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Decay Resistance of the Acetylated Tropical Hardwood Species

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Abstract

Chemical modification of wood is an effective method to enhance the biological durability of wood with no toxic effect on the environment. In this study, wood of Triplochiton scleroxylon was modified using acetylation techniques. A total of one hundred wood blocks, (each 20×20×60 mm) obtained from a 22-year old T. scleroxylon tree were conditioned and acetylated at 120°C in a bioreactor containing acetic anhydride for 60, 120, 180, 240 and 300 minutes. The percentage weight gain of acetylated wood was determined. The untreated (control) and treated blocks were exposed to Pleurotus ostreatus (white rot fungus) and Fibroporia vaillanti (brown rot fungus) after which moisture content (MC) and weight loss (WL) was monitored for 16 weeks. Data were analysed using descriptive and inferential statistics at $p<0.05$ level of significance. The percentage weight gain of acetylated wood samples increased with time from 10.4% (60 minutes) to 22.7% (300 minutes). MC of untreated blocks inoculated with Pleurotus ostreatus was significantly higher than those of Fibroporia vaillantii after 16 weeks exposure. There was no significant difference in the MC of the of the acetylated samples for the two fungi after 300 minutes reaction time. The WL of untreated blocks inoculated with Fibroporia vaillantii was higher than those of Pleurotus ostreatus, however, the two fungi showed no significant difference in the WL for the acetylated samples after 16 weeks exposure. Acetylation prevents moisture absorption and inhibition of fungi growth in acetylated wood compared to untreated wood, thereby enhancing the durability of Triplochiton scleroxylon.

Key Words: acetic anhydride, fibroporia vaillantii, pleurotus ostreatus, triplochiton scleroxylon, weight loss

Introduction

The fibrous nature of wood has made it one of the most versatile raw material for a variety of uses (Rahman 2018). However, the use of wood is limited by factors such as biodegradability, flammability, swelling and shrinking. This is due to the fact that the hemicellulose polymers in wood are accessible, hygroscopic and contain sugar residues that attract microorganisms which degrade wood (Rowel 2012). As a result, wood-destroying fungi (white rot, white-rot and soft-rot) invade wet wood that remains damp for long periods. It is therefore necessary to prolong the service life of wood by conventional methods using wood preservatives. Wood preservatives are substances that effectively prevent the development and action of fungi, termites and pests, for a reasonable length of time (Thomasson et al. 1989). Conventional wood impregnation methods (water or oil type preservatives) are based primarily on the use of toxic chemicals. Environmental concerns, particularly with regard to disposal of treated wood at the end of product life,

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are now causing restrictions to be imposed upon the utilization of conventional chemical treatments (Papadopoulos 2010).

In Nigeria, serious concerns have been raised on the use of non-biodegradable and hazardous preservative chemicals which are common in the timber processing industry. Globally, increased awareness on the need for proper disposal of treated wood at the end of their product life, has led to restrictions on the use of these conventional chemical preservatives. Consequently, the search for environmentally friendly wood treatment techniques have become a central focus of research (Van Acker and Hill 2003; Fojutowski et al. 2013). One of such techniques is chemical modification of wood. This modification process enhances the long-term service of the wood while reducing the risks to human health (Militz et al. 1997). Acetylation is the most widely used reaction for chemical modification of wood. It involves the use of, acetic anhydride or acetyl chloride for modifying the chemical structure of the wood (Hill 2006). It has been documented that wood could be modified chemically so as to enhance some properties of wood in a more or less permanent ways (Rowell 1983; Papadopoulos 2010). Chemical modification involves the formation of a chemical bond between the reagent and the cell wall polymers of wood. The general principle of chemical wood modification is the reaction of a chemical agent with functional group (mostly hydroxy groups) of the cell wall polymers and the formation of covalent bonds. This causes a change in the chemical and physical characteristics of wood (Rowell 1983; Norimoto 2001; Hill 2006). As a result of introduction of bulkier groups within the cell wall, the dimensional stability increases, weathering resistance improves and equilibrium moisture content decreases (Eaton and Hale 1993; Paul and Robert 2001).

