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Development of eco-friendly antimicrobial textile finish on Bamboo Union Fabrics with Saraca asoca Bark and its Phytochemical assay

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ABSTRACT

There is a growing demand for medical textiles globally, which has created a great need for antimicrobial textile finish. The antimicrobial finish is given to fabrics to protect them from microbial damage and also to provide protection to the wearer. The current study is conducted to develop an eco-friendly antimicrobial finish with Saraca asoca (Ashoka tree) bark. Two different union fabrics Bamboo/ Linen and Bamboo/ Cotton were woven and pretreated with citric acid as crosslinking agent and the bark extract was applied. The antimicrobial efficacy against gram positive (Bacillus subtills), gram negative (E. coli) bacteria and a fungus (Aspergillus niger) were tested. The durability of the finish was tested by conducting washing tests and subjecting to antimicrobial tests. The results showed that, 10% methanol extract of Ashoka exhibited good antimicrobial efficacy and resistance to washing up to 10 washes. The FTIR characterization and quantitative Phytochemical Analysis showed the presence of antioxidants, flavonoids, saponins, phenols and tannins, which may be the causative of antimicrobial activity of the bark. Though well diffusion test of extracts yielded very good ZOI, the treatment fabrics didn't exhibit good ZOI with disc diffusion, hence the parallel streak method was used. The current study contributes to the present knowledge of natural antimicrobial agents and the textile finish developed can be applied on medical textiles, surgical gowns and health care uniforms.

Keywords: *Eco-friendly, antibacterial, antimicrobial efficacy; phytochemical, bark extract, Zone of Inhibition, antimicrobial agents, textile finish*

Introduction

The antimicrobial finish inhibits the growth of bacteria and prevents decay and damage from perspiration. Textiles are the most suitable materials to impart antimicrobial properties. The availability of large surfaces and moisture retaining property of textile materials provide a nourishing environment for microbial growth, causing undesirable effects on the textile as well as the wearer [1]. The natural fibres provide favourable conditions for the growth of microbes [2]. Hence, textile finishes, which impart properties such as anti-fungal, anti-mildew, and anti-bacterial are given to medical textiles, bandages used for wound healing, and protective clothing, Synthetic antimicrobial agents are very effective in imparting protection against a wide range of microorganisms, but are non-biodegradable, bio accumulators, and may cause skin irritation, cancer, also destroy desirable microbes. Another drawback of chemical antimicrobial agents is most of the pathogens are developing resistance to them. Many studies are being carried out to develop eco-friendly antimicrobial finishes prepared from plant extracts, essential oils, and other natural products. The current study is conducted to develop eco-friendly, antimicrobial finish on union fabrics of natural fibres, with *Saraca asoca* (Ashoka) tree bark extract. The bark extract constituents were characterized with FTIR test, phytochemicals were quantified, assessment of the antimicrobial efficacy and durability of the finish were conducted.

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Materials and Methods

Materials

Three sustainable natural fibres viz., Bamboo, Linen and cotton unbleached yarns were selected for the study to apply the eco-friendly antimicrobial finish. Two Union fabrics, Viz., Bamboo 2/20s/ Linen 40Lea (UF1) and Bamboo2/20s/ Cotton 2/20s(UF2) were woven at Karnati Murali, master Weaver's Unit, Sattenapalli, Guntur District, Andhra Pradesh, on a handloom (frame loom). Tree bark of *Saraca asoca* (Ashoka) was selected as the source for antimicrobial textile finish after conducting a pilot project. Ashoka bark was procured from the local market. The bark of this tree is traditionally known to have antimicrobial, anticancer, antihemorrhagic, and anti-oxytocic activities**.**

Methods

The Union Fabrics, UF1 and UF2 after weaving, were given eco-friendly pre-treatment to remove impurities. The fabrics were treated with a neutral soap of 2g/ litre at 90°C for 30 minutes, with MLR of 1: 20, washed thoroughly and dried under sun. The Ashoka tree bark was pounded in a stone grinder to small pieces, and then made into powder in a mixer grinder, sieved several times to remove large particles, until smooth fine powder was obtained. Three methods of extraction, viz. Soxhlet extraction,

maceration and aqueous extraction were used to prepare the extracts. Three solvents, methanol, acetone, and distilled water were evaluated. Maceration was conducted separately with methanol and acetone solvents, for 24 hours continuously stirring at 550 RPM, with a magnetic stirrer and allowed to stand still for eight hours. The aqueous extraction was conducted at 99°C on a hotplate for 30 minutes and the solution was kept aside to cool down to room temperature. The supernatant liquid was filtered with filter paper.

