

Developing Antimicrobial Finishing of Cotton Fabrics using Different Medically Significant Herbs

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Abstract

Herbal medicines were gaining lot of interest in the field of medical textiles and hence, the present research was aimed to develop antimicrobial textiles using cotton fabrics finished with different types of medicinal herbs in the process of reactive dye method. Herbal extracts were prepared and finished onto cotton fabric samples using Pad-dry cure process. Finished fabrics were subjected to antibacterial activity and antifungal activity using standard EN ISO 20645 test method against respective bacteria (*Micrococcus luteus* and *Staphylococcus aureus*) and fungi (*Candida albicans* and *Candida tropicalis*). Test results of antibacterial activity of herbal extract finished fabrics revealed good inhibitory action against both test bacteria and fungi. Against bacteria, 35mm and 34mm of inhibitory zones were recorded for respective bacteria (*Micrococcus luteus* and *Staphylococcus aureus*). Similarly, against fungi, 35mm and 34mm of inhibitory zones were recorded for respective fungi (*Candida albicans* and *Candida tropicalis*). Physical properties such as air-permeability, tensile strength and absorbency between finished and unfinished fabrics did not showed any significant difference indicating that the nativity of the fabric was not altered even after finishing with herbal extracts. The present study would be considered highly significant by providing an eco-friendly and sustainable products for developing medical textile products and smart home textile products.

Keywords: Herbal extracts, Pad-dry cure, Antibacterial activity, Anti-fungal activity, Air-permeability.

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Introduction

Textile finishing is the application of active agents onto a fabric to improve the look and quality of the fabric by changing its appearance [Anik et al., (2025) and Moges et al., (2025)]. The purpose is to make textiles more flexible during their end-use, such as coloring and antimicrobial effects (Berkli et al., 2024). Microorganisms capable of degrading textiles are found everywhere, which makes the researchers to develop antimicrobial textiles for the prevention of healthcare-associated infections caused by some of the pathogenic bacteria such as *Micrococcus luteus* and *Staphylococcus aureus* usually found on textile surface and its interstices (Vincent et al., 2016). People are aware of pathogenic bacteria, which has led to the growing popularity of textiles treated with antibacterial agents, not just to prevent fabric deterioration, but to actively protect the wearer from bacterial contamination, combining personal health protection with enhanced fabric durability and hygiene in everyday use (Gulati et al., 2022). One of the most promising fields of new textile materials is the manufacturing of antimicrobial medical textiles (Yilmaz et al., 2023). The antibacterial properties of several secondary metabolites, including terpenoids, alkaloids, and phenolic chemicals, are present in plants (Othman et al., 2019). Roots, leaves, petals, and seeds are among the plant parts from which extracts have antimicrobial qualities (Voon et al.,

2012). The use of natural antimicrobial agents in medical and healthcare textiles has grown significantly because of their advantages over synthetic ones, including being safe, non-toxic, skin-friendly, and environmentally friendly (Bibi et al., 2024). Hence, developing a novel antibacterial agents that has a dual function of preventing the process of drug resistance and improving the properties of antimicrobial textiles against infectious disease are highly essential. From different literature survey, the emphasis of World Health Organization on the replacing antibiotics with natural ingredients was noticed (Son et al., 2022). The organization instructs the researchers to use rich reservoirs of useful compounds from nature to replace chemical and synthetic antibacterial agents (Bulut et al., 2021). The natural compounds shall be processed in the pharmaceutical industry for the production of natural antimicrobials with the very negligible side effects (Yonjalli et al., 2020). Based on these concept, in our present study, different herbs were selected. Herbal extracts were developed on solvent based extraction process and made into reactive with textile using standard methods for imparting durable antimicrobial finishes. Antimicrobial finish is an important property required in Meditech textiles (Sonu Rani and Alka Goel, 2023). Athimadhuram (*Glycyrrhiza glabra*), Amukkara (*Withania somnifera*), Karisilankanni (*Eclipta prostrata*), Keezhanelli