Much work has been done to determine the decay resistance of acetylated wood to different fungi (brown rot and white rot) attack (Perterson and Thomas 1978; Suittle et al. 1999) and also differences in the hygroscopicity of brown rot and white rot has been reported (Cowling 1961; Anagnost and Smith 1997; Chauhan and Nagaveni 2010). It was found that wood decay by brown rot and white rot absorbed moisture differently as compare to sound wood. Although, several studies have been conducted on the effect of acetylation on durability of wood against fungi attack, however, little or no information is available on the effect of acetylation on moisture absorption and durability of tropical wood species exposed to brown rot and white rot fungi.

Triplochiton scleroxylon K. Schum (Obeche) is a high profit, indigenous, non-durable timber species in the Nigerian timber market. This versatile raw material has a huge volume supply, because of its ability to grow under plantation management (Ogunsanwo and Onilude 2000). However, its low durability predisposes it to fungal attack which results in wood decay. Hence, wood treatment with preservatives is essential for prolonging its durability (Adetogun 1998). Some of the chemical used for preserving Obeche wood include creosote, arsenic, zinc, copper, chromium, and biocides (Ogunsanwo and Adedeji 2010). Hence, the potential use of acetylation for modification of *Triplochiton scleroxylon* wood to enhance its durability has not been explored. The purpose of this study is to chemically modify *Triplochiton scleroxylon* wood at varying acetylation periods to determine the moisture absorption and decay resistance of the acetylated wood to white (*Pleurotus ostreatus*) and brown rot (*Fibroporia vaillanti*) fungi.

Materials and Methods

Wood preparation and acetylation procedures

Wood specimens obtained from a 22-year-old tree (*Triplochiton scleroxylon*) were converted into 20×20×60 mm (radial×tangential×longitudinal) wood blocks with no defects. The wood blocks were weighed, then oven dried at 105±2°C till a constant weight was reached and this was recorded as W_1. Chemical modification was achieved following the methods of (Anne-Marie et al. 1987; Mohebby 2008; Giotra 2009). For acetylation, wood samples (MC ~7%) were introduced into stainless steel pressure reactor vessels containing the acetylation liquid, (i.e., acetic anhydride and acetic acid (92:8)). In order to achieve pre-impregnation of the wood with the reagent, the temperature was set at 25°C and 10 -15 bar for 30 minutes Then, the reaction temperature was set at 120°C for 0, 60-, 120-, 180-, 240- and 300 minutes with 20 wood blocks per period of exposure. After the acetylation reaction, the reactor was cooled in an ice bath. The wood samples were extracted and washed several times to remove excess acetic anhydride and the by product (i.e. acetic acid) until no acid smell was de-

tected at pH 7.4. The wood samples were air-dried and then oven dried at 105±2°C for 24 h, the weight was recorded as W_2. The weight gain (WG) and volume gain (VG) were calculated using Equation 1.

$$\text{WGP}\,(\%) = \frac{W_2 - W_1}{W_1} \times 100 \quad (1)$$

Where,
W_1 = Oven − dry weight of samples before treatment (g)
W_2 = Oven − dry weight of samples after treatment (g)

Preparation of culture medium and infection of treated wood samples

A nutrient medium of Potato Dextrose Agar (PDA) was prepared by mixing 35 g of PDA with 1 litre of distilled water in a conical flask and then homogenizing the solution. After homogenization, 40 ml of PDA was poured into McCartney bottles and sterilized by autoclaving at 0.1 N/mm^2 and 120°C) for a period of 20 minutes. The bottles were laid sideways so that the medium could be retained in the neck of the bottle. The inoculums of white rot (*Pleurotus ostreatus*) and brown rot (*Fibroporia vaillantii*) fungi were obtained from Department of Microbiology, University of Ibadan, Nigeria. These were inoculated into the medium within 6 days after preparation of the bottles (Arun 2006; Sarker et al. 2006). The bottles were incubated at room temperature (27±2°C) in the laboratory.

