

## Winner of DR Balyal Memorial Best Paper Award 2019

### Utilization of plant extract as antimicrobial finish for healthcare textiles

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#### ABSTRACT

Textile materials are good media for the growth of the microorganisms. The consumers these days are increasingly being aware of the hygienic lifestyle and there is a necessity of development of a wide range of textile products finished with antimicrobial properties. Thus an innovative approach was made to utilize the plant sources as eco-friendly and renewable source for production of microbial resistant fabric. The developed environment-friendly finish prevents the growth of bacteria. It also prevents garments from unpleasant odour. For this the chemical nature of Mousambi and Harshingar leaf extracts was determined and extraction of active substances from the leaves was done using ethanol, methanol and aqueous medium. The herbal extracts were applied on 100 per cent cotton fabric with and without the use of cross linking agent. Eleven concentrations of both the plant extracts were used to study the reduction in bacterial colonies on the fabric. The antimicrobial activity of the finished fabric was assessed using quantitative and qualitative methods against bacteria and fungi that normally exist in the textile environment. The finished cotton fabrics showed the maximum bacterial reduction percentage of 74.7 for Harshingar against Gram positive and 71.2 for Mousambi against Gram negative bacteria. With the increase in concentration of the plant extract a decrease in turbidity was noticed.

**Keywords:** Textile; fabric; plant extracts; bacteria; fungi; Mousambi; Harshingar

#### INTRODUCTION

In the recent scenario there has been an upsurge interest in apparel technology all over the world for much demanding functionality of the products like wrinkle resistance, water repelling, fade resistance and resistance to microbial invasion. Among these development of antimicrobial textile finish is highly indispensable and relevant since garments are in direct contact with the human body (Sathianarayanan et al 2010).

An antimicrobial source is an agent that kills microorganisms or inhibits their growth. Antimicrobial medicines can be grouped according to the microorganisms they act primarily against. Antibacterial, commonly known as antibiotics are used against bacteria and antifungal are used against fungi (Cowan 1999). Plant products are the principle source of pharmaceutical agents used in traditional medicine (Khond et al 2009, Bandow et al 2003).

Phytochemicals, the naturally occurring compounds in plants are responsible for colour and other organoleptic properties. Plants produce various compounds which are responsible for protection against pathogens. Some plants may contain compounds of potential medical use (Yadav et al 2012, Ahmad and Beg 2007). Growing interest in substitution of synthetic antimicrobial agents by natural ones has fostered research on vegetable sources. Screening of plant materials in order to identify new compounds or test natural chemicals already known for important activities have not been discovered so far (Ait Ouazzou et al 2011). Essential oils (EOs) extracted from different plant genera are in many cases biologically active. These aromatic compounds are relatively inexpensive and there is abundant raw material with practical applications for different industries. Textile can be a potential microorganism's propagation site because it comes in contact with human skin and endows optimal conditions by combination of nutrient sources,

temperature and moisture providing conducive environment for microorganisms' growth. Thus the development of antibacterial textile is crucial to inhibit the growth of microorganisms for protection of health and safety of wearer.

Cotton fabrics provide ideal environment for microbial growth. Apparel researchers have several challenges due to increasing global demand in textile. Therefore textile finishes with added value particularly for medical clothes are greatly appreciated and there is an increasing demand for them on global scale. The consumers are aware of hygienic lifestyle and there is a necessity of textile product with antimicrobial properties. Several antimicrobial agents viz triclosan, quaternary ammonium compounds and recent nano-silver are available for textile finishing (Thilagavathi and Kannaian 2010). However due to their cost and synthetic nature which create environmental problems, plant sources in textile finishing are gaining significant momentum. The effect of various plant extracts on bacteria and fungi has been studied by a number of researchers (Lioliou et al 2007, Pereira et al 2011, Jasso de Rodriguez et al 2007). Although certain natural antibacterial agents are available, at present only few studies have been explored for their antibacterial activity on textile materials and progressive and consolidated data on antimicrobial finished products of textiles are also required. In the present study two plant sources viz Harshingar and Mousambi were selected.

## MATERIAL and METHODS

The leaves of sweet orange or Mousambi, *Citrus sinensis* (Rutaceae) and night jasmine or Harshingar (*Nyctanthes arbor-tristis*) were selected on the basis of their properties for the functional finish. The plant leaves were collected, washed in reverse osmosis (RO) water, dried in hot air oven at  $40 \pm 1^\circ\text{C}$ , powdered in a blender and refrigerated. The plant source powder was soaked in distilled water, ethyl alcohol and methyl alcohol separately for 24 hours. The supernatants were centrifuged and filtered. This extract was applied on the cotton fabric. Two methods were used for finishing the fabric viz direct and pad-dry-cure method.