(*Phyllanthus niruri*) and Nochi (*Vitex negundo*) was selected as significant medicinal herbs based on the literature survey as these herbs were not used for antimicrobial finishing elsewhere. However, antibacterial activity of these medicinal herbs were studied by many researchers. Roy et al., evaluated the antibacterial activity of *Glycyrrhiza glabra*. Bishit and Rawat, (2014) studied the antibacterial activity of *Withania somnifera* against Gram-Positive pyogenic bacteria. Megala et al., (2021) reported on the antioxidant and antimicrobial activity of *Eclipta prostrata*. Sunitha et al., (2017) highlighted the antimicrobial effect of leaves of *Phyllanthus niruri* against different MTCC microbial strains in vitro. Recently, Vishal et al., (2024) studied antibacterial and anti-inflammatory activity of *Vitex negundo*. Hence

these herbs were selected for functional finishing onto cotton fabrics in the present study with the aim to develop antibacterial and antifungal finishes by imparting its reactive dyes and also to investigate its physical and comfort properties using textile standard methods.

Materials and methods

2.1 Fabric selection

Cotton (100%) was procured commercially with 30s count and 140 GSM.

2.2 Pre-treatment

All desized fabric materials was cut in to 10cm X 10cm. Five series of steps were carried out to pre-treat all the five fabrics separately. In Table-1, the fabric pre-treatment process for all the five fabrics were illustrated.

Table-1: Pre-treatment Steps

S. No.	Steps
1	Soaking fabrics in water @ 70°C for 20min
2	Rinsing in running water @ 50°C
3	Relaxation and drying @ room temperature
4	Non-Ionic detergent (Machine washing)
5	Shade drying

Initially, the fabrics were soaked in normal fresh water at 70°C for 20min. The washed fabrics were rinsed well in the running water to remove surface dirt and other impurities. All the fabrics were squeezed (relaxed) gently in hand and dried at room temperature. To improve excess starchy like components from fabric surface and interstices, non-ionic detergent (sodium lauryl sulphate – 1% (10g/L) was used. Washing was done in a normal washing machine kept at low speed

for 10min. Washed fabrics were relaxed in hand and shade dried for 120min.

2.3 Selection of herbs

As medicinal herbs are considered very significant to inhibit the growth and metabolism of microbes such as bacteria and fungi, five different herbs were selected in our present study. The common names and scientific names of the herbs were presented in Table-2, below.

Table- 2: Medicinal herbs used in the study

S. No	Common name	Properties
1	Athimadhuram	<i>Antibacterial, Antifungal, Antiviral, Anti-inflammatory, Anti-cancer and Anti-oxidant</i>
2	Amukara	
3	Karisilankanni	
4	Keezhanelli	
5	Nochi	

2.4 Processing of herbs

All the selected herbs (Table-2) were collected from the farm house, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India. All the leaves of the selected herbs were pre-washed in deionised water and kept under shade until completely dried for milling. The dried leaves were arranged separately in aluminum trays and oven dried at 40°C to 50°C for 2h until the moisture content reach below 10%. The dried parts were grounded to powder and sieved using metal sieve. Sieved particles were stored at room temperature prior testing. Sieved herbs were presented in Fig. A.

Fig. A: Medicinal herbs used in the study



Athimadhuram

Karisilankanni



Keezhanelli

Nochi



Amukkara

2.5 Soxhlet extraction and evaluating the antimicrobial properties of herbal extracts

Athimadhuram powders (Fig) was selected for the present research. Powdered herbs were further processed for getting extractions. Extraction of each herbs were carried out using a standard Soxhlet method. In the Soxhlet extraction method, finely ground sample - *Athimadhuram* powder was placed in a porous bag or “thimble” made from a strong filter paper or cellulose, which is placed, in thimble chamber of the Soxhlet apparatus. Extraction solvent (acetone and water) is heated in the bottom flask, vaporizes into the sample thimble, and condenses in the condenser and drip back. When the liquid content reaches the siphon arm, the liquid contents is emptied into the bottom flask again and the process is continued. For the study, infusion method of Soxhlet Extraction had been adopted. The powdered herbs were filled in the thimble and placed in the soxhlet extractor. The

