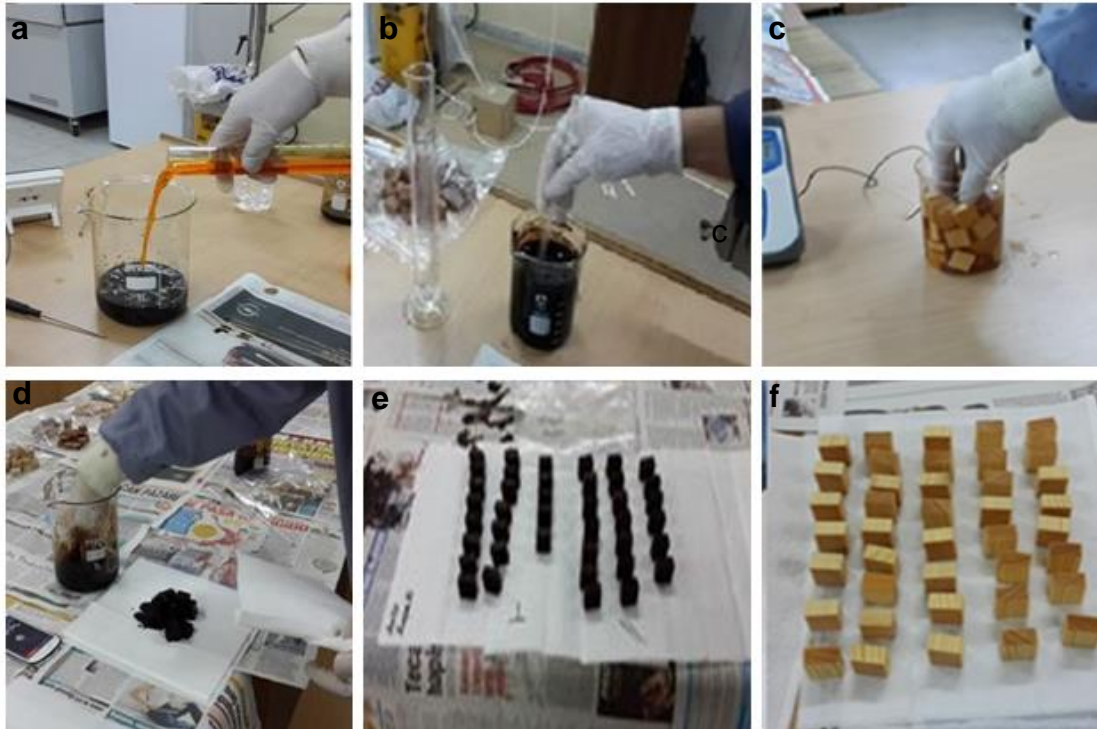


Fig. 2. The processes of stain preparation (a), mixing (b), application (c, d), and drying (e, f)

A sterilized cloth was used to remove excess stain from the surfaces after the dipping process. Following staining, all samples were stored in a climate cabinet at 20 ± 2 °C and $65 \pm 3\%$ relative humidity until they reached a constant weight.

Preparation of Growth Medium

An infestation of brown rot (*Fomitopsis palustris*) and white rot (*Coriolus versicolor*) fungi was conducted on stained and unstained (control) samples. Protective measures such as gloves and masks were taken throughout the entire process to prevent cross-contamination of samples. In addition, the work bench and equipment were washed with 70% ethanol before the process. Both fungi were cultured on 3.7% malt extract agar. On a magnetic stirrer-heater device, 37 g of malt extract agar were homogeneously mixed with 963 g of water to produce the growth medium. An autoclave was used to sterilize the mixtures at 121 °C for 20 min under 1.1 atm pressure. A nutrient solution of approximately 14 mL was placed on each Petri dish.

Exposure of Samples to Fungal Decay

Test samples were exposed to fungal decay according to TS 5563 EN 113 (1996). The micelles cut in 1 mm² pieces were inoculated into nutrient medium in Petri dishes in the biohazard safety cabinet. Using a culture chamber at 28 °C and 75 to 80% relative humidity, micelles were kept on the nutrient medium until they completely covered it. After 48 h at 60 °C, the weights of samples to be placed in the culture medium were determined. Then the samples were sterilized for 20 min in an autoclave at 110 °C under 1.1 ATM pressure. All samples were transferred to Petri dishes in a biosafety cabinet (Fig. 3). The growth of mycelium on the wood was observed for 3 months in a culture room with a temperature 28°C and relative humidity 75 to 80% (Fig. 3).