**Direct method:** The pre-treated cotton fabric was immersed in antimicrobial stock solution containing cross linking agent for one hour and was shade-dried.

**Pad-dry-cure method:** The fabric was immersed in the antimicrobial stock solution containing cross linking agent for ten minutes and passed through pneumatic padding mangle at a speed of 3 m/min with a pressure of  $1 \text{ kg/cm}^2$  to remove excess solution ensuring the wet pick up of 70 per cent (Selvi and Rajendran 2014). Later the fabric was shade-dried and cured for 3 minutes at  $140^\circ\text{C}$ .

Antimicrobial activity of the plant source and the finished textile fabric was done. Quantitative assessment of antimicrobial activity exhibited on cotton fabric against both the microorganisms was carried out by AATCC Test 100-2004 (<https://wenku.baidu.com/view/a4a461c62cc58bd63186bdd5.html>). To evaluate the antimicrobial activities of the treated fabrics the reduction in the number of bacterial colonies formed with respect to the untreated control after incubation ( $37 \pm 1^\circ\text{C}$  for 24 hours) was determined.

The percentage reduction was calculated using the following equation:

$$\text{Reduction rate, } R (\%) = (B - A)/B \times 100$$

where R: Reduction in bacterial count, A: Number of bacterial colonies recovered from the inoculated treated test specimen swatches in the jar incubated for 24 hours contact period, B: Number of bacterial colonies recovered from the inoculated untreated control test specimen swatches in the jar immediately after inoculation (at 0 contact time)

**Minimum inhibitory concentration (MIC):** A minimum inhibitory concentration is considered the good standard for determining the susceptibility of organisms to antimicrobials. The MIC is defined as the lowest concentration of an agent that will inhibit the visible growth of an organism after overnight incubation (Tripathi 2013).

**Stock solution:** Stock solution was prepared using 100 ml of antimicrobial solution.

**Culture:** For 24 hours subcultures of the test organisms *Staphylococcus aureus* (Gram positive) and *Escherichia coli* (Gram negative) were serially diluted and  $1 \times 10^{-5}$  dilution was selected for MIC study.

**Preparation of tubes for MIC:** A separate test tube containing nutrient broth alone was kept as control. All the test tubes and control samples were incubated at  $37^\circ\text{C}$  for 24 hours. After the period of incubation the

MIC was determined based on the turbidity that occurred.

## RESULTS and DISCUSSION

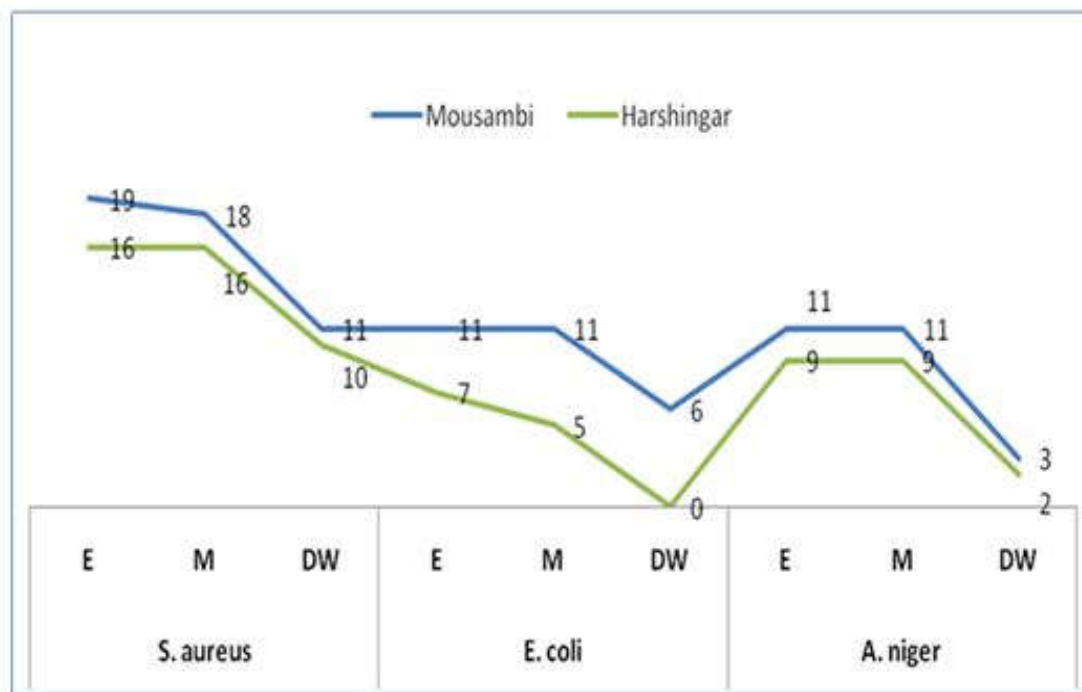
The antimicrobial activity of the extracts of plant sources was observed and zone of inhibition was noted. Lack of zone of inhibition does not necessarily mean an absence of activity. A zone is generally shown by antimicrobial agents that are leaching type ie they leach out of the fabric and kill the microbes present on as well as around the treated fabric. The results of antimicrobial activity of the herbal plant extracts are presented as zone of inhibition in Fig 1. The Gram positive bacteria chosen for the study were *Staphylococcus aureus* and Gram negative bacteria were *Escherichia coli*.

