

Efficiency Of Ricinus Communis And Rosmarinus Officinalis L. Extracts On Cotton Fabrics And Its Physio-Mechanical Assessments

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Abstract: These Biologically active compounds are used to control undesirable growth of bacteria, fungi, mold, mildew and algae. Their control reduces or eliminates the problems of skin diseases that are caused by the micro-organisms. In this study the single jersey cotton knitted fabric was treated with rosemary (*Rosmarinus officinalis* L.) and castor seed (*Ricinus communis*) extract and two bacterial strains namely *Staphylococcus aureus* and *Escherichia coli* were used. After treatment the fabric was subjected to anti-microbial assessment of finished fabric by agar diffusion method. The physical properties, morphological and structural are investigated of treated cotton fabric samples. 1mg/ml, 1.5mg/ml and 2mg/ml rosemary extract showed the zone of inhibition in 1.20 ± 0.17 mm, 2.07 ± 0.12 mm, and 3.00 ± 0.00 mm respectively for gram-positive bacteria and similarly 1.00 ± 0.00 mm 1.87 ± 0.12 mm and 2.63 ± 0.21 mm for gram-negative bacteria respectively. Castor seed extract showed the zone of inhibition in 2.20 ± 0.20 mm, 3.00 ± 0.10 mm, and 3.93 ± 0.12 mm respectively for gram-positive bacteria and 2 ± 0 mm, 2.77 ± 0.25 mm, 3.30 ± 0.26 mm for gram-negative bacteria respectively. The results for qualitative assessment of microbiological activity on a cotton fabric with in different concentration of rosemary oil and castor oil were more energetic against gram-positive bacteria (*S. aureus*) as compared to untreated fabrics. On the opposing, the gram-negative bacteria were slightly affected (*E. coli*). The extract showed increasing inhibitory activity with an increase in concentration. Treated knitted cotton fabric still showed weakness in washing durability and there are no significance variations in physical properties of fabric.

Index Terms: Antimicrobial, Castor Seed, Rosemary, Rosemary, Tensile Strength, Textiles, Wicking.

1. INTRODUCTION

ALL items of apparel and home textiles are susceptible the problems of hygiene in normal daily use, for example, socks, sportswear, and working clothes as well as mattresses, floor coverings, and shoe linings. Textiles for outdoor use are constantly exposed to the influence of microbes and bacteria. Most textile materials are currently used in hospitals and hotels are conducive to cross infection or transmission of diseases caused by microorganisms [1].

The growth of microorganisms on textiles can lead to functional, hygienic and aesthetic difficulties. The most trouble-causing organisms are fungi and bacteria. Under very moist conditions, algae can also grow on textiles but are troublesome only because they act as nutrient sources for fungi and bacteria. The use of antimicrobial finishes to prevent unpleasant odors on intimate apparel, underwear, socks and athletic wear is an important market need. The odors are produced by the bacterial decomposition of sweat and other body fluids, and controlling bacterial growth by hygiene finishes reduces or eliminates the problem [2]. Bacteria cause multiple problems to textiles including discoloration, colored stains, and fibre damage. Bacteria are not damaging to fibres, but can produce some fibre damage, unpleasant odors and a slick, slimy feel. Often, fungi and bacteria are both present on the fabric in a symbiotic relationship. Substances added to fibres, such as lubricants, antistats, natural-based auxiliaries (for example size, thickener and hand modifiers) and dirt provide a food source for microorganisms [3]. Antimicrobial agents are used to control the undesirable growth of bacteria, fungi, mold, mildew and algae. Their control reduces or eliminates the

problems that are caused due to microorganisms. Antimicrobial agents vary in their chemical nature, mode of action, cost, substrate to which they are applied, durability and their impact on people and the environment. The unpleasant odor develops when among other things; bacteria convert human perspiration into foul-smelling substances, such as carboxylic acid, aldehydes and amines. Gram-negative bacteria *P. vulgaris* is known to be able to metabolise urea to form ammonia and is the cause for generation of odor in baby diapers. Several products can be used to tackle the odor problem in textiles. Use antimicrobials to prevent the formation of odor causing compounds by inhibiting the growth of bacteria. In many personal care products around the world, such as underarm deodorants, antimicrobial agents such as triclosan have already been widely used with satisfactory results [4]. Textiles are easily attacked by microorganisms, which mean that they quickly become damaged. Microbial degradation of fabrics depends primarily on their chemical composition. Fabrics of natural origin are particularly susceptible to attack by microorganisms. Bacterial decomposition of cellulose takes place from outside to inside, but it cannot digest cellulose directly. Cellulolytic microorganisms secrete enzymes, which make cellulose soluble followed by the diffusion of microbes inside the cell. Carbon heterotopy type of bacteria degrade polysaccharide chains into shorter ones and these are eventually hydrolyzed to shorter oligomers and then finally to cellobiose and D-Glucose. As a result of enzymatic degradation, the strength of cotton is reduced [5], [6]. The *Rosmarinus officinalis* L. is widely known for its numerous applications in the food field and also for the increasing interest in its pharmaceutical properties [7]. Two groups of compounds are mainly responsible for the biological activities of the plant: the volatile fraction and the phenolic constituents. The latter group is mainly constituted by rosmarinic acid, by a flavonoid fraction and by some diterpenoid compounds structurally derived from the carnosic acid. Rosemary (*Rosmarinus officinalis* L.) originally grows in southern Europe. Its herb and oil are commonly used as spice and flavoring agents in food processing for its desirable flavor, high antioxidant activity and lately as antimicrobial agent. High