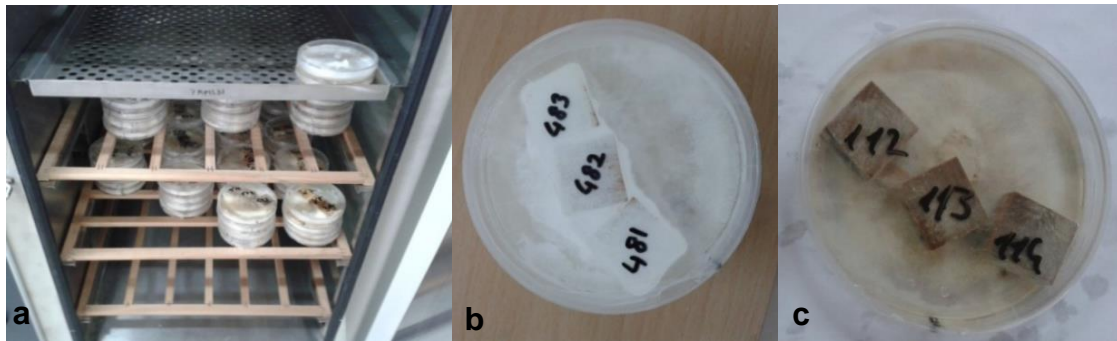


Fig. 3. Exposure of samples to fungal decay in a) biosafety cabinet and b) white rot and c) brown rot infected samples

Execution of Tests

The changes in the hardness, color, and gloss tests were determined. The average of three different measurements from the same sample was recorded as a single value for each test. Total color change and gloss change values were determined using the BYK - Gardner Spektrο-Guide 45/0 device (Spectro-guide sphere gloss meter, model CD-6834. BYK-Gardner GmbH, Geretsried, Germany). The color measurements were conducted according to ASTM D 2244 (2015) standard using the CIEL*a*b* color scheme. The CIEL*a*b* color system scheme is shown in Fig. 4.

In the scheme, L^* is on the black-white axis ($L^*=0$ is black, $L^*=100$ is white), a^* is on the red-green axis (positive values are reddish and negative values are greenish), and b^* is on the yellow-blue axis (positive values are yellowish and negative values are blueish). The total color change values, ΔE^* , were calculated using the Eq. 2,

$$\Delta E^* = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (2)$$

where ΔE^* is the total color change after fungal decay. The other “ Δ ” values also represent the difference between the values before and after the fungal decay, in the same way.

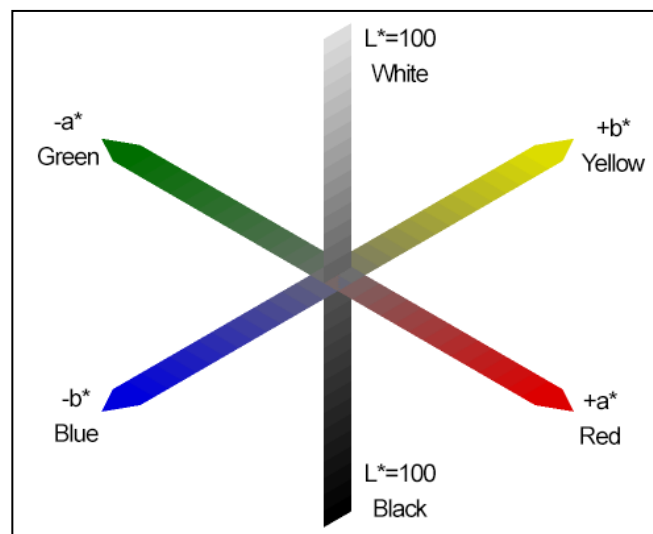


Fig. 4. CIEL*a*b* color scheme

The gloss measurements of the samples were made with the BYK - Gardner Spektro-Guide 45/0 device using 60-degree angle according to ASTM D523 (2014). The gloss change values (Δ_G) were calculated using Eq. 3,

$$\Delta_G = G_{AF} - G_{BF} \quad (3)$$

where G_{AF} is the gloss value after fungal decay, and G_{BF} is the gloss value before fungal decay.

