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ANTIFUNGAL EFFECTIVENESS OF *Cassia fistula* L. SEED OIL ON *Ficus mucuso* Welw. ex Ficalho WOOD

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ABSTRACT

The prone of wood to biological damage during use in service necessitate its preservation using environmental friendly preservative. This study therefore investigates *Cassia fistula* seed oil (CFSO) against fungi attack on *Ficus mucuso* wood. The CFSO was obtained using soxhlet apparatus while ethanol was used as solvent and phytochemical screening of CFSO was carried out qualitatively. Five different concentration levels (0, 25, 50, 75, and 100 %) were prepared and used on *F. mucuso* wood (20 X 20 X60 mm) while total immersion was adopted for the treatment method. Retention of CFSO was determined for each of the concentration levels and the resistance of the wood against fungal attack (*Pleurotus florida Pleurotus saju caju*) was evaluated using weight loss. Data was analysed using analysis of variance at $a_{0.05}$. Analysis of the CFSO by phytochemical screening indicates the presence of tannin, flavonoids, terpenoids phenols. The preservative absorption rate of CFSO observed in this study decreases as the concentration increases. The retention of the CFSO by the wood ranged between 9.47 to 119.58 kgm^3 and weight loss ranged between 12.46 to 43.59 % and 12.85 to 44.56 for *Pleurotus florida Pleurotus saju caju* respectively. The maximum wood protection against fungi was obtained at 50 %. However, all the treatments proved to be effective over the untreated wood of *F. mucuso* wood as CFSO significantly contained antifungal properties.

Keywords: phytochemical, seed oil, preservative, fungi

Introduction

Wood as a hard fibrous material is prone to biological damage caused by mold, insect, such a beetles and termites and fungi, thus it is generally accepted that seasoning and preservative of wood and wood products is required in order to increase the longevity of wood in service against biological damage (Hickey and King, 2001; Byre, 2008). Fungal decay is one kind of the most serious microbiological deterioration because the strength of wood decreases rapidly (Howell *et al.*, 2011; Ayrilmisa *et al.*, 2015; Karim *et al.*, 2016). Reported by Ejechi *et al.*, (1996) and Irbe *et al.*, (2011) explained that the fungal decay can be classified into three categories: brown-rot,

white-rot, and soft-rot based on degradation mode in wood cell wall. The Brown-rot and white-rot are severer compared with softrot considering the decay time, extent, and resulting damages to wood. In brown-rot, the celluloses and hemicelluloses of wood are broken down, while the lignin remained (Kin *et al.*, 1996). Therefore, the decayed wood cracks into cubes and finally crumbles into powder (Babaee *et al.*, 2015). In case of ther white-rot, the fungus can degrade lignin as well as cellulose and hemicelluloses. However, lignin is degraded earlier in the decay process than cellulose and hemicelluloses (Schwarze and Engels, 1998).

However, changes ranging from simple discoloration to alterations of living trees, logs and wood products rendered wood completely useless in appearance, structure and chemical composition Ajala *et al*, 2021). Zabel and Morrell (1992) opined that degradation by fungi could cause losses as much as 15 to 25% of marketable wood volume in standing trees and 10 to 15%in wood products during storage and conversion. Akinyemi *et al.*, (2004) reported that thousands of cubic metres of wood are lost annually in Nigeria due to devastating effect of wood-rotting basidiomycetes and other insect infestations of logs and lumber products. Research has shown that toxic chemical preservative of wood against fungal decay posed a hazardous injury to human health and environment. This however has necessitated the development of eco-friendly preservative that is of biological origin. Plant extract (leaf, stem, root and seed) are safe, non-harmful to man and environment and still effective against pathogens (Arango *et al.*, 2005).

In recent time, it has been reported by (Amienyo and Ataga, 2007; Osman *et al.*, 2007; Kirker *et al.*, 2013, Tascioglu *et al.*, 2013; Adegoke *et al.*, 2015, Brocco *et al.*, 2017; Adebawo and Adegoke, 2019; Adegoke *et al.*, 2020; Okanlawon *et al.*, 2020) that plant has greater potentials in the use of plant and oil extractives as natural preservatives as many components of their extracts are very toxic to organisms imparting decay resistance to wood

Beside, *Ficus mucuso* wood is susceptible to rapid deterioration of fungi (Lamb and Ntima, 2000). Since *Cassia fistula* seed oil has been reported to possess anti-fungi and anti-termite (Awal *et al*, 2010) it is therefore, important to investigate the potential of *Cassia fistula* seed oil as preservative against fungi attack on *Ficus mucuso* wood.