After the growth of the mycelia of the test fungi in the McCartney bottles, the blocks consisting of sixteen (16) replicates for each treatment were placed in the bottles in such a way that they were in contact with the aerial mycelium of the fungus and not the medium, in order to avoid the leaching of the chemicals into the PDA substrate (Adegeye et al. 2009). The experiment was laid in a completely randomised design with factorial arrangement {2 (fungi)×6(acetylation periods)×4(time of exposure)} and then variation in moisture content and weight loss were monitored, monthly for 16 weeks at 27±2°C.

Moisture content and weight loss determination

At the end of each monthly incubation period, four (4) test blocks were randomly removed from the McCartney bottles, carefully cleaned of the adhering mycelium, with special care in order not to remove the splinters of wood. They were weighed immediately and recorded as W_3, then oven-dried at 105±2°C, until a constant weight was reached. The samples were allowed to cool over a silica gel in a desiccator and then weighed, W_4. The moisture content and weight loss of the samples were determined using equation 2 and equation 3 respectively.

$$\text{Moisture Content}\,(\%) \frac{W_3 - W_4}{W_4} \times 100 \quad (2)$$

$$\text{Weight loss}\,(\%) = \frac{W_2 - W_4}{W_2} \times 100 \quad (3)$$

Data analysis

Analysis of variance was used to determine the main and interaction effects of the fungi (white rot and brown rot), acetylation period and time of exposure to decaying agents on the weight loss and moisture content of Obeche blocks. Data obtained were analysed using analysis of variance (ANOVA). Treatment means were separated using the Duncan Multiple Range test at 95% level of significance.

Results and discussion

Weight gian (WG) of acetylated Triplochiton scleroxylon wood samples

Weight gain ranged from 10.37-22.74%, as presented in Fig. 1. Wood samples treated for 300 minutes had the highest WG (23%). There was a linear relationship between reaction time and WG. Hence, increased penetration of the chemical reagent into the cell wall occurred with increased reaction time. The WG indicates the chemical reaction that occurred inside the cell wall rather than just within the cell lumen. Thus, the wood samples did not only increase in weight but also swelled with the acetyl group occupying space in the cell wall. WG of 7.7% for two hardwood species (*Tectona grandis* and *Gmelina arborea*) at a reaction time of 360 min at 70°C was reported (Pardo 2013). Similarly, 11.96% and 12.18% WG have been observed for,

Calophyllum brasiliense and *Enterolobium cyclocarpum* respectively, at a reaction time of 420 min and 90°C. The study further opined that higher reaction time coupled with lower reaction temperature, did not significantly increase WG (Blanco and Alfaro 2014). Nevertheless, the temperature of the wood should not be permitted to go above 160°C, which may lead to overheating, resulting in darkening or degradation of the wood (Neogi et al. 2007).

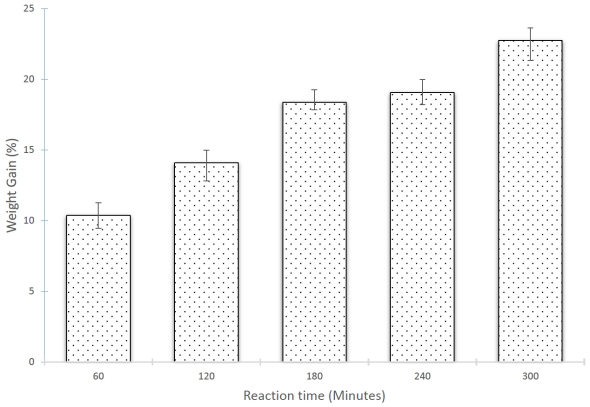

Fig. 1. Weight gain percentage of acetylated *Triplochiton scleroxylon* wood.