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percent of the antimicrobial activity they attributed to carnosic acid and carnosol. It is clear that rosemary extracts have bioactive properties, but their antimicrobial activities have not been deeply characterized. Antimicrobial activities of plant essential oils have been known for centuries, but their strong flavor limited their use in food [8]. The biological properties of rosemary have been attributed to its phytochemical composition rich in (poly) phenolic compounds, main diterpenoids such as carnosic acid and carnosol [9]. The (poly) phenolic profile of the proprietary rosemary extract rich in carnosic acid was evaluated using an UHPLC-ESI-MSn untargeted method consisting of two complementary mass spectrometry (MS) conditions [10]. Castor plant, *Ricinus communis*, is a species of flowering plant in the spurge family, Euphorbiaceae. Its seed is the castor bean which, despite its name, is not a true bean [11]. Leaf essential oil and leaf methanol extract of castor plant have recently been reported to possess potent antibacterial, antifungal and leishmanicidal activity [12], [13]. Isolated lectin, from the seed of *R. communis* has previously been reported to exert anti-proliferative activity against tumor cells both in vitro and in vivo condition. With the advancement of nanocarrier's research, scientists are finding great prospect in cancer therapy with castor lectin (ricin) to selectively target the cancer cells using nano-carriers [14].

2 MATERIALS AND METHODS

2.1 Sample Collection and Preparation

The fresh seed of castor and rhizomes of rosemary were collected from Arba Minch town of the Southern Nations, Nationalities, and Peoples Region, Ethiopia and washed with clean water and rinsed with distilled water. Washed castor seeds were prepared for use by removing the endocarp, and sun drying was implemented for sample to reduce the moisture contents; examining to separate the shell from the nibs (cotyledon). This was carried out using a tray to blow away the cover in order to achieve very high yield and the seeds were crushed by using mortar and pestle. The powdered samples were kept for further analysis. The collected rosemary sample was air dried, cut into small pieces and crushed in a mechanical blender. Powdered plant material was used for the preparation of methanol extracts. Desized, scoured and bleached 100% cotton with yarn count of 32^s Ne, GSM of 168, loop length of 2.3 mm course count per cm of 22.0 and wale count per cm of 12.00 single jersey knitted fabric was used selected for the study and bacterial species viz., Gram-positive *Staphylococcus aureus* (ATCC 6538) and Gram-negative *Escherichia coli* (ATCC 8739) were created at microbiology laboratory.

2.2 Final Stage

Methanolic extract of the sample was extracted using dried powder of rosemary leaves and castor seed by employing methanol as a solvent using soxhlet apparatus. There are different methods of essential oil extraction from plant material. Typically, Soxhlet extraction is used when the desired compound has a limited solubility in a solvent, and the impurity is insoluble in that solvent. It allows for unmonitored and unmanaged operation while efficiently recycling a small amount of solvent to dissolve a larger amount of material, therefore in the present study soxhlet apparatus was used. The extraction was continued for 6 hours. The obtained extract was filtered using filter paper (Whitman no.1) and the

remaining solvent was evaporated under reduced pressure at 60°C with the help of rotary evaporator. The dry fractions of sample were stored in refrigerator at 4°C in airtight glass bottles [15].

2.3 Quantitative Determination of Phytochemical Constituents

2.3.1 Determination of Alkaloids

Plants sample (5 gram) was taken in a beaker and the solution of ethanol and 10% of acetic acid of 200 ml was added into the plant sample. The mixture was covered with aluminum foil and allowed to stand for 4 hours and finally filtered. The extract was placed in a water bath until it reaches 1/4 of the native volume, the extract was enabled to become concentrated and then concentration NH₄OH was added until the precipitation completed. Resolved the whole solution then collected the precipitate and wiped with dilute NH₄OH and finally filtered. Then the samples were dried and the percentage of alkaloid was determined. [16].

2.3.2 Determination of Alkaloids

Flavonoid determination was by the method reported by Eijkeme et al. [17]. Exactly 50ml of 80% aqueous methanol was added to 5gram of sample in a 250ml beaker, covered, and allowed to stand for 24 hours at room temperature. After discarding the supernatant, the residue was re-extracted (three times) with the same volume of ethanol. Whatman filter paper number was used to filter the whole solution of each plant sample. Each plant sample filtrate was later transferred into a crucible and evaporated to dryness over a water bath. After constant weight obtained, the percentage of flavonoid was calculated [18].

2.3.3 Determination of Lipid

Exactly 2.5gram powder each rosemary and castor added into a thimble connected to a Soxhlet extractor chamber with a pre-weighed flat bottom and connected to a condenser. Petroleum ether (100mL) enough to cause a reflux was added to the flask and the lipid from the sample was extracted for 3 hours by heating on an electric hot plate at 50°C. The petroleum ether was distilled off and the lipid recovered by cooling the flask in a desiccator and its value calculated by reweighed flask and content. This lipid quantitative analysis was done according to literature procedure [17], [19].