The hardness values of the samples were determined with the Shoremeter-D hardness durometer in accordance with ASTM D 2240 (2006) before and after deterioration. The hardness change values, Δ_H , were calculated using the Eq. 4,

$$\Delta_H = H_{AF} - H_{BF} \quad (4)$$

where H_{AF} is the hardness value after fungal decay, and H_{BF} is the hardness value before fungal decay.

Statistical Analysis

The MSTAT-C 2.1 software (Michigan State University, East Lansing, MI, USA) was used for statistical analysis. Analysis of variance (ANOVA) tests were performed to determine the effects of the wood, fungus, stain type, and their interactions on some surface properties of wood materials exposed to fungal decay. Duncan's multiple range tests (DMRT) using the least significant difference critical value (LSD) were used to determine the significant differences between the variables.

RESULTS AND DISCUSSION

Total Color Change (ΔE^*)

The antifungal effects of the coloring process on the total color change value were different in terms of wood species, fungus species, and stain types. An ANOVA was used to determine statistical significance of the total color change values in terms of these factors and their interactions. The results are provided in Table 2.

Table 2. ANOVA Results of Total Color Change (ΔE^*) Values

Factors	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Level of Significance ($p \leq 0.05$)
Wood Type (A)	3	778.66	259.55	17.51	0.000*
Fungus Type (B)	1	757.67	757.67	51.14	0.000*
Stain Type (C)	3	5224.65	1741.55	117.55	0.000*
Interaction (AB)	3	437.69	145.89	9.847	0.000*
Interaction (AC)	9	1449.31	161.03	10.86	0.000*
Interaction (BC)	3	1504.80	501.60	33.85	0.000*
Interaction (ABC)	9	1444.56	160.50	10.83	0.000*
Error	160	2370.43	14.81		
Total	191	13967.81			

*Significant at 95% confidence level

The ANOVA results indicate that all factors and their interactions were statistically significant ($P \leq 0.05$). The fungus, wood and stain type factors each affected the total color change value differently. The Duncan's Multiple Range Test (DMRT) using the LSD

critical value were conducted between all factors to see where the significant differences occurred. The results are given in Tables 3 and 4.

Table 3. The DMRT Comparison Results for the Wood, Fungus and Stain Types of the Total Color Change (ΔE^*) Values

Wood Type	\bar{x}	HG
Scotch Pine	15.90	B
Eastern Beech	17.42	A*
Sessile Oak	15.19	AB
Mahogany	11.91	C
LSD \pm 1.552		
Fungus Type	\bar{x}	HG
Brown Rot	13.12	B
White Rot	17.09	A*
LSD \pm 1.097		
Stain Type	\bar{x}	HG
Aniline	23.46	A*
Van Dyke Brown	15.58	B
Chemical	9.13	D
Control	10.83	C
LSD \pm 1.552		
\bar{x} : Average value, HG: Homogeneity group, *: The highest total color change value		

Table 4. The DMRT Comparison Results for the Interactions of Wood, Fungus, and Stain Types of the Total Color Change (ΔE^*) Values

Factors WFS**		Aniline		Van Dyke Brown		Chemical		Control	
		\bar{x}	HG	\bar{x}	HG	\bar{x}	HG	\bar{x}	HG
Scotch Pine	BR	18.49	D-F	13.81	G-J	4.35	M	23.13	BC
	WR	22.72	B-D	14.17	F-I	11.75	E-G	8.18	K-M
Eastern Beech	BR	20.24	C-E	15.70	FG	5.42	LM	10.67	H-K
	WR	41.54	A*	20.78	C-E	12.59	F-I	10.39	I-K
Sessile Oak	BR	21.41	CD	12.95	G-J	7.52	K-M	13.78	G-J
	WR	22.34	B-D	26.41	B	12.68	G-J	8.15	K-M
Mahogany	BR	15.04	F-H	7.02	K-M	7.88	G-I	5.50	L-M
	WR	25.87	B	13.78	G-J	9.47	J-L	4.67	M
LSD \pm 4.389									
\bar{x} : Average value, HG: Homogeneity group, *: The highest total color change value, **W: Wood Type, F: Fungus Type, S: Stain Type, BR: Brown Rot, and WR: White rot									

Among the wood types, beech had the highest total color change value, while mahogany had the lowest value. Previous studies indicated that the white rot fungus can completely deteriorate beech wood. Moreover, it consumes cellulose and hemicellulose after destroying lignin (Tsoumis 1968; Kollmann *et al.* 1975). The higher value of beech wood was attributed to this fact.