The Ashoka bark extracts obtained were applied on the union fabrics using pad-dry-cure method. Then the fabric samples were evaluated for antibacterial efficacy against *E. coli* and *Bacillus subtilis*. Based on the antimicrobial test results and extensive literature review, an eco-friendly crosslinking agent, citric acid, was selected as the crosslinking agent. Three concentration s of citric acid, viz., 8%, 9% and 10% were evaluated and antibacterial tests were conducted to find out the best concertation. The soaking time for pretreatment of fabrics in citric acid solution was optimized. Three soaking times, 60 minutes, 75 minutes, and 90 minutes were evaluated and the best soaking time was selected based on antibacterial test result. Three Ashoka bark extracts made with three different solvents were made in different concentrations, 5%, 10% and 20%. The suitable concentration and solvent were selected, based on antibacterial test results. The duration of soaking the union fabric samples also influences the antimicrobial efficacy. Hence, 45 minutes, 60 minutes, 75 minutes, and 90 minutes durations of soaking were evaluated, optimal soaking time was selected based on antibacterial test results.

Antimicrobial tests

The AATCC test method 147-2004, parallel streak method was used to test the antibacterial efficacy of the control and treatment fabric samples of size 25mm X 50mm. The clear zone of Inhibition (ZOI in mm) was measured. These tests were conducted at the Dept. of Veterinary Microbiology, CVSc, PVNRTVU, Hyderabad. The antifungal tests against *Aspergillus niger* using diffusion plate method AATCC 30, were carried out. The percentage of area of discs covered with growth of *Aspergillus niger* were recorded. The assessment was done based on the standard observations. Different ratings were formulated based the extent of % fungus growth. The sample with "no growth (0%) was given a rating "4". Less than 10% growth was rated as "3", 10%-30% growth was given the rating "2", 60% to 90% growth (complete coverage) was rated "1" and 90%-100% growth was given a rating of "0".

Washing tests

The durability of antimicrobial finish was tested by washing tests, as per the Bureau of Indian Standards IS 687-1979, using a Launder-O-meter, at the Dept. of Apparel and Textiles, CCSc, PJTSAU, Hyderabad. The samples were evaluated after 1 wash, 5 washes, 10 washes and 15 washes.

Phytochemical Analysis of Ashoka Bark extract

The Fourier-transform infrared (FTIR) spectroscopy was conducted using the UV Double Beam Spectrophotometer at the center for Nano Science and Technology, JNTUH.The infrared spectrum of Ashoka bark extract (methanol 10%), and the peaks in the FTIR spectrum were analysed.Quantitative phytochemical analysis of Ashoka bark extract (methanol 10%) was conducted to detect the primary and secondary metabolites present in the bark extracts, which may contribute to the antimicrobial efficacy of the bark extract. The tests conducted were, the total Antioxidant Activity [3], total saponins [4], total flavonoids [4,5], total phenols [4] and total tannins [4], present in Ashoka bark extract.