Moisture content of acetylated Triplochiton scleroxylon wood samples inoculated with pleurotus ostreatus (white rot) and Fibroporia vaillantii (brown rot)

The moisture content of the untreated samples inoculated with *Pleurotus ostreatus* and *Fibroporia vaillantii* was above 30% after 4 weeks of exposure (Tables 1 and 2). As the period of exposure increased, the moisture content of the untreated samples also increased to 73% and 65% for *Pleurotus ostreatus* and *Fibroporia vaillantii* respectively at 16 weeks (Tables 1 and 2). However, the moisture content of

Table 1. Moisture content of *Triplochiton scleroxylon* exposed to *Pleurotus ostreatus* for 16 weeks

Reaction time (min)	Moisture content (%) (*Pleurotus ostreatus*)			
	Exposure period (weeks)			
	4	8	12	16
Untreated	36.80 (3.9)^{a}	56.44 (7.9)^{a}	68.79 (3.4)^{a}	73.74 (6.2)^{a}
60	27.21 (3.1)^{b}	42.2 (5.5)^{b}	49.95 (7.5)^{b}	55.42 (7.7)^{b}
120	27.18 (2.1)^{b}	48.75 (3.0)^{b}	53.81 (4.7)^{b}	50.43 (7.1)^{b}
180	24.89 (2.9)^{b}	33.38 (2.6)^{c}	42.51 (1.9)^{c}	51.07 (10.3)^{b}
240	22.87 (1.7)^{b}	32.8 (4.8)^{c}	39.68 (5.3)^{c}	45.77 (3.3)^{c}
300	24.82 (3.0)^{b}	32.57 (5.6)^{c}	41.2 (4.4)^{c}	43.77 (3.4)^{c}

Standard deviations are in parentheses. Means in the same column with same alphabet are not significantly different from each other at $\alpha=0.05$.

Table 2. Moisture content of *Triplochiton scleroxylon* exposed to *Fibroporia vaillantii* for 16 weeks

Reaction time (min)	Moisture content (%) (*Fibroporia vaillantii*)			
	Exposure period (weeks)			
	4	8	12	16
Untreated	37.55 (6.1)^{a}	48.15 (4.7)^{a}	57.2 (4.8)^{a}	64.74 (8.5)^{a}
60	26.12 (1.9)^{b}	37.47 (2.8)^{b}	37.78 (2.7)^{c}	52.26 (4.6)^{b}
120	23.73 (2.1)^{b}	36.84 (5.1)^{b}	42.49 (4.3)^{b}	47.61 (1.1)^{bc}
180	28.37 (4.9)^{b}	35.51 (3.5)^{b}	40.12 (6.1)^{b}	46.19 (0.8)^{bc}
240	24.72 (4.3)^{b}	32.44 (1.3)^{b}	34.97 (4.2)^{c}	39.88 (4.0)^{c}
300	23.00 (2.3)^{b}	35.53 (3.3)^{b}	37.19 (4.2)^{c}	43.86 (5.8)^{c}

Standard deviations are in parentheses. Means in the same column with same alphabet are not significantly different from each other at $\alpha=0.05$.

acetylated wood samples after 4 weeks ranged from 22.87-27.21% and did not exceed 43.77-55.42% range after 16 weeks for *Pleurotus ostreatus* and *Fibroporia vaillantii*. The moisture content of the untreated and acetylated samples inoculated with white and brown rot fungi increased as the period of exposure increased (Tables 1 and 2). However, the untreated samples inoculated with white rot fungus exhibited significant higher moisture content than the samples inoculated with brown rot fungus after 16 weeks. Statistical analysis revealed that at 5% level there are variations in the moisture content for fungi used, period of exposure and acetylation levels. There was no significant difference in the moisture content of all the acetylated samples over the period of exposure. However, the untreated samples had significantly higher values than other treatments for the two fungi.