According to fungus type, samples exposed to white rot fungus exhibited a greater total color change than samples exposed to brown rot fungus. Previous studies indicated that the white rot fungus degrades pentosans and lignin in the early stages of decay and consumes cellulose and pectin in the middle lamella during the later stages of decay (Tsoumis 1968; Schmidt 2006). This phenomenon, which changes all the characteristics of

the wood, is effective in changing the color properties. There are also previous studies reporting that fungi have an impact on the total color change value of wood and wood-based products (Gorbushina *et al.* 1993; Okino *et al.* 2015; Li *et al.* 2005; Nandika *et al.* 2020).

In comparison of the stain types, aniline yielded the highest value, and chemical yielded the lowest value. Both fungi species use the carbon and nitrogen in aniline as nutrients and can reproduce in environments where these elements exist, which explains why the highest total color change value is obtained when samples are colored with aniline (Boyle 1995). The acidity of the chemical, however, may prevent fungi from spreading on the surface, resulting in less total color change.

The highest total color change value was found in beech samples that were exposed to white rot fungus after staining with aniline at the level of interaction between wood type, fungus type, and stain type factors. White rot fungus is believed to be responsible for this phenomenon due to its ability to rot beech wood completely (Skyba *et al.* 2009). Scotch pine samples stained with chemical stain and then exposed to white rot fungus had the lowest values. Also, unstained mahogany samples exposed to white rot fungus were also in the same homogeneity group. This means that the difference between the groups was insignificant and both groups were at the same level. For Scotch pine, this may be due to the acidity of potassium in the chemical stain and the antifungal properties of the resin it contains (Hu *et al.* 2013; Broda 2020). In mahogany wood, extractives are thought to be effective. Previous studies have also reported that extractives contained in mahogany species provide resistance to fungi (Reilly and Robertson 2006; França *et al.* 2016).

Gloss Change

The arithmetic averages obtained to determine the antifungal effects of the coloring process on the gloss change value were different in terms of wood species, fungus species, and stain types of factors. To analyze the effect of these factors and their interactions on gloss change values, an ANOVA was performed, and the results are given in Table 5.

ANOVA results showed that wood type-fungus type and fungus type-stain type interactions were not significant, but other factors were. The Duncan's Multiple Range Test (DMRT) using the LSD critical value were run between all factors to see where the significant differences occurred. The results are given in Tables 6 and 7.

Table 5. ANOVA Results of the Gloss Change Values

Factors	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Level of Significance ($p \leq 0.05$)
Wood (A)	3	1.97	0.658	22.314	0.000*
Fungus Type (B)	1	0.89	0.89	30.178	0.000*
Stain Type (C)	3	11.5	3.83	130.12	0.000*
Interaction (AB)	3	0.11	0.037	1.259	0.280
Interaction (AC)	9	1.66	0.032	6.255	0.000*
Interaction (BC)	3	0.09	0.079	1.096	0.350
Interaction (ABC)	9	0.7	0.029	0.673	0.000*
Error	160	4.71			
Total	191	21.66			

*Significant at 95% confidence level

Table 6. The DMRT Comparison Results for the Wood, Fungus and Stain Types of the Gloss Change

Wood Type	\bar{x}	HG
Scotch Pine	-0.039	C
Eastern Beech	0.214	A*
Sessile Oak	0.049	B
Mahogany	0.177	A
LSD \pm 0.069		
Fungus Type	\bar{x}	HG
Brown Rot	0.032	B
White Rot	0.168	A*
LSD \pm 0.048		
Stain Type	\bar{x}	HG
Aniline	0.329	A*
Van Dyke Brown	0.195	B
Chemical	0.188	B
Control	-0.313	C
LSD \pm 0.069		
\bar{x} : Average value, HG: Homogeneity group, *: The highest gloss change value		

The highest gloss change was observed in beech and mahogany samples, as shown in Table 6. Scotch pine samples, however, showed a slight decrease in gloss value after fungal degradation. This may have been caused by the yellowish-white color of Scotch pine, which is brighter in its natural state (Sidorov *et al.* 2020). This negative change may also be caused by Scotch pine's natural resin, which has antifungal properties (Ross 2010).