extractor had been filled with solvent solution of acetone and the temperature of 60°C was set and left for 6 hours. Slowly and steadily the temperature was increased up to 100°C. The extract from the thimble was collected in the round bottom flask kept in the heating mantle below by passing through a side arm tube. Similar procedure was carried out for the other herbs under controlled conditions to obtain the acetone extracts. The collected extracts of medicinal herbs were stored at room temperature for further antibacterial and antifungal screening tests

2.5.1 Antibacterial activity of the extracts (One best solvent selected)

The antibacterial activity of Herbal extracts was evaluated against the test organisms by well diffusion method separately. All the test cultures (*Micrococcus luteus* and *Staphylococcus aureus*) were inoculated in a sterile Nutrient broth* and allowed to attain the growth

for 24 to 48 hours. Sterile Mueller-Hinton Agar* plates (MHA) were prepared and allowed to solidify. About 0.1% inoculum suspensions of the test organism (*Micrococcus luteus* and *Staphylococcus aureus*) were swabbed uniformly over the agar surface separately. Under sterile conditions, 6mm wells were cut on the agar surface of each MHA plates. About 20µl of Herbal extract fractions (100µg/ml prepared using Dimethyl sulfoxide - DMSO) were loaded into the well separately for all medicinal herbs. And all the plates were incubated at 37°C for 24h. Standard antibiotic streptomycin is used as positive control in one well to compare the results. Dimethyl sulfoxide - DMSO was used as negative control. The antibacterial activity was evaluated in terms of zone of inhibition around the wells in all the inoculated NA plates. The inhibition clear zones were measured and recorded in millimeter.

2.5.2 Antifungal activity of the extracts (One best solvent selected)

The antifungal activity of Herbal extracts was evaluated against the test organisms by well diffusion method. All the test cultures (*Candida albicans* and *Candida tropicalis*) were inoculated in a sterile Nutrient broth* and allowed to attain the growth for 24 to 48 hours. Sterile Mueller-Hinton Agar* plates (MHA) were prepared and allowed to solidify. About 0.1% inoculum suspensions of the test organism (*Candida albicans* and *Candida tropicalis*) were swabbed uniformly over the agar surface separately. Under sterile conditions, 6mm wells were cut on the agar surface of each MHA plates. About 20µl of Herbal extract fractions (100µg/ml prepared using Dimethyl sulfoxide - DMSO) were loaded into the well separately for all medicinal herbs. And all the plates were incubated at 37°C for 24h. Standard antifungal agent, amphotericin was used as positive control in one well to compare the results. Dimethyl sulfoxide - DMSO was used as negative control. The antifungal activity was evaluated in terms of zone of inhibition around the wells in all the inoculated NA plates. The inhibition clear zones were measured and recorded in millimeter.

2.6 Method of finishing (Dip and dry method)

The pre-treated cotton fabric samples were finished with the developed herbal extracts using dip and dry method. Each finishing was done separately by immersing in a dye bath containing the herbal extracts at liquor ratio of 1:50 at room temperature without heating. Different set of fabric samples were selected for finishing each type of herbal extracts. After finishing, all the samples were dried under shade for 20min at room temperature and followed by curing in dry oven at 50°C to 60°C for 15 min for effective binding. All the samples were stored at room temperature for further analysis.