2.3.4 Determination of Saponins

Exactly 100ml of 20% aqueous ethanol was added to 5 grams of each plant sample in a 250mL conical flask. The mixture was heated over a hot water bath for 4 hours with continuous stirring at a temperature of 55°C. The residue of the mixture was re-extracted with another 100ml of 20% aqueous ethanol after filtration and heated for 4 hours at a constant temperature of 55°C with constant stirring. The combined extract was evaporated to 40ml over a water bath at 90°C. 20ml of diethyl ether was added to the concentrate in a 250ml separator funnel and vigorously agitated from which the aqueous layer was recovered while the ether layer was discarded. This purification process was repeated twice. 60ml of n-butanol was added and extracted twice with 10ml of 5% sodium chloride. After discarding the sodium chloride layer the remaining solution was heated in a water bath for 30 minutes, after which the solution was transferred into a

crucible and was dried in an oven to a constant weight. The saponin content was calculated as a percentage [17].

2.4 Finishing Procedures of Fabrics Using Plant Extracts

Prior to finishing process the fabric was washed well with dilute soap solution to remove the impurities so that they do not interrupt in further finishing process. The fabric was washed at 60°C in a bath containing distilled water and non-ionic detergent for 30 min. After washing the fabric was rinsed thoroughly with water to remove the soap solution completely and dried at room temperature. Each of the different samples of methanolic extract was mixed thoroughly with citric acid as a crosslinking agent; subsequently, it was added into distilled water. For making the finishing of fabric, pad dry cure method was used. Sample fabrics were dipped in a prepared antimicrobial solution for 30 min at 40°C subsequently; padding was carried out in padding mangle by maintaining the temperature with 3.5 Kg/cm² pressure. Curing of finished fabric was carried out at 100°C for 20 min in a hot air oven and dry at room temperature.

2.5 Anti-bacterial Assessment of Finished Fabric by Agar Diffusion Method

Culture medium (lysogeny broth, LB) was prepared by dissolving 0.45g lysogeny broth powder by 5mL distilled water and the solution was sterilized. The bacteria stock was transferred to lysogeny broth, and also the nutrient broth (NB) was prepared by dissolving 0.36gram nutrient broth powder by 5mL distilled water after preparation the NB and LB was sterilized at 121°C for 15 min. and then incubated at 37°C for 24hr. 4.7gram of plate count agar (PCA) powder was measured and dissolved with 200ml of distilled water and sterilized at 121°C for 15 min. The solution of PCA was transferred to sterilized petridishes, after some minutes it's solidified. On these petridishes the prepared bacterial solution was added. Fabrics of treated with natural plant and untreated (control) fabric samples were placed in intimate contact with plate count agar and lysogeny broth agar, which has been previously inoculated with an inoculum of test organisms in petridishes. Two test organisms namely *Staphylococcus aureus* and *Escherichia coli* were used for this study. The plates incubated at 37°C for 18-24 hrs. After incubation, a clear area of uninterrupted growth underneath and along the side of the test material indicated the antibacterial effectiveness of the fabrics.

2.6. Investigations of Treated Cotton Fabric Samples

2.6.1. Scanning Electron Microscopy (SEM)

Scanning electron microscopy (SEM) is a powerful technique that produces images of a sample by scanning the surface with a focused beam of electrons. The scanning electron microscopy creates magnified images by using electrons instead of light waves. The electrons interact with the sample atoms producing signals that contain information about the sample's surface topography and composition. When a beam of electrons strikes the surface of the sample and interacts with the atom of the sample, signals in the form of secondary electrons, backscattered electrons and characteristic X-rays are generated that contains information about the samples surface topography, composition, etc. The scanning electron microscope (SEM) is a type of electron microscope that images the sample surface by scanning it with a high-energy

beam of electrons in a raster scan pattern. Completely dry the sample in the drying oven at 60°C for 3 hours. The sample was loaded into the SEM holder and turning on the SEM. SEM images of the treated and untreated samples were obtained. In the present study, the particle morphology of the fabrics was performed using a scanning electron microscopy images taken from JEOL JSM-6610LV instrument.

2.7. Physico-mechanical Characterization

The physico-mechanical properties of knitted fabric can be changed due to the use of various count of yarn, type of yarn (ring, rotor, and compact), quality of yarn, Loop length / Stitch length, structural geometry, fibre composition of yarn etc. some physico-mechanical properties of the fabrics are wicking property, tensile strength, wash durability, air permeability etc. The antibacterial treated cotton fabric samples were tested to assess some of the physico-mechanical properties compared with untreated knitted cotton samples.

2.8 Data Analysis

Once the results were acquired, they were treated statistically with the help of one way analysis of variance (one way ANOVA) tables to indicate the significance of the finishing treatment effects. Considered significant if the probability level was 0.05 or less and highly significant if the probability level was 0.01 or less.

3 RESULTS AND DISCUSSIONS

3.1 Phytochemicals Characterization of Rosemary and Castor Seed Extract

The extraction yield is a measure of the solvent efficiency to extract specific components from the original material. The rhizomes of rosemary plants and castor seed were extracted using methanol. In this study the extractive values were found 21.67 ± 0.12% and 49.57 ± 0.24%, of rosemary and castor seed extract respectively (Table 3.1) based on the weight of plant material and weight of extract. The percentage yield of rosemary essential oil was calculated by using percentage of extraction yield equation. The extraction yield of methanol extracts of rosemary was 19.4%, which was lesser than the current findings [20]. The extraction yield varied in each plants and their solvent extracts. The extraction yield of castor seed was 66.67% which was higher than our finding [21]. The extraction yield is dependent on the solvent and method of extraction [22]. The extracts were subjected to quantitative phytochemical investigation for the presence of alkaloids, flavonoids, saponins and lipids were the result obtained from the quantitative determination of alkaloids, flavonoids, saponins and lipids from methanol extracts are illustrated in (table 3.2). Generally, small quantities of saponins were found in castor plant extracts examined with the highest quantities obtained in rosemary extract (2.4 ± 0.1%), the highest quantities of alkaloid were obtained in castor oil (0.28 ± 0.11%), rosemary (0.25 ± 0.11%). Flavonoid value of castor oil is higher than the methanolic extract of rosemary. In general castor plant extract had more lipids value than rosemary (4 ± 0.1%) [23].