White rot fungus had a much stronger effect on gloss change than brown rot fungus. White rot fungus consumes dark colored lignin while leaving behind light colored cellulose (Kollmann *et al.* 1975; Mai *et al.* 2004; Schmidt 2006). Pure cellulose has a poor light absorption property (Hon 1975). Cellulose is able to absorb light because of the acetal or ketonic carbonyl groups situated at the first carbon atom of the non-reducing glucose unit. Hemicellulose possesses similar characteristics as a result of its structural similarities to cellulose. In contrast to cellulose and hemicellulose, lignin is better at absorbing light and can therefore be more easily degraded (Kılıç and Hafizoğlu 2007). The results may have been influenced by this feature of lignin. There are also previous studies reporting the effect of fungi on the gloss value of wood and wood-based products (Can and Sivrikaya 2019; Peng *et al.* 2021).

Aniline-stained samples had the highest gloss change, while unstained control samples had the lowest gloss change. This may be due to the excessive fiber swelling after staining with aniline. In addition, the aminyl radicals in the aniline stain may have had a reducing effect after fungal degradation. The acid, alkali, and strong oxygen-laden chemicals used can damage the main components of the wood material and cause the loss of its natural gloss (Sönmez 2005).

Table 7 indicates that all unstained control samples decreased in gloss value after fungal decay, while all stained samples increased in gloss value. Particularly striking is the parallelism between the effect of aniline stain on the gloss and total color change values. The strong adhesion of fungi to the aniline stained surface may have resulted in smoother and brighter surfaces.

Table 7. The DMRT Comparison Results for the Interactions of Wood, Fungus and Stain Types of the Gloss Change

Factors WFS**		Aniline		Van Dyke Brown		Chemical		Control	
		\bar{x}	HG	\bar{x}	HG	\bar{x}	HG	\bar{x}	HG
Scotch Pine	BR	0.18	B-H	0.08	E-I	0.05	F-I	-0.71	N
	WR	0.21	B-G	0.13	D-H	0.33	B-D	-0.6	MN
Eastern Beech	BR	0.16	C-H	0.41	B	0.11	D-H	-0.25	J-L
	WR	0.7	A*	0.38	BC	0.23	B-G	-0.05	H-J
Sessile Oak	BR	0.31	B-E	0.01	G-I	0.01	H-I	-0.45	L
	WR	0.33	B-D	0.12	D-H	0.31	B-E	-0.28	KL
Mahogany	BR	0.30	B-E	0.11	D-H	0.26	B-F	-0.11	I-K
	WR	0.41	B	0.23	B-G	0.23	B-G	-0.03	HI
LSD ± 0.1942									
\bar{x} : Average value, HG: Homogeneity group, *: The highest gloss change value **W: Wood Type, F: Fungus Type, S: Stain Type, BR: Brown Rot, and WR: White rot									

Hardness

The hardness changes were different in terms of wood species, fungus species, and stain types of factors. To determine the statistical significance of the hardness change values, an ANOVA test was performed considering wood type, fungus type, and stain type as well as their interactions. The results are given in Table 8.

Table 8. ANOVA Results of the Hardness Change Values

Factors	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Level of Significance ($p \leq 0.05$)
Wood Type (A)	3	1794.29	598.097	10.46	0.000*
Fungus Type (B)	1	772.005	772.05	13.50	0.000*
Stain Type (C)	3	385.15	128.38	2.24	0.030*
Interaction (AB)	3	1938.8	646.26	11.30	0.000*
Interaction (AC)	9	980.92	108.99	1.90	0.080
Interaction (BC)	3	233.35	77.78	1.36	0.250
Interaction (ABC)	9	1216.45	135.16	2.36	0.010*
Error	160	9144.91	57.156		
Total	191	16465.91			

*Significant at 95% confidence level

The ANOVA results showed that wood type-stain type and fungus type-stain type interactions were not significantly different. Other factors and interactions, however, differed significantly. DMRT was applied to the interaction of all factors based on the LSD critical value, and the results are shown in Tables 9 and 10.

The highest hardness change was observed in sessile oak samples, while the lowest was found in mahogany samples, as shown in Table 9. All samples except mahogany had decreased hardness values. Hardwood species growing in tropical regions differ physically from those growing in temperate zones. This wood types are characterized by higher extractive and ash amounts, and lower acetyl levels (Pettersen 1984; Gérard *et al.* 2019). It is possible that the results were influenced by the high density and extractive-rich composition of Mahogany samples.