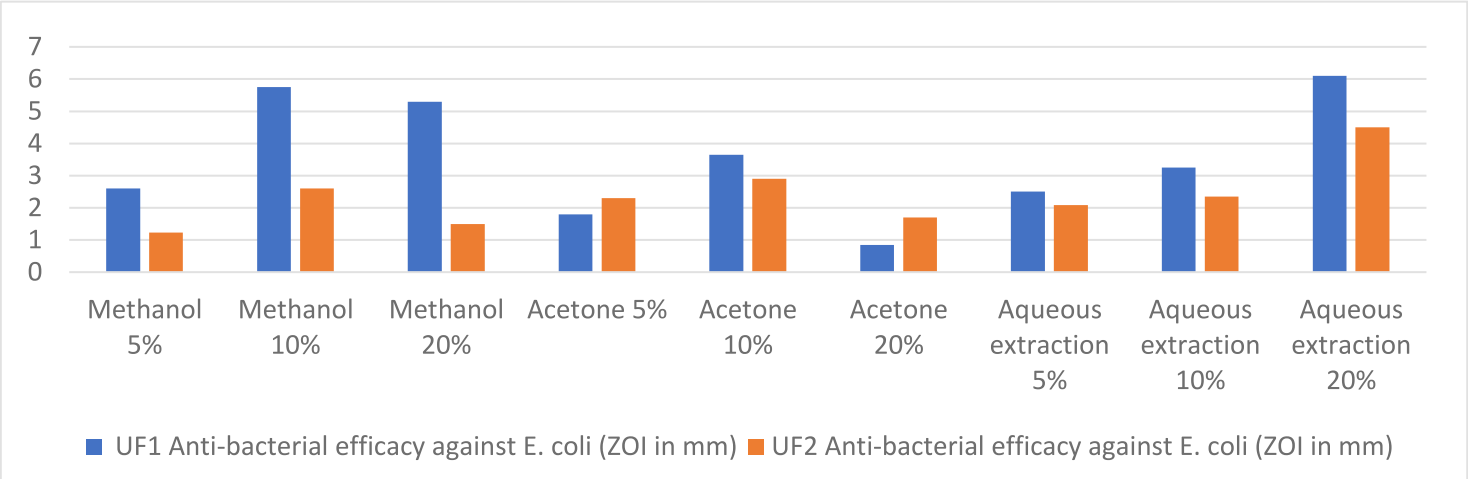
Results and Discussion

Application of Ashoka bark extract on union fabrics: Ccitric acid, an eco-friendly crosslinking agent was tested in three concentrations, viz., 8%, 9%, 10% The 9% citric acid showed best ZOI. Hence, 9% concentration of citric acid was selected for the study. The best duration of soaking the fabric samples in 9% citric acid solution was found to be 75 minutes based on antimicrobial test results. Three solvents (Methanol, Acetone and Distilled water) and three concentrations (5%, 10% and 20%) were used to prepare extracts and applied by pad-dry-cure method and evaluated for antibacterial efficacy. The optimum soaking time for soaking in Ashoka bark extract was found to be 75 minutes. The test results (mean ZOI in mm of five streaks) is given below.

Table 1. Antibacterial tests (parallel streak method) against E. coli

Extract sample	sample	*E. coli* (ZOI in mm)	sample	*E. coli* (ZOI in mm)
	UF1C	0	UF2 C	0
Methanol 5%	UF1T	2.6	UF2 T	1.23
Methanol 10%	UF1T	5.75	UF2 T	2.6
Methanol 20%	UF1T	5.3	UF2 T	1.5
Acetone 5%	UF1T	1.8	UF2 T	2.3
Acetone 10%	UF1T	3.65	UF2 T	2.9
Acetone 20%	UF1T	0.85	UF2 T	1.6
Aqueous 5%	UF1T	2.5	UF2 T	2.08
Aqueous10%	UF1T	3.25	UF2 T	2.35
Aqueous 20%	UF1T	4.1	UF2 T	2.4

Fig.1. Ashoka bark extract with different solvents and concentrations– antibacterial tests (parallel streak method) against E coli