The decrease in moisture content of acetylated wood samples when compared to untreated wood samples, indicates that acetylation increases hydrophobation (wood resistance to water). Fojutowski et al. (2013) studied the effect of acetylation of alder, beech, birch and poplar wood on decay caused by *Coniophora puteana* and reported decrease in moisture content of all the acetylated hardwood inoculated with fungi compared with the untreated samples. In this present study, moisture content of the acetylated *Triplochiton scleroxylon* wood increased above 40% after 16 weeks of exposure to brown rot and white rot fungi. This could be because the Fibre Saturation Point (FSP) of wood (approximately 25-35%) is an important factor for fungal colonization. Fungi require wood moisture content to be above FSP for colonisation to take place. Under these conditions, free water is available in the lumen of the wood cells and thus mycelia growth will be encouraged (Eaton and Hale1993). Since fungal attack is related to moisture content, weight loss resulting from fungal attack is used to determine the effectiveness of a chemical treatment to protect wood from decay.

Fungi attack on acetylated Triplochiton scleroxylon

The weight loss increased with increase in exposure period and ranged from 1.94-9.19% for treated wood samples exposed to *Pleurotus ostreatus* (white rot) while those exposed to *Fibroporia vaillantii* (brown rot) ranged from 0.92-14.31% after 4 weeks (Tables 3 and 4). After 16 weeks of exposure, the weight loss of the untreated samples was 44% and 50.16% for blocks exposed to white rot and brown rot fungi, respectively. However, the weight loss of acetylated samples at different reaction time ranged between 3.54-11% and 3.47-19.91% for wood exposed to white rot and brown rot fungi respectively. There was a decrease in weight loss with increase in period of acetylation (Tables 3 and 4).

At 4, 8, 12 and 16 weeks, weight loss of untreated samples exposed to brown rot and white rot were significantly higher than acetylated wood samples, irrespective of the reaction time. The percentage weight loss of 300 min acetylated wood samples exposed to brown rot was higher than those exposed to white rot fungus. There was no significant difference between the weight loss of all acetylated wood samples exposed to white rot and brown rot fungi.

Table 3. Weight loss of *Triplochiton scleroxylon* exposed to *Pleurotus ostreatus* for 16 weeks

Reaction time (min)	Weight loss (%)			
	Exposure period (weeks)			
	4	8	12	16
Untreated	9.19 (1.6)^{a}	13.39 (2.1)^{a}	20.77 (2.8)^{a}	44.01 (3.3)^{a}
60	2.73 (1.3)^{b}	6.55 (0.9)^{b}	7.89 (0.6)^{b}	11.04 (2.3)^{b}
120	2.13 (0.6)^{b}	3.16 (0.8)^{c}	5.09 (0.7)^{bc}	5.01 (0.7)^{c}
180	2.5 (0.6)^{b}	3.00 (0.8)^{c}	4.83 (0.8)^{c}	5.07 (0.8)^{c}
240	2.04 (0.6)^{b}	3.39 (0.9)^{c}	3.62 (0.7)^{c}	3.13 (0.5)^{c}
300	1.74 (0.7)^{b}	3.35 (0.4)^{c}	3.25 (0.7)^{c}	3.54 (0.8)^{c}

Standard deviations are in parentheses. Means in the same column with same alphabet are not significantly different from each other at $\alpha=0.05$.