Table 9. The DMRT Comparison Results for the Wood, Fungus and Stain Types of the Hardness Change

Wood Type	\bar{x}	HG
Scotch Pine	-5.331	B
Eastern Beech	-5.229	B
Sessile Oak	-7.250	A*
Mahogany	0.395	C
LSD \pm 1.91		
Fungus Type	\bar{x}	HG
Brown Rot	-2.849	B
White Rot	-6.859	A*
LSD \pm 2.155		
Stain Type	\bar{x}	HG
Aniline	-7.094	A*
Van Dyke Brown	-4.750	B
Chemical	-3.385	C
Control	-4.188	B
LSD \pm 1.72		
\bar{x} : Average value, HG: Homogeneity group, *: The highest hardness change value		

The samples destroyed by white rot fungus had the greatest change in hardness, and the samples destroyed by brown rot fungus had the least change. The cell wall contains lignin as well as cellulose and hemicellulose. The hardness of the woody structure in plants is largely related to the amount of lignin (Lebo *et al.* 2001; Young 2008; Hatakeyama and Hatakeyama 2010). By destroying the lignin, the white rot fungus leaves behind a cottony structure rich in cellulose. In contrast, brown rot fungus destroys cellulose and leaves behind a lignin-rich, brittle, crumbly but relatively sturdy structure (Geib *et al.* 2008). It can be argued that these phenomena are the reason for the difference in the effects of fungal species. Aniline stained samples showed the greatest hardness change when compared to samples stained with other stains. There were no significant differences between the Van Dyke brown stain group and the control group. Samples colored with chemical stain showed the least change. Results may have been affected by carbon-based petrochemical nature of aniline stain and physical coloring feature.

Table 10. The DMRT Comparison Results for the Interactions of Wood, Fungus and Stain Types of the Hardness Change

Factors WFS**		Aniline		Van Dyke Brown		Chemical		Control	
		\bar{x}	HG	\bar{x}	HG	\bar{x}	HG	\bar{x}	HG
Scotch Pine	BR	-24.42	I	-2.41	B-E	-4.167	C-G	-5.75	C-H
	WR	-4.66	B-G	-3.41	B-F	-3.58	B-F	-1.25	B-D
Eastern Beech	BR	3.83	AB	-4.16	B-F	-1.16	B-D	-1.33	B-D
	WR	-14.01	H	-11.02	E-H	-7.01	GH	-9.04	D-H
Sessile Oak	BR	-9.00	D-H	-0.83	A-D	-2.50	B-E	-3.16	B-F
	WR	-8.83	D-H	-13.83	H	-8.33	D-H	-11.50	F-H
Mahogany	BR	1.66	A-C	-1.66	B-D	-2.83	A-C	7.66	A*
	WR	-1.33	B-D	0.66	A-D	-4.16	B-F	-1.16	B-D
LSD \pm 8.62									
\bar{x} : Average value, HG: Homogeneity group, *: The highest hardness change value **W: Wood Type, F: Fungus Type, S: Stain Type, BR: Brown Rot, and WR: White rot									

Aniline stained beech samples exposed to white rot fungus showed the greatest changes in hardness, as shown in Table 10. Unstained (control) mahogany samples exposed to brown rot fungus showed a positive change in hardness. Brown rot fungus primarily destroys the secondary wall of cells, which contains a small amount of lignin. The spreading rate slows down when it reaches the primary wall due to the high content of lignin (Carlquist 1988; Richter 2015). In wood exposed to brown rot fungus, the cell structure is preserved for a considerable period of time because the primary wall remains intact. Extractives increase density, hardness, and compressive strength (Carlquist 1988; Kollmann *et al.* 1975; Gérard *et al.* 2019). The high amount of lignin and extractives in mahogany wood may explain its high hardness.

CONCLUSIONS

1. The staining process reduced the hardness and gloss values of the samples that were exposed to both fungus types.
2. In comparison to the control group, chemical staining produced a positive effect on color change values. Conversely, aniline and Van Dyke stains produced inferior results.
3. Chemical staining resulted in positive hardness change values, whereas aniline staining resulted in negative hardness change values.
4. Contrary to the aim of the research, the samples stained with aniline failed compared to the unstained (control) samples.
5. Chemical stains have an antifungal effect on rot fungi. They can be used for coloring purposes and as a preservative in some situations.

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