Materials and Methods

Seed Collection, Preparation and Extraction of Oil

Cassia fistula seeds were collected within the Federal College of Forestry, Ibadan, Nigeria, It was de-shelled to retrieved the seeds from the pod, sun-dried to remove moisture, and blended with laboratory blender to fine powder. The fine powder was subjected to solvent extraction.

Two hundred (200 ml) of ethanol was poured into a round bottom flask. In the center of the extractor, 30 g of sample was placed in the thimble and heated at 65^{o}C. The vapor rises through the vertical tube into the condenser at the top while solvent was boiling. The liquid condensate drips into the thimble in the center, which contains the solid sample to be extracted. The extract seeps through the pores of the thimble and fills the siphon tube, where it flows back down into the round bottom flask. This was allowed to continue for 3 h. The thimble was then removed from the siphon tube, allowed to cool and after evaporation of the ethanol the oil left was weighed to determine the amount of oil extracted.

Phytochemical Screening of *Cassia fistula* seed Oil (CFSO)

The methanol crude extract was used for the phytochemicals, by dissolving 5 g of crude oil in 40 ml of distilled water. The presence of Alkaloids, Saponins, Tannins, Phenol, Flavonoids, Terpenoids, and Steroids were determined by using standard analytical procedures as described by Sofowora, (1993) and Trease and Evans, (2002).

Preparation and Treatment of Wood Test Block

The wood samples of *Fiscus mucuso* were obtained from the Bodija plank market in Ibadan, Nigeria and brought to the wood Workshop of Department of Wood and Paper Technology proper identification. The wood samples were dimensioned into 20 x 20 x 60 mm (radial x tangential x longitudinal directions). Each concentration level had twenty (20) replicates of the wood samples and they were properly labeled, weighed, and dried at a temperature of 103±2^{o}C for 24 h in an oven until excess moisture content was removed.

Dilution of the CFSO

Four (4) different concentration level of CFSO using volume by volume method in which kerosene was used as the diluents. 20 ml of CFSO and 80 ml of kerosene equal 20 % concentration level hence 0, 25, 50, 75, and 100 % while untreated samples (without immersion) served as the control (Adebawo *et al.*, 2018).

0 % = 0ml of CFSO with 100% of Kerosene.
25 % = 25 ml of CFSO with 75% of Kerosene.
50 % = 50 ml of CFS0 with 50% of Kerosene.
75 % = 75 ml of CFSO with 25% of Kerosene.
100 % = 100 ml of CFSO with 0% of Kerosene
Control = Untreated samples (Control)

Each of the diluted concentration levels has twenty (20) replicates of wood samples and total immersion was adopted for wood treatment with CFSO for 24 h after which , they were removed,

conditioned and the final weight was recorded.

Absorption Rate

The rate of absorption of wood samples when immersed in CFSO was determined using equation 1:

$$\text{Retention } (\text{kg/m}^3) = \frac{\text{G X C X 10}}{\text{V}} \qquad (1)$$

Where;

G = (T_2 –T_1) - amount of treated solution absorbed by the wood specimen (g).

T_1 = initial weight of the conditioned wood specimen before impregnation (g).

T_2 = initial weight of the conditioned wood specimen after impregnation (g).

C = grams of preservative in 100 g of solution.

V = volume of wood specimen (cm).

Preparation of Fungal Culture Medium and Innoculation

The inoculums of white rot fungi (*Pleurotus florida*) and brown rot fungi (*Pleutorus saju caju)* were cultured using Potato Dextrose Agar as a culturing medium. The control wood samples, and the treated wood samples at each concentration level of CFSO were placed in different McCartney bottles containing the actively growing test fungi (Adebawo and Adegoke, 2019). All *F. mucuso* wood was exposed to the fungi at room temperature (27±2^{o}C) for 14 weeks. At the end of incubation period, the blocks were removed from the culture bottles and the adhering mycelia were carefully cleaned off the wood surface.

Resistance to Fungi Attack by Weight Loss

The *F. mucuso* wood after exposure to fungal attack was oven-dried at 103^{o}C for 24 h and then re-weighed to determine the weight loss. The percentage weight loss was estimated using equation 2:

$$Weight\ Loss = 100\ \frac{W_{be} - W_{ae}}{W_{be}} \qquad (2)$$

Where;

W_{be} = Wight (g) of treated wood before exposure to fungi.

W_{ae} = Weight (g) of treated wood after exposure and final conditioning.