2.7 Evaluation of finished fabrics

2.7.1 Qualitative antibacterial activity by ENISO 20645

The herbal extract finished samples were taken as test specimens. Antibacterial activity was tested for both top and middle layer separately as per EN ISO 20645 standard methods. All the samples were cut into pieces (20mm in diameter). Sterile Nutrient agar plates* were prepared and allowed to solidify. Using sterile 4mm inoculating loop, one loop full of 12 hours old bacterial culture (*Micrococcus luteus* and *Staphylococcus aureus*) was transferred on to the centre of the Nutrient Agar Media. Using sterile cotton swabs, the bacterial aliquots were swabbed all around the surface of the agar plate. For each test organism, separate Nutrient agar plates were used in a sterile zone. Test specimens [herbal extract finished samples] were placed over the bacterial cultured inoculated plates separately. All the plates were incubated at 37°C for 24 hours. The test plates were examined for the clear zone of inhibition around each samples. The average width of the zone of inhibition around each type of fabric specimen was measured in millimeter (mm).

2.7.2 Qualitative antifungal activity by ENISO 20645

The herbal extract finished samples were taken as test specimens. Antifungal activity was tested for both top and middle layer separately as per EN ISO 20645 standard methods. All the samples were cut into pieces (20mm in diameter). Sterile Nutrient agar plates* were prepared and allowed to solidify. Using sterile 4mm inoculating loop, one loop full of 12 hours old fungal culture (*Candida albicans* and *Candida tropicalis*) was transferred on to the centre of the Nutrient Agar Media. Using sterile cotton swabs, the bacterial aliquots were swabbed all around the surface of the agar plate. For each test organism, separate Nutrient agar plates were used in a sterile zone. Test specimens [herbal extract finished samples] were placed over the bacterial cultured inoculated plates separately. All the plates were incubated at 37°C for 24 hours. The test plates were examined for the clear zone of inhibition around each samples. The average width of the zone of inhibition around each type of fabric specimen was measured in millimeter (mm).

2.8 Comfort and physical properties

2.8.1 Fabric weight

According to ASTM D 3776 standard, fabric weight of the fabrics was carried out. It measures the fabric mass per unit area (weight). It is expressed as grams per square meter. Samples were cut using the GSM cutter avoiding selvage and creases. The samples were allowed to condition in standard atmospheric condition for one hour after which the weight was taken on a digital balance having 0.01g sensitivity. The mass per unit area of the sample was calculated using the formula.

$$M_{ua} = W \times 100$$

2.8.2 Tensile strength test (ASTM D 5035-2006 test method)

Tensile strength is the measure of the resistance of the fabric tensile load or stress in either warp or weft

direction. It is the strength shown by a specimen subjected to tension as distinct from torsion, compression, or shear. Elongation defines the length to which a fibre may stretch before breaking. A sample of 12” X 2” was taken for the test. The tensile strength of the fabric was determined by cloth tensile strength tester. Tensile strength is performed using cut strip method. This test is used for finished or heavily sized fabrics. Three readings for every sample were taken and the average was calculated.

2.8.3 Air- permeability test (ASTM D 737-96 test method)

Air permeability of a fabric is the volume of air measured in cubic cm passed per second through 1 sq. cm for the fabric at a pressure of one cm. head of water. Air permeability can be measured using an instrument called Shirley Air Permeability Tester. Air permeability was determined in accordance with Test Method ASTM D-737-96. The conditioned specimens in the specimens to transport liquid along and/or through them” Wicking rate is a particularly important property that measures a fabric’s ability to remove sweat/liquid from contact with the skin. The wickability of herbal extract finished fabric was evaluated by time for wetting. During the analysis, the fibre samples were conditioned in a standard atmosphere of 22°C under 65% relative humidity for 24 hours. The pre-measured size (1.5cm x 5cm) of each test mounted on the glass slides was kept at immersed condition inside a reservoir containing distilled water. The wicking height of the advancing liquid front as a function of time was recorded by visual observation after 5 minutes. Using a standard ruler scale, the colour of water absorbed on the fibre surface was measured for each sample and the values were recorded.

standard atmosphere for testing textiles, 21 ± 1°C and 65 ± 2 % relative humidity was tested unless otherwise specified in a material specification or contract order. The test specimens were carefully handled to avoid altering the natural state of the material. Placed each test specimen onto the test head of the test instrument, and performed the test as specified in the manufacturer’s operating instructions. Read and recorded the individual test results in SI units as cm³/s/cm².

