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EVALUATION OF HONEY BEE PROPOLIS AS WOOD PRESERVATIVE ON *Triplochiton scleroxylon* USING MAXIMUM COMPRESSIVE STRENGTH

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ABSTRACT

This study was carried out in order to investigate the fungi toxic potentials of Honey bee Propolis, using the wood of *Triplochiton scleroxylon.* Propolis Extract (PE) was obtained using absolute ethanol as solvent. The inhibition of PE over two decay fungi, *Coriolopsis polyzona* and *Coniophora puteana* was tested using four different concentration levels. The efficacy of PE was evaluated using Maximum Compressive Strength (MCS) on wood blocks of *T. scleroxylon* after 16weeks of incubation. Data obtained were analyzed using descriptive statistics and 2-way Analysis of Variance. Results were significant ($P<0.01$), recording the mean Maximum Compressive Strength (MCS) of blocks exposed to *C. polyzona* of 18.64±0.24N/mm^2 for control and 25.45±0.28 N/mm^2 at 50% concentration, while test blocks exposed to *C. puteana* was 16.31±0.20N/mm^2 for control and 21.22±0.08N/mm^2 at 50% concentration respectively. The study concluded that Propolis extract was effective for controlling the test fungi and recommended the use of other solvents and fungal strains for the evaluation of propolis antifungal activity.

Keywords: Propolis; wood preservative; maximum compressive strength

INTRODUCTION

In order to ensure a long, useful and safe life, timber needs protection from the hazards of fungal decay and weathering (Hyvonen *et al.,* 2005). Wood treated with conventional preservative is classified as hazardous waste (Hyvonen *et al*., 2005). The current trend therefore is to seek alternatives to synthetic chemicals with attention focused on the use of natural products of plant origin; which are not only effective, but also biodegradable (Ogunsanwo *et al.,* 2008).

Studies have shown that Propolis of the Honeybee (*Apis mellifera*) has antifungal and insecticidal properties (Sforcin *et al*., 1995; Obasa *et al.,* 2007; Osipitan *et al*., 2010). Typically, propolis contains approximately 50% resins and vegetable balsams, 30% waxes, 10% essential oils and 5% pollen (Orsi *et al.,* 2005). The use of propolis in wood utilization is given little attention in developing countries, especially in West Africa. Most honey

hunters harvest only honey, paying little attention to the valuable antibiotic called Propolis. In hives (wood) propolis reinforces the structural stability, reduces vibration, make hives more defensible by sealing alternative entrances and prevention against diseases and parasites (Krell, 1996). It therefore, becomes necessary to investigate the fungicidal potentials of Propolis for the development of environmentally friendly wood protecting chemicals.

Fungi are the major causes of decay in wood. As these organisms grow through the wood, they alter its chemical structure and remove mass thereby altering its mechanical properties (Adetogun *et. al.*, 2003). Raberg *et al*., (2005) observed that strength tests are the most effective for detecting decay in wood. According to Levi (1978) in Adetogun *et al*., (2003), wood derives its strength from a combination of highly oriented cellulose micro fibrils and encrusting hemicelluloses. Any change in these carbohydrates often cause sharp reduction in wood strength and properties, and since these properties drop much more rapidly than its weight, it is theoretically and practically possible to determine the effectiveness of a preservative much more rapidly using strength rather than weight loss (Adetogun *et al.*, 2003). This study assesses the preservative potential of propolis using strength loss assessment method.

MATERIALS AND METHODS

Study Area

The wood for the study was sourced from a natural forest patch within Cocoa Research Institute of Nigeria (CRIN), Ibadan. The Institute lies on latitude $07^0$25'N and longitude 3^0 53'E. It is approximately 12km from Ibadan city. The annual rainfall is 1257mm. The relative humidity ranges between 84.5% (June to September) and 78.8% (December to January). The mean annual temperature ranges from 21.0 to 31.3^{o}C (Shomade, 2000).