TABLE 3.1
PERCENTAGE YIELD OF CRUDE EXTRACT OF ROSEMARY AND CASTOR SEEDS

Plant material	Wt. of plant material (g)	Vol. of sol. (ml)	Wt. of extract (g)	% yield of extract
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Rosemary	40 ± 0.00	250 ± 0.00	8.67± 0.67	21.67 ± 0.12
Castor seed	40 ± 0.00	250 ± 0.00	19.83 ± 0.34	49.57 ± 0.24

and	1.50	3.60±0.10	3.40± 0.40
50% castor	2.00	4.90± 0.10	4.00± 0.00

TABLE 3.2

TABLE OF QUANTITATIVE PHYTOCHEMICALS OF CASTOR AND ROSEMARY PLANT EXTRACT

Extract sample	Alkaloids (%)	Flavonoids (%)	Saponins (%)	Lipids (%)
Castor oil	0.28±0.11%	0.03± 0.12%	0.19±0.12%	4.00±0.10%
Rosemary	0.25± 0.11%	0.02± 0.11%	2.40±0.10%	3.20± 0.24%

3.2. Anti-bacterial Fabrics Characterization

The results for qualitative assessment of microbiological activity on cotton fabric with in different concentration of rosemary oil and castor oil by The AATCC test method 147 agar well diffusion test were shown in (Table 3.3). The antibacterial activity was measured by the zone of inhibition (mm). Totally two bacterial strains, gram-positive (*S. aureus*) and gram-negative bacteria (*E. coli*) were used in this investigation. The extract solution was prepared with in 1mg/mL, 1.5mg/ml and 2mg/ml concentration. 1mg/ml, 1.5mg/mL and 2mg/mL rosemary extract treated fabrics showed the zone of inhibition in 1.20 ± 0.17 mm, 2.07 ± 0.12 mm, and 3.00 ± 0.00 mm respectively for gram-positive bacteria and similarly 1.00 ± 0.00 mm 1.87 ± 0.12mm and 2.63 ± 0.21mm for gram-negative bacteria respectively. Castor seed extract treated cotton fabrics showed the zone of inhibition in 2.20 ± 0.20mm, 3.00 ± 0.10mm, and 3.93 ± 0.12mm respectively for gram-positive bacteria and 2 ± 0mm, 2.77 ± 0.25mm, 3.30 ± 0.26mm for gram negative bacteria respectively. Generally, the bigger the zone, the higher in it's the antibacterial activity [24]. The results of the antibacterial test showed that the methanolic extracts of rosemary and castor were more energetic against gram-positive bacteria (*S. aureus*). On the opposing, the gram-negative bacteria were slightly energetic (*E. coli*). 50% of rosemary and castor oil exhibited better growth of inhibition against both gram-positive and gram-negative 4.90± 0.10 and 4.00± 0.00 respectively at 2mg/ml. The rosemary generally showed low action against test organisms (3.00± 0.00 and 2.63 ± 0.21 at 2 mg/ml concentration) for gram-positive and gram-negative bacteria, compared to the castor and the combination of the two.

TABLE 3.3

ANTIBACTERIAL EFFICIENCY OF THE EXTRACTS FINISHED COTTON KNITTED FABRIC WITH IN DIFFERENT CONCENTRATION

Samples	Concentration (mg/ml)	Zone of inhibition (mm)	
		<i>Staphylococcus aureus</i>	<i>Escherichia coli</i>
Rosemary	1.00	1.20± 0.17	1.00 ± 0.00
	1.50	2.07± 0.12	1.87± 0.12
	2.00	3.00± 0.00	2.63 ± 0.21
Castor	1.00	2.20 ± 0.20	2.00 ± 0.00
	1.50	3.00± 0.10	2.77± 0.25
	2.00	3.93± 0.12	3.30 ± 0.26
50% rosemary	1.00	3.00± 0.00	3.17± 0.15

The extract showed increasing inhibitory activity with an increase in concentration (1 to 2mg/ml). However, the activity of current extract is less than the literature reported result. Castor extract exhibited maximum zone of inhibition against *S. aureus* (14 ± 1.83 mm) followed further, it was observed that, castor (12 ± 1.63 mm) extracts showed greater zone of inhibition against *E. coli* [25]. But there is a difference with-in extraction solvent and the literature report was reported in antibacterial activity of its own extract but the current study was reported as the efficiency of an extract with-in the fabric. During application of extract into fabric some antimicrobial compounds are damaged. Results of a minimum inhibitory concentration of rosemary and extract were 1mg/ml, with respect to all the test bacteria. The variation in the antibacterial activity of the plant extracts can be attributed to inoculum size, type of media used, type of solvent used for extraction, extraction procedure, incubation time and temperature, part of the plant used and its time of collection, method of extraction procedure, incubation time and temperature, method of antimicrobial assay and strain activity [25].