Table 4. Weight loss of *Triplochiton scleroxylon* exposed to *Fibroporia vaillantii* for 16 weeks

Reaction time (min)	Weight loss (%)			
	Exposure period (weeks)			
	4	8	12	16
Untreated	14.31 (1.5)[a]	20.47 (2.3)[a]	31.32 (2.3)[a]	50.16 (3.5)[a]
60	3.94 (0.4)[b]	4.69 (1.1)[b]	6.78 (1.1)[b]	19.91 (2.9)[b]
120	3.94 (0.63)[b]	2.71 (0.3)[b]	6.87 (0.3)[b]	9.39 (1.6)[c]
180	3.94 (0.9)[b]	2.82 (0.6)[b]	2.94 (0.6)[c]	3.39 (1.1)[d]
240	3.94 (0.5)[b]	2.61 (0.5)[b]	2.9 (0.5)[c]	3.76 (0.8)[d]
300	3.94 (0.5)[b]	1.01 (0.4)[b]	2.72 (0.8)[c]	3.47 (0.7)[d]

Standard deviations are in parentheses. Means in the same column with same alphabet are not significantly different from each other at $\alpha=0.05$.

Weight loss is an important parameter for assessing decay resistance of solid wood (Lomelí-Ramírez et al. 2009). The cumulative increase in weight loss due to the microbial activities of white rot and brown rot fungi highlights the protective effect of the chemical modification of the wood samples (Sandberg et al. 2017). The results are similar to those reported by Calonego et al. (2010), that weight loss in untreated Eucalyptus grandis wood were 50.33% and 34.32% for the samples incubated in substrate containing brown-rot and white-rot fungi. In this study, increased reaction time increased the WG and resulted in a decrease in weight loss after exposure to fungi attack.

Brown-rot fungus resulted in higher weight loss in untreated and acetylated wood samples when compared with white-rot fungus. This could be attributed to the fact that brown-rot fungus tends to degrade cellulose and hemicelluloses by depolymerization without extensive loss of lignin. Hemicellulose degradation has a high impact on the mechanical properties of wood during brown-rot decay is significant (Curling et al. 2000; Curling et al. 2001). Since white-rot decay fungus degrades the lignin component of wood before other components, wood samples exposed to it may not experience any appreciable reduction in weight during the early stages of decomposition. This is because the lignin component of wood has less of structural role as compared to cellulose.

It has been reported that levels of acetylation that caused 17-20% WG generally increased wood resistance to decay caused by fungi. An 18% WG achieved after 180 min of acetylation, resulted in a low weight loss which was not significantly different from those of samples treated for 240 and 300 minutes. Hence, acetylation that confers a WG of 18% and above provided sufficient protection against fungi attack. This corroborates the assertion of Mohebby (2003) that no decay pattern was observed for acetylated beech wood (fagus) with17.15% WG, that was inoculated with *Trametes versicolor*. In addition, Beckers et al. (1994) determined the decay protection threshold levels for acetylated Scot pine (*Pinus sylvestris*) exposed to a variety of wood decay fungi and reported that a 18% WG provided resistance against *Coniophora puteana* and *Gloeophyllum trabeum*. The high resistance of chemically modified wood against fungal decay has been attributed to the reduction of moisture content (fibre saturation point), modifications of the cell wall polymers (polymers become unrecognizable to enzymes) and a lowering of the micro-pore size in the wood cell wall, such that the blockage of the micro-pores and cell wall bulking effectively inhibit the penetration of low molecular weight diffusible agents, which are required for fungal degradation (Papadopoulos and Hill 2002; Hill et al. 2005; Hill 2006). Nevertheless, the high resistance against fungal decay is assumed to be due to changes in the material properties of wood rather than toxic effect on fungal physiology. Invariably, acetylated wood could be colonised by decay fungi, since modified wood is not toxic to the fungi, but degradation of the cell wall is impeded by chemical modification (Hill 2006).

Conclusion

The results from this study have demonstrated that decay resistance of *T. scleroxylon* against *P. ostreatus* (white rot fungus) and *F. vailantii* (brown rot fungus) wood is dependent on the repellence of the acetylated wood to moisture absorption. As the weight gain due to reaction with acetic anhydride increases, moisture absorption reduces and resistance to attack by white and brown-rot fungi increases. Thus, it could be concluded that acetylation modified the wood structure such that it reduced moisture absorption and inhibited the growth of the fungi, thereby protecting the wood species against white and brown rot fungi.

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