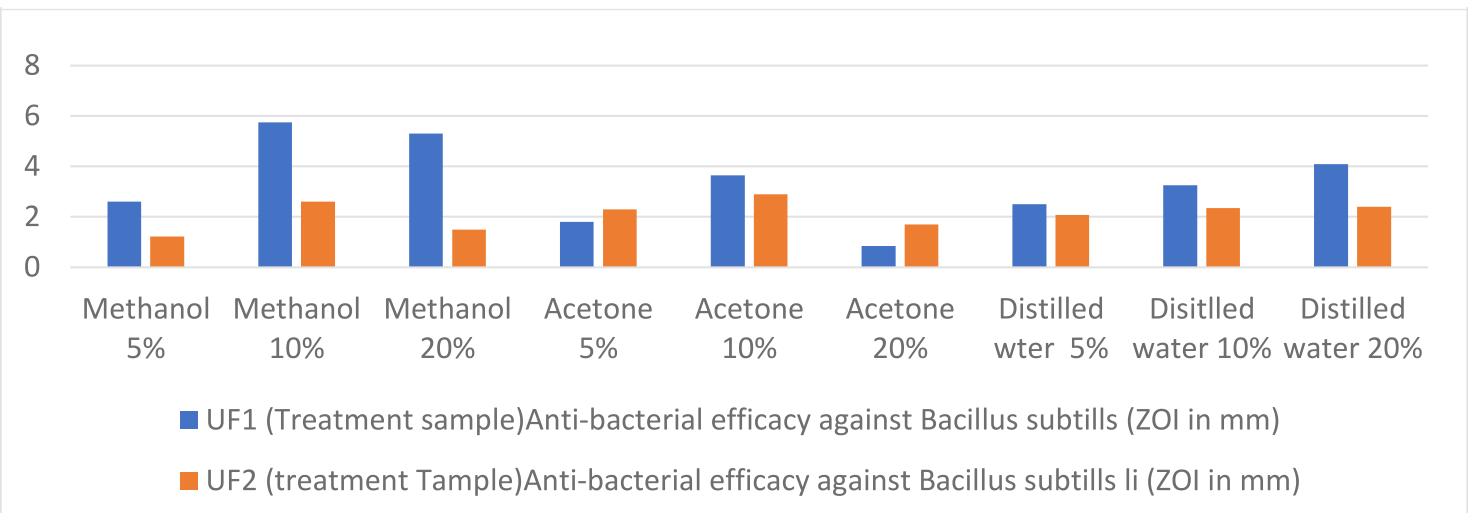


Overall analysis of all solvents and concentrations revealed that Methanol 10% solvent has given the best antibacterial efficacy against E. coli bacteria.

Table 2. Antibacterial tests (parallel streak method) against Bacillus Subtills

Extract sample	sample	*Bacillus subtills* (ZOI in mm)	sample	*Bacillus subtills* (ZOI in mm)
	UF1C	0	UF2 C	0
Methanol 5%	UF1T	1.75	UF2 T	6.25
Methanol 10%	UF1T	5.75	UF2 T	6.4
Methanol 20%	UF1T	1.45	UF2 T	3.75
Acetone 5%	UF1T	2.9	UF2 T	1.75
Acetone 10%	UF1T	2.1	UF2 T	1.3
Acetone 20%	UF1T	2.5	UF2 T	2.5
Aqueous5%	UF1T	1.25	UF2 T	2
Aqueous 10%	UF1T	3.75	UF2 T	1.25
Aqueous 20%	UF1T	4.1	UF2 T	2.4

Fig.2. Ashoka bark with different solvents and cocnentrations -antibacterial tests (parllel streak method) against Bacillus subtilis



The overall test results against Bacillus subtilis revealed that Methanol 10% solvent has given best antibacterial efficacy, when compared with other solvents and concentrations.

Washing Tests

Table 3. Antibacterial Test Results – Washing Test Samples

No. of washes	*E. coli* (ZOI in mm)				*Bacillus subtilis* (ZOI in mm)		
	UF1C	UF1T	UF2C	UF2T	UF2C	UF2T	UF2T
1 Wash	0	4	0	3	0	1	3
5 washes	0	3	0	5	0	1	3
10 washes	0	3	0	5	0	1	3
15 washes	0	2	0	1	0	2	2

Antifungal tests: The methanol 10% treated UF1 and UF2 fabrics were evaluated for antifungal efficacy against *Aspergillus niger*. The washing test samples, viz., 1 wash, 5 washes, 10 washes and 15 washes U1C, UF1T and UF2C, UF2T were tested for antifungal efficacy. The control samples have shown 90-100% growth. The treatment samples have exhibited excellent antifungal efficacy, with zero fungus growth.

Table 4. Antifungal Test Results – Washing Test Samples

No. of washes	*Aspergillus Niger*			
	UF1C	UF1T (ZOI in mm)	UF2C (ZOI in mm)	UF2T (ZOI in mm)
1 Wash	0	3	0	3
5 washes	0	3	0	3
10 washes	0	2	0	2
15 washes	0	1	0	1

The above results show that the treatment samples after one wash, five washes and fifteen washes exhibited good resistance to fungus growth when compared to control samples.