Fig 3.1 Minimum inhibition zones for antibacterial activity of castor seed, rosemary and the combination of two extracts respectively against *S. aureus*



Fig 3.2 Minimum inhibition zones for antibacterial activity of castor seed, rosemary and the combination of two extracts respectively against *E. coli* against *S. aureus*

Meanwhile, among the bacterial species, higher inhibition zone was observed against Gram positive (*Staphylococcus aureus*) bacteria than Gram negative (*Escherichia coli*) bacteria. Similar observations have been made by many researchers who reported that Gram positive bacteria are more susceptible to plant's extracts as compared to Gram negative bacteria [25], [26], [27]. The inhibition zone for antibacterial activity of all extracts against *staphylococcus aureus* and *Escherichia coli* with the different concentrations are summarized in the Fig 3.3. There was a significant difference between each concentration that means the P value is less than 0.05. Inhibition zone of both *S. aureus* and *E. coli* on the fabric has a significant mean difference at a different concentration which is shown in the ANOVA multiple comparisons table. As shown in ANOVA Table 3.5 below the concentration of extract has a significant effect on the

inhibition zone of both bacteria (*S. aureus* and *E. coli*). The analysis was done by using ONE WAY ANOVA and gives significant value for each bacteria in which for *S. aureus* $F = 104.571$ and $Sig (P) = 0.000$ and for *E. coli* $F = 453.571$ and $Sig (P) = 0.000$. The above ANOVA analysis shows that the result is due to the effect of concentration in which mean square between groups is greater than mean square within groups.

TABLE 3.4

MULTIPLE COMPARISONS OF ANTIBACTERIAL ACTIVITY OF TREATED SINGLE JERSEY COTTON FABRIC

Dependent Variable	(I) conc	(J) conc	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
zone of inhibition to <i>s.aureus</i>	1mg/ml	1.5mg/ml	-.86667*	.09813	.000	-1.1678	-.5656
		2mg/ml	-1.80000*	.09813	.000	-2.1011	-1.4989
	1.5mg/ml	1mg/ml	.86667*	.09813	.000	.5656	1.1678
		2mg/ml	-.93333*	.09813	.000	-1.2344	-.6322
	2mg/ml	1mg/ml	1.80000*	.09813	.000	1.4989	2.1011
		1.5mg/ml	.93333*	.09813	.000	.6322	1.2344
zone of inhibition to <i>s.coli</i>	1mg/ml	1.5mg/ml	-.86667*	.11222	.001	-1.2110	-.5224
		2mg/ml	-1.63333*	.11222	.000	-1.9776	-1.2890
	1.5mg/ml	1mg/ml	.86667*	.11222	.001	.5224	1.2110
		2mg/ml	-.76667*	.11222	.001	-1.1110	-.4224
	1mg/ml	1.5mg/ml	1.63333*	.11222	.000	1.2890	1.9776
		2mg/ml	1.5mg/ml	.76667*	.11222	.001	.4224

*. The mean difference is significant at the 0.05 level

TABLE 3.5
ANOVA ANALYSIS

ANOVA		Sum of Squares	df	Mean Square	F	Sign.
Zone of inhibition to <i>s.aureus</i>	Between Groups	4.880	2	2.440	104.571	.000
	Within Groups	.140	6	.023		
	Total	5.020	8			
Zone of inhibition to <i>e.coli</i>	Between Groups	7.056	2	3.528	453.571	.000
	Within Groups	.047	6	.008		
	Total	7.102	8			

3.3 Scanning Electron Microscope Analysis

The morphological analysis of cotton knitted fabric which is treated and untreated with the methanolic extract samples were employed by using Scanning Electron Microscope (SEM). SEM study was employed to investigate the surface morphology of samples and the obtained images are shown in (Fig 3.4). SEM images in Figure show that the surface morphology of the treated and untreated fabrics showed small differences in surface morphology is due to the presence of uniform coating of the finishing agent on the fibres. The untreated fabric was a little bit smooth, but the treated fabric has some cracking and the fibrillar structure also rupture. The surface of a substrate was changed due to a chemical process of the fibre and the antimicrobial compound from the extract.

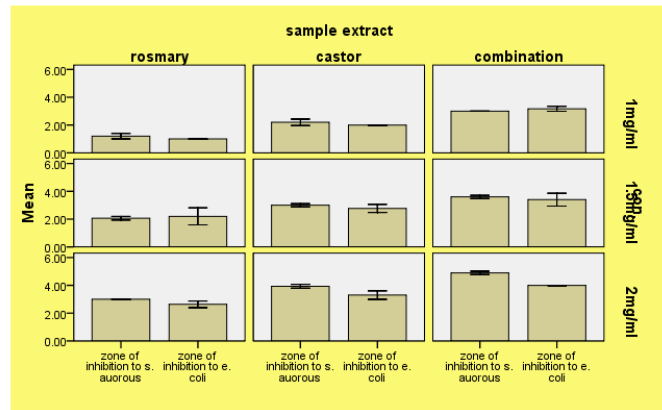


Fig 3.3 Antibacterial activity of different extract with different concentration summary

3.4. Physio-mechanical Characterization

The physio-mechanical characterization was done for the untreated and treated fabrics (minimum inhibitory concentration fabric). Some physio-mechanical properties of the fabrics are wicking property, tensile strength, wash durability, air permeability etc.