It is evident from the figure that Mousambi had the maximum zone of inhibition when extracted in ethanol extract against *S. aureus* bacteria. Bains et al (2019) also revealed that ethyl alcoholic extract was more effective than aqueous extract for anti-bacterial finish when applied on the cotton fabric. Quantitative estimation of biological activity was carried out to determine the minimum inhibitory concentration (MIC) of herbal extracts on cotton fabric. Estimation was done by counting the colony forming units in the culture

incubated with treated samples for 24 hours. Eleven concentrations (1 to 11%) of both the plant extracts were used to study the reduction in bacterial colonies on the fabric. The results are shown in Table 1. It can be observed that cotton treated with 11 per cent Harshingar and 5 per cent Mousambi extracts showed maximum (74.7 and 63.1% respectively) reduction in microbial colonies against *S. aureus* while 11 per cent Mousambi and 11 per cent Harshingar extract showed maximum reduction against *E. coli*. Negligible reduction in bacterial colonies was found with lower concentrations of the plant extracts.

Lowest concentration of the extract which inhibited any visual microbial growth after treatment with plant extract was considered to be minimum inhibitory concentration (MIC). It is evident from the Table 2 (Plate 1) that MIC for Mousambi was 5 per cent against *S. aureus* while it was 11 against *E. coli*. The MIC was 11 per cent for Harshingar against both Gram positive and Gram negative bacteria.

**Visual inspection of samples by scanning electron microscope analysis:** The surface morphological features of the fabric samples were studied through high energy beam of electrons using scanning electronic microscope (Joshi et al 2007) and the surfaces of the treated fabric samples were photographed (Plate 2).

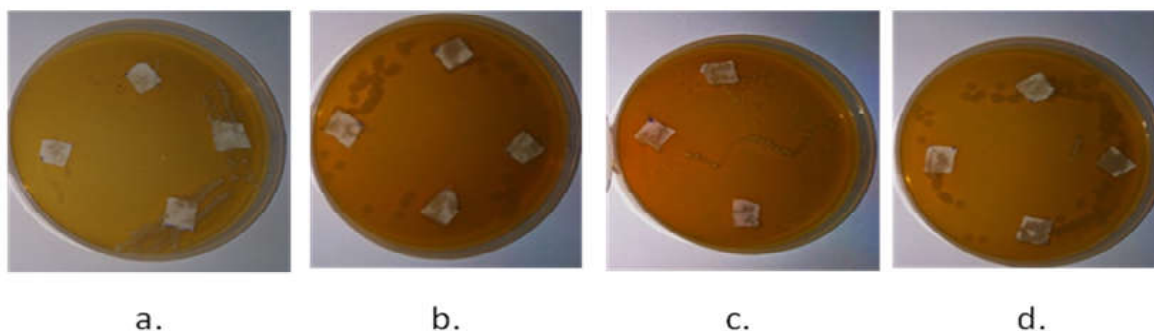


E: Ethanol, M: Methanol, DW: Distilled water

Fig 1. Microbial activity (Zoi) of the plant extracts

Table 1. Reduction in bacterial colonies using various concentrations of plant extract

