

Doi: HTTPS://DOI.ORG/10.23910/IJEP/2018.5.1.0229

Biopreservation of *Pinus roxburghii* and *Bombax ceiba* Using Aqueous Extract of *Acorus calamus*

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Article History

Article ID: IJEP0229

Received in 27th October, 2017

Received in revised form 3rd February, 2018 Accepted in final form 13th February, 2018

Abstract

Due to biological origin, wood is one of the most complex constructions materials and has affinity for moisture which can lead to biological deterioration. Biological damage to wood and wood products is mainly caused by the mould, stain, decay fungi, and insects such as beetles and termites. Therefore, wood preservation is required after harvesting to reduce attack by wood deteriorating agents. Development of low environmental impact technologies for the elimination of biological damage is one of the vital goals of wood protection industry. In the present study, effect of Acorus calamus extract on fungal growth was tested on wooden samples of Pinus roxburghii and Bombax ceiba at different concentrations.

Keywords: Acorus calamus, biopreservation, Bombax ceiba, Pinus roxburghii

1. Introduction

As industrial technology advanced, wood was used more frequently in exterior structural applications. Wood species that did not possess inherent decay resistance properties failed in service due to biological attack, creating a need for preservative-treated wood (Hunt and Garratt, 1967). Wood preservation is an art of protection of wood against any factor whatsoever that may damage and ultimately destroy it. In practical sense it refers to improvement of wood natural durability by treatment with chemicals. It is an outstanding practice to improve the serviceability of wood by chemical treatment under condition that favours early deterioration by decay.

One of the approaches to enhance the durability of wood for its efficient utilization is through preservation (Wegner et al., 2010). Chromium copper arsenate (CCA) was the important wood treatment that had been widely used since 1940 to prevent the decomposition of wooden timbers. This treatment consists of chromium and arsenic salts that inhibit insect attack and microbial decay of the wood so that the lifetime of the timber is increased. However, CCA has been banned or limited for some application in some European countries, U.S. and Japan (Kartal et al., 2004).

In 1928, Iwanowski and Turski discovered the use of di, tri and polychlorinated phenols for wood preserving purposes. In 1929, in the United States, L.P. Curtin patented

the use of chlorine derivatives of coal-tar acids of higher molecular weight than the cresols expressing a preference for chlorinated phenols. Three commercially available nonarsenical systems appear poised to replace CCA for residential applications: alkaline copper quat (ACQ), amine copper azole (CA), and copper bis-(N-cyclohexyldiazeniumdioxy) (Cu-HDO or copper xyligen). Many toxic chemicals were used in wood preservation and wood processing industries. These chemicals were harmful for many species of our biodiversity including plants, beneficial microbes, nematodes, invertebrates, etc. So, scientists are working on biological preservative having no harmful effect on biodiversity. Wood preservatives can extend the life of wood and reduce the need for forest resources, but proper use is important. Some preservatives can slowly leach into the surrounding soil or water. Sometimes, touching the wood can leave residue on exposed skin. There is growing concern about the environmental impacts and increasing difficulty to dispose preservative treated wood products at the end of their service life.

2. Materials and Methods

2.1. Preparation of herbal extract for wood samples treatment In this study, extract of Acorus calamus was prepared with solvent (methanol, benzene, alcohol etc.) in soxhlet apparatus on a boiling water bath at 60-70 °C. Thereafter, 2.5 L stock solution of 10% concentration was prepared with 5% methanol. From this stock solution, solutions of different concentrations were made i.e., 0.25%, 0.50%, 1.0%, 1.50% and 2.0%. Wood samples of both hardwood (Bombax ceiba) and softwood (Pinus roxburghii) species were treated in the above prepared concentrations for 72 hours. The samples meant for control were dipped in distilled water. After dipping treatment, wood samples were dried at 105±2 °C in oven up to constant weight.

2.2. Inoculation and colonization of fungus on treated wood samples

The pure culture of any fungus was revived and transferred to the agar slant which was purified by hyphal tip technique. For the growth of fungus, malt agar solid media was prepared directly by adding 2% of malt and 2% of agar in 1000 ml distilled water. About 100 ml of the medium was poured in glass jars of 500 ml and autoclaved at 15 lb pressure square inch-1 (psi) for 20 minutes at 121 °C. The media was allowed to solidify. The glass jars were then inoculated with fungal culture bits (5 mm) taken from 10 days old vigorously growing culture followed by incubation at 25±1 °C. Wood samples were sterilized by keeping them under UV light in Laminar Air Flow (LAF) for 20 minutes prior to fungus decay tests. All instruments used under LAF were sterilized with alcohol then with spirit lamp to avoid contamination and for better results. Inspection and evaluation of fungus colonization on wood was made by visual assessments after one month (Chauhan, 2013).