3.4.1. Anti- bacterial Finish Wash Durability Test

The results of the wash durability of the fabrics treated with rosemary extract alone retained its antimicrobial activity up to 2 wash cycles, which gradually decreased and became nothing after 3 washes. Fabrics treated with castor and combination of two extract reflected excellent ratings. The wash durability of fabric treated with castor oil and both extract was retained the effect up to 4 wash cycle. Most literatures reported about natural plant dye was retained the effect up to the maximum of 6 wash cycle. This result is somehow in good agreement with literatures reported values [28], [29]. The wash durability of the treated fabric was summarized below in Table 3.6.

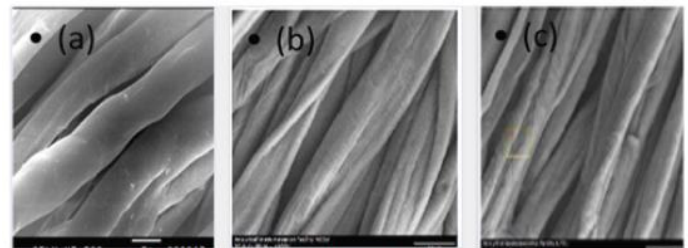


Fig 3.4 SEM images for cotton knitted fabric (a) untreated, (b) treated with rosemary, (c) treated with castor

Treated knitted cotton fabric with rosemary extract still showed weakness in washing durability compared to the literatures for direct industrial application. The fabric samples treated with rosemary had a ratting maximum of 2wash cycle. This weakness in fastness against washing is due to the poor bond or incorporation the plant extract material into the cotton fibers.

TABLE 3.6

Sample extract	Wash cycle	Zone of inhibition (mm)	
		Staphylococcus aureus	Escherichia coli
		Rosemary	2nd
	4th	No zone of inhibition	No zone of inhibition

	6th	No zone of inhibition	No zone of inhibition
Castor oil	2nd	1.5	1
	4th	0.5	0.5
	6th	No zone of inhibition	No zone of inhibition
Combination of two	2nd	1.5	1
	4th	1	No zone of inhibition
	6th	No zone of inhibition	No zone of inhibition

Air permeability (mm/s)	985	987	986	988
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The air permeability of knitted cotton fabrics as control and cotton treated with rosemary, castor and combination of two was studied and the result illustrated in the above table. The results were almost similar as shown in the Fig 3.6. The extract has insignificant effect on the air permeability of the cotton fabric.

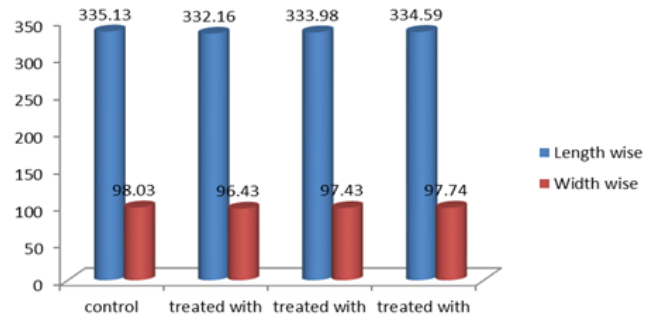


Fig 3.5 Tensile strength of the treated and untreated cotton knitted fabrics length and width wise

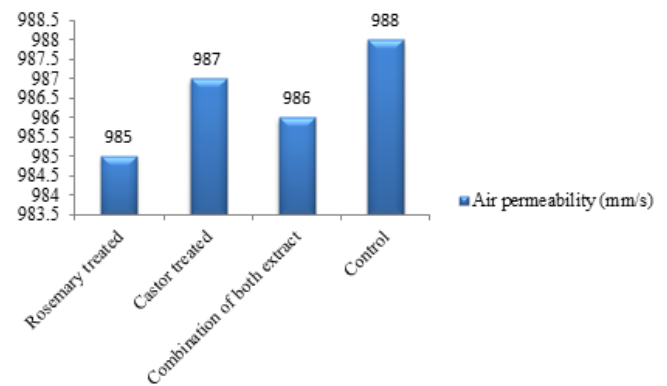


Fig 3.6 Air permeability of a sample fabric

3.4.2. Wicking property

The untreated cotton fabric is showing wicking time, 15 minutes, with consistency in comparison to its performance at treated fabric with castor oil, where the wicking had stopped after 12 minutes, in each minute 0.4 inches are wetted. Similarly, the wicking time of treated fabric with rosemary extract have a related result with the castor oil treated fabric, which means 12 minutes. The wicking time of the treated and untreated knitted fabric have a small change this indicates that the fabric treated with the extract has shown water repellency. This might be due to the hydrophobic nature of the extract which forms a thin coating of film on the surface of the fabric.

3.4.3. Tensile Strength

The tensile strength of untreated single jersey knitted fabric made from 100% cotton has maximum breaking force between 335 N and 335.2 N with an average of 335.13 ± 0.11 N along the length of the fabric and between 98 N and 98.1 N with an average of 98.03 ± 0.06 N across the width of the fabric. There is very negligible change in the tensile strength of the treated and untreated cotton fabric for all in addition to the combination of the two. Literatures also suggest that natural antimicrobial agent have checked some success in terms of efficiency and wash durability with minimal effect on the tensile strength of fabrics. [30] The tensile strength of the treated and untreated fabric was summarized in fig 3.5.