Bacterium	Bacterial colonies under plant extract (%)										
	1	2	3	4	5	6	7	8	9	10	11
<b><i>Staphylococcus aureus</i></b>											
Mousambi	12.2	14.3	33.7	58.9	63.1	61.8	59.5	59.3	58.6	58.2	57.5
Harshingar	22.3	22.9	24.0	24.3	24.4	29.9	35.2	57.3	59.4	68.1	74.7
<b><i>Escherichia coli</i></b>											
Mousambi	37.2	37.9	39.5	44.7	57.5	58.3	60.0	62.5	65.7	67.9	71.2
Harshingar	0.9	1.6	3.4	7.3	19.1	10.2	11.4	12.8	13.3	14.0	47.2



**Plate 1. Bacterial colonies under minimum inhibitory concentration of herbal extract a. *S aureus* under Mousambi (MIC 5%), b. *E coli* under Mousambi (MIC 11%), c. *S aureus* under Harshingar (MIC 11%), d. *E coli* under Harshingar (MIC 11%)**

Table 2. Minimum inhibitory concentration of herbal extracts against bacteria

Herbal extract	Minimum inhibitory concentration (%)	
	<i>S aureus</i>	<i>E coli</i>
Mousambi	5	11
Harshingar	11	11

**Turbidity test:** The turbidity decreases with the increase in the concentration of the antimicrobial agent. Hence the turbidity rate decreases as the bacterial growth reduces. The fabric was found to be non-turbid at 5 per cent concentration against *S aureus* and at 11 per cent concentration against *E coli* in the case of Mousambi and at 11 per cent concentration in case of Harshingar against both the bacteria.

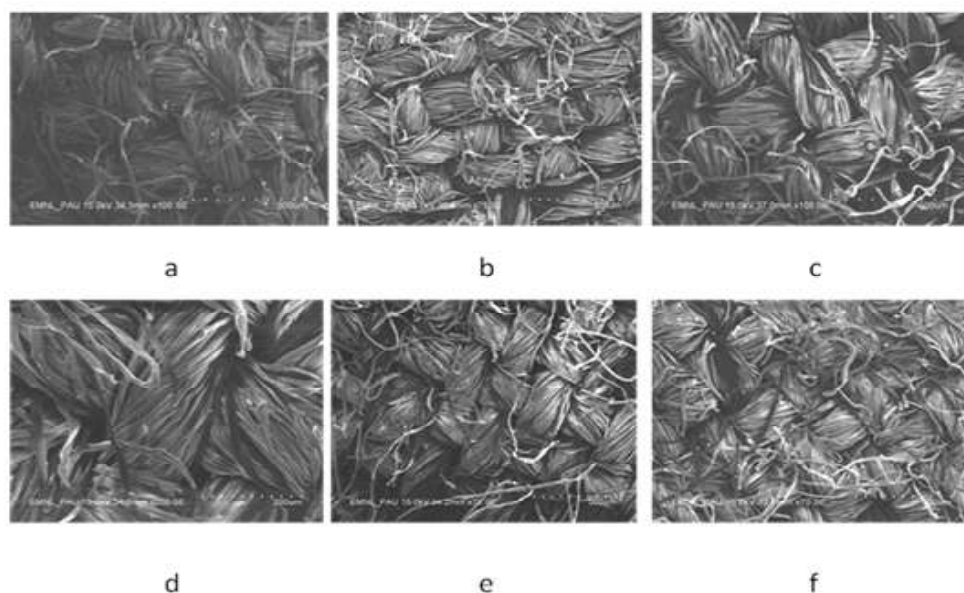
## CONCLUSION

Environment-friendly natural sources for textile application are gaining interest worldwide. The lower

cost of these sources can be exploited as an attractive eco-friendly alternative to synthetic agents for textile wet processing. The study showed that the antibacterial activity varies with solvent used for extraction. Cotton fabric treated with the leaf extract of Mousambi and Harshingar showed bacterial reduction against Gram positive and Gram negative bacteria. The finish proved cost effective and user friendly because it is natural, easily available and eco-friendly. It can be repeatedly applied as a renewable finish. It would also lay the foundation for healthcare textiles.

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**Plate 2. Scanning electron microscope (SEM) analysis of herbal-treated fabrics a. SEM image under Mousambi (control), b. SEM image under Mousambi (cross linking agent: direct method), c. SEM image under Mousambi (cross linking agent: pad-dry-cure method), d. SEM image under Harshingar (control), b. SEM image under Harshingar (cross linking agent: direct method), c. SEM image under Harshingar (cross linking agent: pad-dry-cure method)**

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