Fig. 3. Antimicrobial test samples

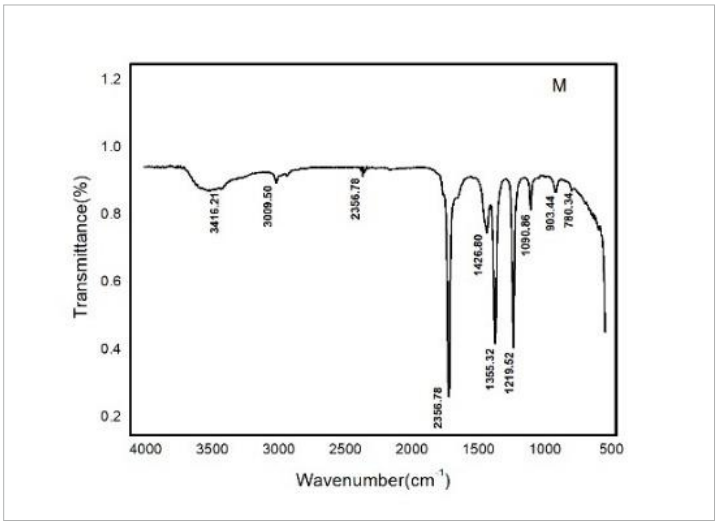
A. Ashoka M 10% UF1 Antibacterial test against Bacillus

B. Ashoka M 10% UF2 Antibacterial test against E. coli

B. Ashoka M 10% UF1 Antifungal test against Aspergillus niger

Phytochemical analysis: The FTIR spectrum of Ashoka bark extract: The image of the spectrum exhibits transmittance percentage on y-axis and on x-axis the wave number (cm^-1). The prominent peaks of the spectrum are marked with their corresponding wave number. The **3328.48 cm^-1** peak is associated with O-H stretching, indicating the presence of hydroxyl groups, which may be due to presence of alcohols, phenols and antioxidants. The **2942.40 cm^-1 and 2841.80 cm^-1** peaks are related to C-H stretching. The peak at 2942.40 cm^-1 is due to the asymmetric stretching of CH2 groups and the symmetric stretching of CH2 groups caused the peak at 2841.80 cm^-1. This indicates the presence of aliphatic chains of lipids, fatty acids and similar compounds in the extract. The **1700-1500 cm^-1** peak shows C=O stretch, around 1700 cm^-1 indicate ketones, aldehydes, or carboxylic acids. The **1500-1000 cm^-1** peak exhibits C-O stretching, C-N stretching, indicating presence of many functional groups [6, 7]. Presence of these functional groups indicates the possibility of the extract containing bioactive compounds like fatty acids, polyphenols, terpenoids and other secondary metabolites. The FTIR spectrum of Ashoka bark extract showed peaks at 3416.21 (O-H), 3009.50 (C-H), 2356.78 (C-N), 1426.80 ©-O, C-N), 1355.32 (C-O, C-N) and 1219.52 (C-O, C-N).

Fig. 4. FTIR Analysis of Ashoka bark extract – FTIR Spectrum Graph



Quantitative Phytochemical analysis of Ashoka Bark Extract: Five major bioactive compounds were detected and quantified using phytochemical analysis test procedures. The results indicate that, Ashoka Bark Extract has the bioactive compounds viz., antioxidants, saponins, flavonoids, phenols and tannins in significant quantities, which may be attributed to the antimicrobial properties of the extract.

Table 5. Quantitative Phytochemical Analysis of Ashoka Bark extract

Sample	Name of the test	Test results (Quantity)
Ashoka Bark Extract	Total Antioxidant Activity	413.79MUGM/100g
	Saponins	13.00%
	Flavonoids	2710.00 Mg of QE/100gm
	Total Phenols	225.55 Mg of GAE/100gm
	Tannins	3.02 Mg of TAE/100gm

Conclusion

All the control samples exhibited zero to very meagre results, implying that the Union fabrics of Bamboo do not possess any inherent antimicrobial properties. The antimicrobial tests exhibited good antimicrobial efficacy. The washing test results showed good durability of the finish, up to ten washes. Hence, it is concluded that Ashoka bark extract can be safely used for textile finishing. The current antimicrobial textile finish is applied on two union fabrics only. However, the textile finish can be applied on other textile fibres as well. Other protective and functional properties of the bark extract can also be assessed.

Author's contribution

The first author has conducted research, compiled the data, and prepared the article under guidance of the co-authors.

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Conflict of interest

The authors declare that there is no conflict of interest.

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