Field Work

The field work involves accelerated laboratory decay tests and strength loss tests, which was carried out at the Organic Laboratory of the Chemistry Department, Obafemi Awolowo University, Ile-Ife and Pathology Laboratory of the Forestry Research Institute of Nigeria (FRIN), Ibadan, respectively.

Extraction of Propolis and Preservative Formulation

Propolis was extracted in alcohol and test preservative was formulated using the volume to volume method where 1ml of extract (propolis) in 99 ml of ethanol (solvent) is equivalent to 1% dilution (Adetogun, 1998). The preservatives were tested using four concentration levels thus: 25%, 50%, 75% and 100%.

Test Block Preparation

Samples were prepared from sapwood of good quality, with the grains of the wood following the longitudinal axis of the tree (Adetogun, 2003). The blocks were prepared for test by drying and sterilizing in the oven at 103^0C until constant weight was achieved (Adetogun *et al.*, 2009; Adetogun, 2011). The weight obtained immediately after oven drying

was considered as initial dry weight (W1), in accordance with Adetogun (1998) and Ajala (2014).

Growth Medium Preparation

Synthetic Potato Dextrose Agar (PDA) was used as fungal culture and prepared according to Ogunsanwo *et al.* (2008) and Ajala (2014). A white and a brown rot-fungi, *Coriolopsis polyzona* (Pers) RYV and *Coniophora puteana* (Schum) Fries, respectively, were obtained from the Mycology unit of the Forestry Research Institute of Nigeria, and prepared in accordance with Adetogun (1998).

Treatment of Test Blocks

Dipping impregnation method (FAO, 1986; Adetogun, 1998; Olajuyigbe, 2007; Ogunsanwo *et al.*, 2008) was used to treat test blocks with the preservatives. The same procedure was used for blocks treated with solvent only. Control blocks were not treated with propolis. The blocks were weighed to determine the rate of absorption.

Laboratory Decay Test

Test blocks were incubated at a temperature of 25±2^{o}C, in Kolle flasks each containing four (4) pairs of test blocks of the same preservative concentration treatment, on PDA medium, with fully grown mycelia of a monoculture of the test fungi (Adetogun *et al*., 2009). Test blocks were incubated for 16weeks. At the end of the incubation period the blocks were cleaned with dry cotton wool to remove adhering fungal mycelia and oven dried. The weight of test blocks after incubation was taken as the final weight.

Strength Test Determination

Test samples measuring 5x2.5x1.5cm (Adetogun, 2011) were used for the study. Both the control (untreated) and fungi infested samples were subjected to strength tests. Wood samples used for weight loss determination after inoculation was also used for the strength test. A Hounsfield tensiometer was used; applying load at the rate of 1.01 mm/s, and the corresponding force at failure point was directly read on the scale and recorded. This force was divided by the cross-sectional area of the test block. Maximum compressive strength was then determined using equation 1, according to Adetogun (1998) and Ajala (2014):

$$\text{MCS} = \frac{p}{bd} \quad \text{..... Eqn 1,}$$

Where: p is the applied load in N, b is the width of specimen in mm, and d is the depth in mm.

Data Analysis

The major factors considered in this study to form sources of variation after inoculation are: five (5) concentration levels: (0%, 25%, 50%, 75% and 100%) and two (2) fungi: (*C. polyzona* and *C. puteana*). Data collected were analyzed using Two-way Analysis

of Variance, Descriptive Statistics and where significant difference exists, Least Significant Difference (LSD) at 1% level of significance ($p<0.01$) was used to separate the means.

RESULTS AND DISCUSSION

Maximum Compressive Strength Parallel to Grain

Raberg *et al*. (2005) observed that strength tests are the most effective for detecting decay in wood. The results of maximum compressive strength parallel to grain (MCS) of wood samples of *T. scleroxylon* treated with various concentrations of propolis and inoculated with *Coriolopsis polyzona* and *Coniophora puteana* are presented in Tables 1 to 3. Table 1 revealed that the mean MCS of samples inoculated with *Coriolopsis polyzona* ranged from 18.64N/mm^2±0.240, for the control to 25.45N/mm^2±0.28, at 50% concentration. The MCS values increased with concentration level up to 50% PE concentration level, before decreasing.