3.4.4. Air Permeability Test

In the present study, the air permeability of single jersey knitted fabric made from 100% cotton was determined by using ASTM D737-18 method. Untreated and treated with rosemary, castor and combination of both extract fabric samples were conditioned in a conditioning room at 65 ± 2 % relative humidity and 20 ± 2 °C for 24 hours. The instrument was calibrated before the experiments using a calibration check plate. The instrument calibrates the testing area at one square meter per second, pressure 200 Pa and adjusts the air flow. After calibration of the plate the test sample was placed on the ground and the test was done each sample and the results of air permeability of sample are recorded in table 3.7

TABLE 3.7
FABRICS SAMPLE AIR PERMEABILITY VALUES

Fabric sample	Rosemary treated	Castor treated	Combination of both extract	Control
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4 CONCLUSION

Although generally, all classes of textiles materials are always affected with microorganisms and degraded by time. So the attempt was made to find a solution for improving the durability of the textile fabric. In this work the cotton fabrics were finished with castor and rosemary extracts and their effectiveness as an antibacterial finish agent were evaluated. Accordingly, the plant extract of rosemary and castor have significantly reduced the growth of bacteria. The textile materials that were treated with the extract showed growth inhibitory activities against *Staphylococcus aureus* and *Escherichia coli*. Rosemary extract coated fabric was found to be less efficient compared to castor this may be due to the intrinsic property of the crude extract or the less binding property of compounds and/or due to sensitivity of the compounds to thermal destruction during application and processing the fiber. The results for qualitative assessment of microbiological activity on cotton fabric with different concentration of rosemary oil and castor oil by agar well diffusion test were more energetic against gram-positive bacteria (*S. aureus*) as compared to untreated fabrics. On the other hand, the gram-negative bacteria were more unaffected (*E. coli*). The extract showed increasing inhibitory activity with an increase in concentration of the extracts of rosemary and

castor oils. It is confirmed that finishing the cotton fabric with the extracts of rosemary and castor seeds reduces the growth of microorganisms and avoids the degradation of the cotton fabric. Antibacterial durability of treated fabrics was limited to four washes and it is considered as a temporary finishing it can be used for low usage garments. All the fabric samples had a rating maximum of 4 wash cycle. Physico-mechanical properties such as wicking property, air permeability, and tensile strength were evaluated and found that the properties have insignificant changed by treating the fabric with the extracts of rosemary and castor seeds. It is thus concluded that the fabrics finished with castor seed and rosemary extracts showed positive enhancement in all the properties. Hence, both extracts can be effectively and efficiently used for antibacterial finish of cotton fabric for commercial purpose as an eco-friendly finish.