2.8.4 Absorbency or Wicking test (AATCC TM 197)

Water or Moisture absorbing abilities of the herbal extract finished fabric sample was studied using AATCC TM 197 standard test method. The method describes the vertical wicking properties of herbal extract finished fabric. The principle of the study describes that, Vertical Wicking of Textiles, is used to measure "the ability of vertically aligned fabric

3. Results and discussion

3.1 Antibacterial activity of the extracts (One best herb selected)

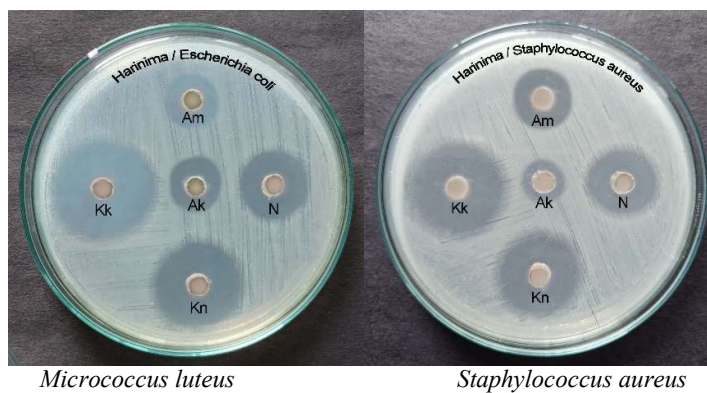
Antibacterial activity of all five types of herbal extracts was observed against both test bacteria. Herbal extracts exhibited maximum inhibitory zones of 18mm and 19mm for karisilankani extracts against respective bacterial cultures, *Micrococcus luteus* and *Staphylococcus aureus*.

All these values were found significant in terms of antibacterial activity when compared to the inhibitory zone size (20mm to 21mm) obtained for streptomycin. Based on these results, karisilankani extracts were selected for finishing the cotton fabric samples (Table-3 and Fig. 1).

Table-3: Antibacterial activity of herbal extracts

S. No.	Control and Samples	Zone of inhibition (mm)	
		<i>Micrococcus luteus</i>	<i>Staphylococcus aureus</i>
1	Athimadhuram	13	14
2	Amukara	12	10
3	Karisilankani	18	19
4	Keezhanelli	16	17
5	Nochi	14	15
6	Positive control (Streptomycin)	20	21

Fig. 1: Antibacterial activity of herbal extracts



3.2 Antifungal activity of the extracts (One best herb and solvent selected)

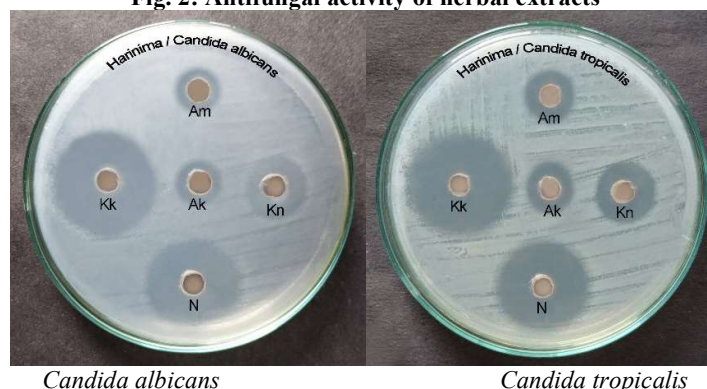
Antifungal activity of all five types of herbal extracts was observed against both test fungi. Herbal extracts exhibited maximum inhibitory zones of 17mm and 18mm for karisilankani extracts against respective bacterial cultures, *Candida albicans* and *Candida tropicalis*.