Data Analysis

The 2 *x* 6 factorial experiment in complete randomized design was adopted in which Factor 2 represent Two (2) fungi – *Pleurotus florida* and *Pleutorus saju caju;* Factor Six represent (6) diluted concentration level - 0, 25, 50, 75, 100 % and untreated samples which served as control. Statistical analyses were performed using Analysis of variance (ANOVA), and the significant difference between means was determined by Duncan's multiple range test. Differences at $P < 0.05$ were considered statistically.

Results and Discussion

Phytochemical Screening of *Cassia fistula* seed Oil (CFSO):

The result for the phytochemical screening showed that the CFSO contain some phytochemical compounds possessing good antimicrobial properties (Table 1). Analysis of the CFSO indicated the presence of tannin, flavonoids, terpenoids phenols, while saponin, steroid and alkaloids are absent. The biological activity of *C. Fistula* extract from bark, leaf, flower and seed has been reported by many scholars (Bahorun *et al.*, 2005; Nagpal *et al.*, 2011; Usha and Bopaiah, 2012; Irshada *et al.*, 2013, Ali, 2014; Abah *et al.*, 2017; Ayesha *et al.*, 2020). Nevertheless, *C. fistula* has been extensively used in various parts of the world against a wide range of ailments, likewise the synergistic action of its metabolite production being most probably responsible for the plant's beneficial effects (Bahorun *et al.*, 2005).

Classes of phytochemicals constituents present in the CFSO

Phytochemical compound	*Cassia fistula* seed oil
Tannin	+
Terpenoid	+
Alkaloid	-
Steroid	-
Saponin	-
Flavonoid	+
Phenol	+

+ = present; - = absent

CFSO absorption by *Ficus mucuso* wood

The Figure 1 shows the results of preservative absorption rate of *F. mucuso* wood species. The absorption rate of CFSO by *F. mucuso* wood species at different concentration level ranged between 9.47 and 119.58 kg/m^3 with 0 and 100 % concentration level showing the highest and lowest absorption rate respectively. Meanwhile, significant difference (p=0.05) existed in the absorption of the CFSO by the *F. Mucuso* and observed was a decreasing trend in the rate of absorption of CFSO by the *F. mucuso* wood from 100 to 0 %, that is, absorption increase with decrease in concentration level (Fig. 1). The 0 % higher absorption rate has been explained by Adenaiya *et al.*, (2016), Adebawo and Adegoke, (2019) due to easy penetration of the solvent into the cell wall.

In addition, the increase in concentration of CFSO as the absorption rate decreases signifies that more CFSO were retained in the cell wall of F. *Mucuso.* This further corroborated by Naveri *et al.*, 2017, Adebawo and Adegoke, (2019) that

absorption/retention depends on the concentration of active substance in solution and more also the penetration ability, viscosity of preservative and wood chemical composition (Onilude *et al.*, 2011 and Adegoke *et al.*, 2020).

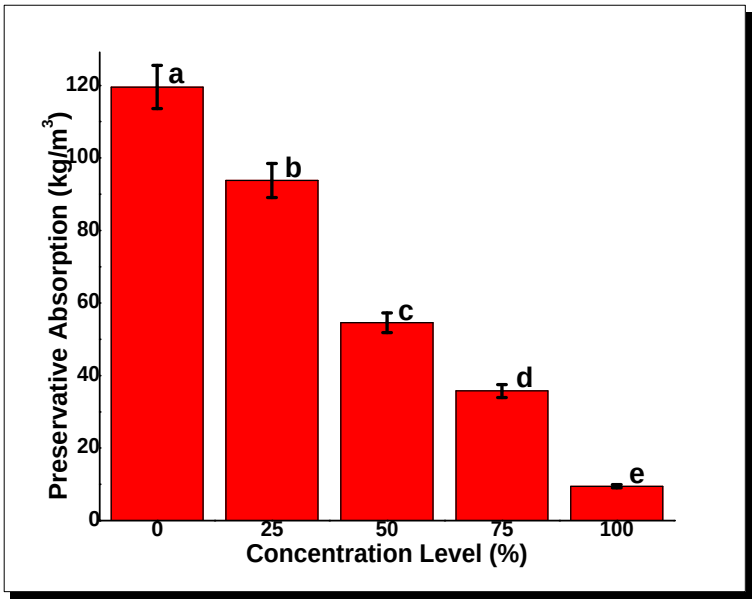

Figure 1: Effect of concentration level on absorption rate of *F.mucuso wood*

Mean values with the same alphabet in each bar are not significantly different (p=0.05) using Duncan multiple range test.