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REFERENCES

- [1] Sheila Shahidi and Jakub Wiener Antibacterial Agents in Textile Industry, Antimicrobial Agents, Varaprasad Bobbarala, Intech Open, Available from: <https://www.intechopen.com/books/antimicrobial-agents/antibacterial-agents-in-textile-industry>, 2012.
- [2] Jain, A. and Tesema, A, "Development of antimicrobial textiles using zinc pyrithione", Research Journal of Textile and Apparel, Vol. 21 No. 3, pp. 188-202. 2017.
- [3] W D Schindler P J Hauser, Chemical Finishing of Textiles (Woodhead Publishing Series in Textiles) 1st Edition, Woodhead Publishing Series in Textiles (Book 32), 224 pages, 2004 ISBN-10: 1855739054, ISBN-13: 978-1855739055.
- [4] G.Thilagavathi, S.Viju, Antimicrobials for protective clothing, Science Direct, Woodhead Publishing Series in Textiles, Chapter 16, Pages 305-317, 2016.
- [5] Deepti Gupta, Antimicrobial treatments for textiles, Indian Journal of Fibre & Textile Research, Vol. 32, pp. 254-263, 2007.
- [6] Beata Gutarowska and Andrzej Michalski. Microbial Degradation of Woven Fabrics and Protection Against Biodegradation, Woven Fabrics, Han-Yong Jeon, IntechOpen, 2012.
- [7] Mulinacci, N., Innocenti, M., Bellumori, M., Giaccherini, C., Martini, V., & Michelozzi, M. Storage Method, Drying Processes and Extraction Procedures Strongly Affect the Phenolic Fraction of Rosemary Leaves: An HPLC/DAD/MS study. *Talanta*, 85(1), 167–176, 2011.
- [8] Rožman, T., & Jeršek, B. Antimicrobial activity of rosemary extracts (*Rosmarinus officinalis* L.) against different species of *Listeria*. *Acta Agriculturae Slovenica*, 93(1), 51–58, 2009.
- [9] Pedro Mena 1OrCID, Martina Cirilini 1OrCID, Michele Tassotti 1, Kelli A. Herrlinger 2, Chiara Dall'Asta 1 and Daniele Del Rio 1, Phytochemical Profiling of Flavonoids, Phenolic Acids, Terpenoids, and Volatile Fraction of a Rosemary (*Rosmarinus officinalis* L.) Extract, *Molecules*, 21(11), 1576, 2016.
- [10] Mena, P.; Calani, L.; Dall'Asta, C.; Galaverna, G.; Garcia-Viguera, C.; Bruni, R.; Crozier, A.; Del Rio, D. Rapid and Comprehensive Evaluation of (poly) phenolic Compounds in Pomegranate (*Punicagranatum* L.) Juice by UHPLC-MSn. *Molecules*, 17, 14821–14840, 2012.
- [11] Momoh, A.O*, Oladunmoye, M.K. and Adebolu, T.T, Evaluation of the Antimicrobial and Phytochemical Properties of Oil from Castor Seeds (*Ricinus communis* Linn) *Bulletin of Environment, Pharmacology and Life Sciences*, Volume 1 [10] 21 – 27, 2012.
- [12] Zarai, Z., Chobba, I.B., Mansour, R.B. et al. Essential oil of the leaves of *Ricinus communis* L.: In vitro cytotoxicity and antimicrobial properties. *Lipids Health Dis* 11, 102, 2012.
- [13] Jumba, B.N., Anjili, C.O., Makwali, J. et al. Evaluation of leishmanicidal activity and cytotoxicity of *Ricinus communis* and *Azadirachta indica* extracts from western Kenya: in vitro and in vivo assays. *BMC Res Notes* 8, 650, 2015
- [14] Tyagi N, Tyagi M, Pachauri M, Ghosh PC, Potential therapeutic applications of plant toxin-ricin in cancer: challenges and advances. *Tumour Biology*, 36(11):8239-46, 2015.
- [15] Shazia Mehtab ; Manisha Gahlot ; Anita Rani, Application of *Justicia adhatoda* L. leaf extract as antibacterial finish on cotton fabric, *International Journal of Basic and Applied Agricultural Research* 2016 Vol.14 No.2 pp.225-228 ref.11, 2016.
- [16] Chukwuma S. Ezeonu¹ and Chigozie M. Ejikeme², Qualitative and Quantitative Determination of Phytochemical Contents of Indigenous Nigerian Softwoods, Hindawi Publishing Corporation *New Journal of Science* Volume 2016, Article ID 5601327, 9 pages, 2016.
- [17] C. M. Ejikeme, C. S. Ezeonu, and A. N. Eboatu, "Determination of Physical and Phytochemical Constituents of Some Tropical Timbers Indigenous to Niger Delta Area of Nigeria," *European Scientific Journal*, vol. 10, no. 18, pp. 247–270, 2014.
- [18] B. A. Boham and A. R. Kocipai, "Flavonoids and Condensed Tannins from Leaves of Hawaiian *Vaccinium vaticulatum* and *V. calycinium*," *Pacific Science*, vol. 48, pp. 458–463, 1994.
- [19] AOAC, Official Methods of Analysis, Association of Official Analytical Chemists, Washington, DC, USA, 14th edition.
- [20] Lagouri, V., Bantouna, A. and Stathopoulos, P, "A comparison of the antioxidant activity and phenolic content of nonpolar and polar extracts obtained from four endemic Lamiaceae species grown in Greece". *Journal of Food Process*. vol. 34, 872–886, 2010.
- [21] Hiwot, T. "Investigation of the chemical composition, characterization and determination of energy content for renewable energy source (biodiesel) produced from non-edible Ethiopian seeds particularly castor seed (*Ricinus communis*) using homogeneous catalysis", *International Letters of Chemistry, Physics and Astronomy*, vol. 37, 63–74, 2014.
- [22] Turkmen, N., Sari, F. and Velioğ̃ Lu, S., "Effects of extraction solvents on concentration and antioxidant activity of black and black mate tea polyphenols determined by ferrous tritrate and Folin-Ciocalteu methods". *Food Chemistry*, vol. 99, 835–841, 2006.
- [23] Anokwuru, Chinedu & Anyasor, Godswill & Olusola, Ajibaye & O., Fakoya & P., Okebugwu. Effect of Extraction Solvents on Phenolic, Flavonoid and Antioxidant activities of Three Nigerian Medicinal Plants. *Nature and Science*, 2011;9(7). 9. 7, 2011.
- [24] Last, H., Blech, B.A., Dahringer, J., Seibel, S., Bioactive Fibre Products. U.S. Patent No, US 7858111 B2, 2010.
- [25] Byadgi, S., Kulloi, S., & Venugopal, C. Phytochemical Screening and Antimicrobial Activity of Plant Extracts for Textile Applications. *International Journal of Biochemistry Research & Review*, 20(3), 1–10, 2018.
- [26] Joana Monte, Ana C. Abreu, Anabela Borges, Lúcia Chaves Simões, Manuel Simões, "Antimicrobial activity of selected

- phytochemicals against *Escherichia coli* and *Staphylococcus aureus* and their biofilms. *Journal of Chemical and Pharmaceutical Research*, 3(2), 473–498, 2014.
- [27] Raho G. Bachir, Benali M., “Antibacterial activity of the essential oils from the leaves of *Eucalyptus globulus* against *Escherichia coli* and *Staphylococcus aureus*”, *Asian Pac J Trop Biomed*; 2(9), 739–742, 2012.
- [28] Rossi, T., Araújo, M. C., Brito, J. O., & Freeman, H. S. “Wash Fastness of Textile Fibers Dyed with Natural Dye from *Eucalyptus* Wood Steaming Waste”, *International Journal of Chemical, Molecular, Nuclear, Materials and Metallurgical Engineering*, vol.9, page 7, 2015.
- [29] Sayed, U. “Application of Essential Oils for Finishing of Textile Substrates”. *Journal of Textile Engineering & Fashion Technology*, 1(2), 42–47, 2017.
- [30] Sathianarayanan M P Bhat N V, Kokate S S, Walunj V E, “Antibacterial finish for cotton fabric from herbal products”, *Indian Journal of Fibre & Textile Research*; Vol. 35 PP 50-58, 2010.