2.3. Observations recorded

The following observations were recorded to study the effect of extract on growth of fungus:

2.3.1. Fungus growth index

The following scale was used for recording the disease severity:

Numerical	% fungus	Description
ratings	colonization on surface	
0	0	No surface coverage
1	0-25	Very less surface coverage
2	26-50	Less surface coverage
3	51-75	Moderate surface coverage
4	>76	Large surface coverage

Disease severity (%) on surface was calculated according to McKinney (1923).

Fungus growth index (%)=
$$\frac{\text{Sum of all disease rating}}{\text{Total no. of rating}} \times 100$$

Maximum disease grade

2.3.2. Fungus growth inhibition (I)

Fungus growth inhibition was calculated by the formula give by Vincent (1947):

Where,

I= Per cent growth inhibition

C= Per cent fungus colonization in control

T = Per cent fungus colonization in treated wood

3. Results and Discussion

Wood samples treated with Acorus calamus L. extract were able to inhibit the fungal growth significantly. Maximum fungal inhibition (75%) was observed in Pinus roxburghii at 2% concentration, followed by Bombax ceiba (66.67%). The next best treatment concentration was 1.50% providing 50% and 58.33% inhibition of fungus on Pinus roxburghii and Bombax ceiba, respectively. The effect of extract was more on softwood as compare to hardwood species. The least effective concentration was 0.25% providing only 16.67 and 8.33% growth inhibition of test fungus (Table 1).

These results are in conformity with Astiti (1998) who found that the water extract of teak leaves inhibited the growth of Monilia sp., Alternaria cajani and Helminthosporium sp. Similarly, Bajwa et al. (2003) evaluated the potential of aqueous extracts of allelopathic weed Parthenium hysterophorus against three pathogenic fungi viz. Drechslera tetramera, Aspergillus niger and Phoma glomerata. Dhyani et al. (2005) used acetone, ethyl alcohol and methyl alcohol extract of neem leaves to inhibit the growth of Poria menticola (brown rot) and Polyporus versicolor (white rot). Sethy et al. (2005) found that alcoholic extract from the heartwood of Dalbergia latifolia have the potentiality to protect wood decay fungi. Reddy et al. (2009) tested the antifungal activity of phytoextracts and plant oils of several plant species such as Azadirachta indica, Allium cepa, Allium sativum. Tegetes erecta, Aloe barbadensis, Eucalyptus globulus against the growth of Cercospora moricola. Salih et al. (2010) used extract of Ageratum conyzoides, Ficus retusa, Lavandula pubescens, Lawsonia alba against fungus. Devi (2013); Chauhan (2013) observed that at 2% concentrations of Ageratum conyzoides L. and Melia azedarach L. extract have minimum fungus colonization in wood and proved to be a good biopreservative.

Table 1: Fungus growth index and growth inhibition of taken wood samples using extract of Acorus calamus

Treatment	Fungus growth index (%)		Fungus growth inhibition (%)	
	Pinus	Bombax	Pinus	Bom-
	rox-	ceiba	rox-	bax
	burghii		burghii	ceiba
A. calamus (0.25%)	83.33	91.67	16.67	8.33
A. calamus (0.50%)	75.00	83.33	25.00	16.67
A. calamus (1.00%)	58.33	75.00	41.67	25.00
A. calamus (1.50%)	50.00	41.67	50.00	58.33
A. calamus (2.00%)	25.00	33.33	75.00	66.67
Control (Untreated)	100.00	100.00	0.00	0.00

4. Conclusion

The wood industry is undergoing a major transition as CCA is replaced in most residential applications. The use of newer process technologies holds promise for new wood preservatives, and breaks ground for modern advances in commercial production plants, and innovation in research opportunities. There is an increasing need to educate the consumer regarding new wood-treating chemistries and new products. Also, there is a need for detail and advance study on wood biopreservation.

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