For samples inoculated with *Coniophora puteana*, the mean MCS ranged from 16.31 N/mm^2±0.20, for the control, to 21.22N/mm^2±0.085, at 50% concentration. The reduction in strength was greater in *Coniophora puteana* (Brown-rot fungi) than in *Coriolopsis polyzona* (white rot fungi).

Table 1: Mean MCS values of *T. scleroxylon* after inoculation with *C. polyzona and C. puteana*

	MCS (N/mm^2)	
Propolis Conc (%)	*C. polyzona*	*C. puteana*
TC (Control)	18.64 ± 0.240	16.31 ± 0.200
0	20.58 ± 0.030	17.24 ± 0.160
25	23.46 ± 0.250	20.77 ± 0.170
50	25.45 ± 0.280	25.45 ± 0.280
75	18.78 ± 0.050	18.89 ± 0.100
100	19.15 ± 0.082	18.23 ± 0.061

Source: field work, 2014

Table.2: ANOVA of the effect of fungi and concentration on MCS of *T. scleroxylon*

Source of Variation	Sum of Squares	DF	Mean Square	F- Value	Sig
Fungi	41.983	1	41.983	258.621	.000***
Concentration	533.624	5	106.725	657.433	.000***
Fungi * Concentration	31.961	5	6.392	39.376	.000***
Error	9.740	60	0.162		
Total	617.308	71			

***: Significant ($p<0.01$)

The analysis of variance (Table 2), revealed that both test fungi and concentration of PE have significant ($p<0.01$) effect on the MCS of wood samples of *T. scleroxylon*. The result

further revealed that the interaction between fungi and concentration of PE have significant ($p<0.01$) effect on the MCS of wood samples. Table 3 revealed that concentrations 75 and 100% are the same, while concentrations 25 and 50% are different from each other; affecting the MCS differently.

Table. 3: Post Mortem Analysis (LSD) showing Averages of MCS for the two fungi

Parameters	Solvent	25%	50%	75%	100%	Control
MCS	18.91 ± 1.7644a	22.12 ± 1.4893b	25.45± 0.3838c	18.84± 0.5674a	18.69± 0.5082a	17.47± 1.2447d

Values along the same row with different superscripts are significantly different at 1% level.

The result follows similar observation of Ajala (2014) in which MCS values increased with concentration up to 50% level of heartwood extract of *Gliricidia sepium* before decreasing. The author further observed that the decrease in MCS at higher concentrations was due to the negative effect of the preservative, by tearing the wood fibres apart. The greater reduction in strength, in *Coniophora puteana* (Brown-rot fungi) than in *Coriolopsis polyzona* (white-rot fungi), conforms to that of Zabel and Morrell (1992), who observed that brown rot fungi usually have a greater strength reduction on wood. Ajala (2014) reported that reduction in the compressive strength is a good indicator for the evaluation of the preservative properties of chemicals.

The reduction in MCS at higher PE concentration conforms with the assertions of Wilcox (1978), Roll (2003) and Ibach (2006), that mechanical properties are affected by wood decay though strength loses are greatly dependent on fungi type and to a lesser extent on the wood species involved, and also conforms with Ajala (2014), who reported that the higher the concentration the higher the reduction in MCS. Test fungi also have significant effect on the mechanical properties of wood samples (Adetogun *et al*., 2003; Humar *et al*., 2006; Larnoy *et al*., 2006; Singh *et al*., 2006; Tanaka *et al*., 2006; Ajala, 2014).

CONCLUSION

The study revealed that the MCS differs for both fungi and is affected by extract concentration. *Coniophora puteana* (brown rot fungi) is more destructive on the wood of *T. scleroxylon* than *Coriolopsis polyzona* (white rot fungi). Propolis is able to control the test fungi that degrade wood.

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