All these values were found significant in terms of antibacterial activity when compared to the inhibitory zone size (19mm to 20mm) obtained for streptomycin. Based on these results, karisilankani extracts were selected for finishing the cotton fabric samples (Table-4 and Fig. 2).

Table-4: Antifungal activity of herbal extracts

S. No.	Control and Samples	Zone of inhibition (mm)	
		<i>Candida albicans</i>	<i>Candida tropicalis</i>
1	Athimadhuram	10	11
2	Amukara	11	12
3	Karisilankani	17	18
4	Keezhanelli	12	13
5	Nochi	15	16
6	Positive control (Streptomycin)	19	20

Fig. 2: Antifungal activity of herbal extracts



3.3 Evaluation of finished fabrics

3.3.1 Qualitative antibacterial activity by ENISO 20645

Antibacterial activity of the Herbal extract cotton fabric (HEc) finished fabric samples expressed good inhibitory zones against both test bacteria for all the samples tested. In Table-5, the zone size measured in millimeter for each sample was presented. In Fig. 3, zone obtained for all the samples were found evident.

Herbal extract finished fabric samples exhibited inhibitory zone size of about 35mm and 34mm against *Micrococcus luteus* and *Staphylococcus aureus* respectively (Table-5 and Fig. 3). Unfinished fabric sample showed no activity when compared to herbal finished sample.

The obtained results indicated that herbal extract contains significant phytochemical compounds that attributes well for its potential Antibacterial properties. These compounds may be alkaloids, flavonoids, phenol, tannins or terpenoids along with some biochemical compounds present in cotton charcoal.

Table-5: Antibacterial activity of the Herbal extract finished fabric samples

Test Bacteria	Zone of Inhibition (mm)	
	Unfinished fabric	HEc finished fabric
<i>Micrococcus luteus</i>	0	35
<i>Staphylococcus aureus</i>	0	34

Fig. 3: Antibacterial activity of Herbal extract finished fabric



3.3.2 Qualitative antifungal activity by ENISO 20645

Antifungal activity of the Herbal extract (HEc) finished fabric samples expressed good inhibitory zones against both test Fungi for all the samples tested. In Table-6, the zone size measured in millimeter for each sample was presented. In Fig. 4, zone obtained for all the samples were found evident.

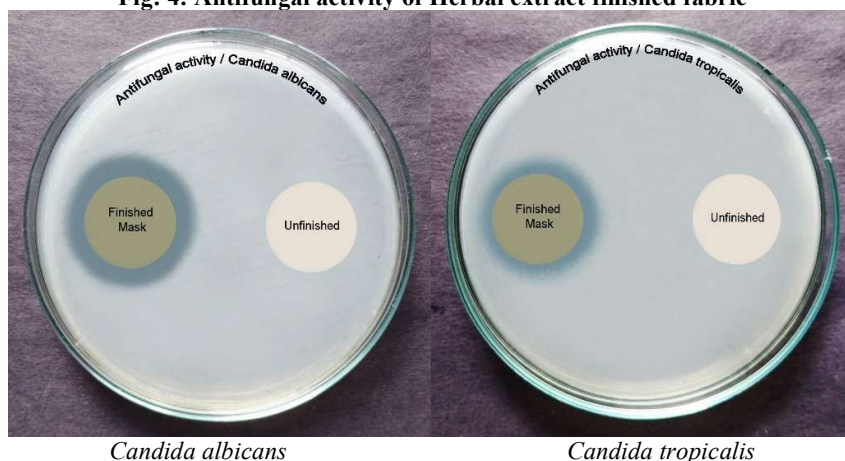
Herbal extract finished fabric samples exhibited inhibitory zone size of about 33mm and 30mm against *Candida albicans* and *Candida tropicalis* respectively (Table-6 and Fig. 4). Unfinished fabric sample showed no activity when compared to herbal finished sample.