Resistance to P*leurotus florida* and *Plerotus saju caju* attack by Weight Loss

The weight loss of *F. mucuso* to white and brown rot fungi (P*leurotus florida* and *Plerotus saju caju)* attack further explain the effect of CFSO on the sampled wood. Meanwhile, the mean weight loss due to *white and brown rot fungi* attack ranged from 5.70 – 43.59 and 7.63 – 44.86 % respectively while the highest mean weight loss due to fungi attack was recorded at untreated wood of *F. mucuso* and 50 % concentration level.

The result of the weight loss of the untreated samples indicated that the two fungi (white and brown rot) significantly attack the wood while treated wood of *F. mucuso* resist the attack. It was obviously observed from the results (Fig. 1) 25 and 50 % concentration level of CFSO treated *F. mucuso* were highly resistance to the fungi attack while 100, 75, and 0 % were resistance to the fungal attack. However, the untreated wood was moderately

resistance to the fungal attack. The figure 2 further explained the effect of concentration level (25, 50, 75, and 100 %) on percentage weight loss *F. mucuso* after the fungi attack which shows significant (p=0.05) differences.

Table 1: Analysis of variance for weight loss of *F. mucuso* wood to fungi attack

Source of variation	df	Sum of Squares	Mean Square	F	Sig.
Fungi (FG)	1	302.94	302.94	7390.11*	0.00
Concentration Level (CL)	5	41831.31	8366.26	204091.63*	0.00
FG * CL	5	192.53	38.51	939.32*	0.00
Error	228	9.35	0.04		
Total	239	42336.12			

* - Significant (p=0.05)

Cassia fistula is found biologically potential by having many pharmacological activities, that is, antifungal (Irshada *et al.,* 2013), antibacterial (Ali *et al.,* 2003), antiparasitic (Sartorelli *et al.,* 2009), antioxidant (Tzekiat and Chiange, 2013) antifertility (Yadav and Jain, 2009), anti-inflammatory (Anwikar and Bhitre, 2010) among others. However, the extent of resistance of *F. mucuso* wood to fungi attack could be proved significantly that antifungal properties posses by CFSO could be responsible (Duraipandiyan and Ignacimuthu, 2007; Panda *et al.,* 2010; Ayesha *et al.,* 2020), More importantly, the availability of phytochemicals constituents such as alkaloids, flavonoids, tannins and saponins in CFSO could promote antifungal activity. Generally, the seed extract that are originated from plant do offer more occurring chemicals that are very toxic and active against biodegradation agents.

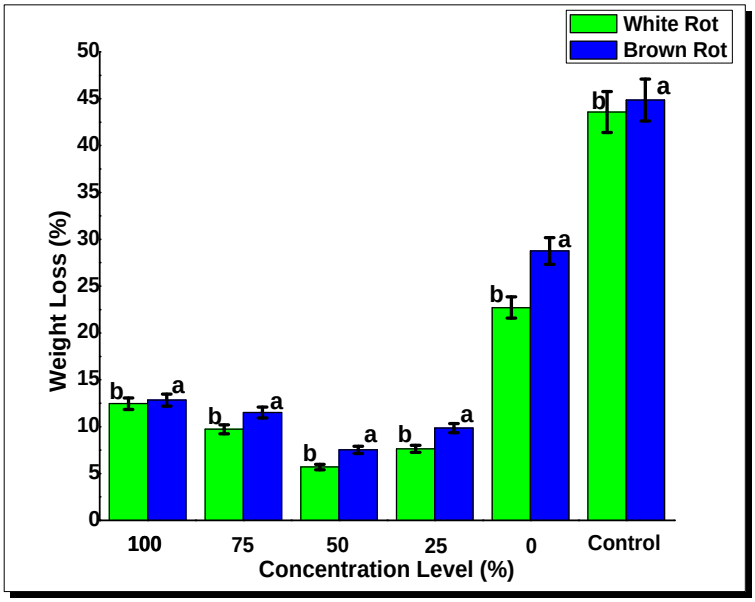

Figure 2: Weight loss of *F. mucuso* wood to fungi attack

Mean values with the same alphabet in each bar are not significantly different (p=0.05) using Duncan multiple range test.

Conclusion

Development of more environmentally acceptable preservatives which is a priority in the wood preservative industry today has opened the door to plant based wood preservatives. *Cassia fistula* seed oil was found to have toxicity towards fungi. *Cassia fistula* seed oil in resisting the decay of both white and brown rot fungi. It was proved further that seed oil extract of *Cassia fistula* could used as biological preservative.

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