The obtained results indicated that herbal extract contains significant phytochemical compounds that attributes well for its potential Antifungal properties. These compounds may be alkaloids, flavonoids, phenol, tannins or terpenoids along with some biochemical compounds present in cotton charcoal.

Table-6: Antifungal activity of the Herbal extract finished fabric samples

Test Fungi	Zone of Inhibition (mm)	
	Unfinished fabric	HEc finished fabric
<i>Candida albicans</i>	0	33
<i>Candida tropicalis</i>	0	30

Fig: 4: Antifungal activity of Herbal extract finished fabric



3.4 Comfort and physical properties

Physical properties of Herbal extract finished fabric samples were tested and compared with unfinished control fabrics based on tensile strength test and wicking test. In Table-7, the values obtained separately for each test were recorded.

Table-7: Physical properties of Herbal extract finished fabrics

S. No	Tensile strength (MPa/kgf)		Wicking height in (cm)		Inference
	Unfinished fabric	Finished fabric	Unfinished fabric	Finished fabric	
1	37.9 ± 0.57	36.6 ± 0.75	2.9 ± 0.57	2.6 ± 0.75	No significant difference

The tensile strength of Herbal extract finished fabric samples did not showed significant difference on their respective values when compared to control unfinished plain fabric. This indicated that the Herbal extract extracts after finishing on to the fabrics did not altered or changed the nativity of the fibres.

The vertical wicking test was done for the unfinished and finished fabrics. The fabric finished with Herbal extract showed wicking height of 2.6 ± 0.75 cm; and the unfinished fabric also showed almost similar absorbency in terms of height in centimetre of 2.9 ± 0.57 (Table-6). This indicated that the Herbal extract finished for Antibacterial and anti-oxidant properties does not have any impact on the absorbency of the fabrics. The nativity of the fabric was not altered significantly.

Table-8: Comfort properties of Herbal extract finished fabrics

S. No	Fabric Weight (GSM)		Air permeability (cm ³ /cm ² /sec)		Inference
	Unfinished fabric	Finished fabric	Unfinished fabric	Finished fabric	
1	141.9 ± 0.57	142.6 ± 0.75	23.9 ± 0.57	22.6 ± 0.75	No significant difference

The fabric weight of the herbal extract finished fabric samples did not showed significant difference on their respective values when compared to control unfinished plain fabric. This indicated that the Herbal extract extracts after finishing on to the fabrics did not altered or changed the nativity of the fibres.

The air-permeability test was done for the unfinished and finished fabrics. The fabric finished with Herbal extract showed air-permeability of 22.6 ± 0.75 cm³/cm²/sec; and the unfinished fabric also showed almost similar air-permeability of 23.9 ± 0.57 cm³/cm²/sec (Table-8). This indicated that the Herbal extract finished for Antibacterial and anti-oxidant properties does not have any impact on the comfort and physical properties of the selected fabrics; and thus the nativity of the fabric was not altered significantly.

Conclusion

Different medicinal herbs were selected on the present study based on their antimicrobial potential against bacteria and fungi as per literature survey. The herbs were also selected because it was not used for antimicrobial finishing on to textiles. Herbal extracts were screened for its antimicrobial efficacy and found *Karisilankanni (Eclipta prostrata)* was more effective against bacterial and fungal strains. Hence *Eclipta prostrata* extracts were finished onto cotton fabrics and its antimicrobial and physical properties were evaluated using textile standards. Finished fabrics showed good inhibitory zones against *Micrococcus luteus* and *Staphylococcus aureus*; and *Candida albicans* and *Candida tropicalis*. Physical properties such as air-permeability, tensile strength and absorbency between finished and unfinished fabrics did not showed any significant difference indicating that the nativity of the fabric was not altered even after finishing with herbal extracts. The present study would be considered highly significant by providing an eco-friendly and sustainable products for developing medical textile products and smart home textile products. To conclude that, the herbal extracts used for finishing the fabrics would be considered highly significant for the development of eco-friendly and sustainable products.

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