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EVALUATION OF ANTIMICROBIAL ACTIVITY OF FLAX FIBRES IMPREGNATED WITH ALOE GEL EXTRACT

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ABSTRACT

The natural textile fibres are prone to be attacked by various micro organisms and provide room for their growth and infestation. Human being can easily be crops infected by these micro organisms. Natural antimicrobial and bioactive agents are available in plenty that will not harm the skin and are found to be very effective against both gram positive and gram negative bacteria. To minimise the risk factor of microbial infections, the study was aimed to provide the natural flax waste fibres an antimicrobial efficiency that can stop the microbial infestation and their growth completely. Waste fibres from various spinning mills are either used in low grade fabrics or as filler into the beddings etc. which can easily attacked by microorganism which are available in environment everywhere. Fibres were treated with alovera gel extract in 10 different concentrations from 100 to 10% and tested for its microbial efficacy against Staphylococcus aureus, Escherichia coli and fungi. The samples were found to be very effective against these micro organisms.

INTRODUCTION

The natural fibres are susceptible of microbial infestation. The inherent properties of natural fibres give them the conditions required for their growth.

The micro-organisms like bacteria, fungi etc. are present in environment everywhere, some of them are harmful to human while others are not. These can survive easily in textile material, and in case of natural fibres they find more favourable conditions. 'Textiles have long been recognized as media to support the growth of micro-organism such as bacteria and fungi' **(Maleknia *et al.*, 2010)**. To avoid infection caused by such micro-organism some kind of antibacterial treatment is needed for textile material. Microbes that damage textiles are also a threat to health of human beings.

A huge number of antimicrobial agents are available in market. Ecofriendliness is gaining attention worldwide and due to which bioactive agents are found to be in vogue now a days. Some effective bioactive agents are neem, aloevera, tulsi, clove, and eucalyptus etc.

The Aloevera (*Aloe barbadensis Miller)* is found to have medicinal and antimicrobial properties and have been used long for wound healing, skin care treatments etc. **Joshi *et al,* 2009**. It has been found very effective against the growth of micro-organisms that is bacteria, both gram positive and gram negative, fungi **(Hamman 2008)**, as it does not support the growth of these micro-organisms.

Acemannan found in aloe vera possess the "immunomodulation, antibacterial, antifungal and antitumor properties". **(Joshi *et al.*, 2009)**

The present study aims at development of an effective finish having antimicrobial and herbal potentials which can be used for various textile applications. Thouroughly cleaned and bleached flax fibres were immerses into the methenolic extract of aloevera gel and tested for antimicrobial activity as per standard test methods.

MATERIALS AND METHODS

Finish extraction

Aloevera leaves were processed to get aloe gel by using hand filleted method. Inner gel was removed with the help of knife and avoiding yellow sap, also called latex which in found next to the rind of the leaf. The clear gel thus collected was crushed and converted to solvent form using methanol as solvent and citric acid (100% concentrated) as cross linking agent.

Application: The extracted finish was applied in 10 different concentrations from 100 to 10 percent on to the fibre samples by immersing directly into the prepared finish till complete wetting of samples. Samples were then removed from liquor and dried in hot air oven at 80^0C.

Assessment

Evaluation of antimicrobial activity and Comparison with cotton filler of commercial products

Antibacterial and antifungal activities of treated samples were tested as per standard test methods of ENISO (5405: 2004-

Agar Diffusion Test) and AATCC (147: 2004- Parallel Streak Method) and AATCC (30: 1993- Antifungal activity, assessment on textile materials). The samples were also compared with traditional cotton filler used in commercial products.

Test Media: Nutrient Agar was used to find out the bacterial growth and PDA for finding fungal growth.

Note: All the Preparation and Activities were performed in a Laminar Flow Chamber to avoid any kind of contamination for getting best results.

Evaluation of antibacterial activity

Bacterium Cultures

Two cultures of Staphylococcus Aureus (gram positive) and Escherichia Coli (gram negative) were selected to perform test for assessment of antibacterial activity.

Preparation of Agar Plate

25 ml of prepared media (Nutrient Agar) was poured in sterile petri disc, allowed to cool.

ENISO 5405: 2004- Agar Diffusion Test

Test specimens (25 mm. diameter) in circular shape were prepared. 200 µm of bacterial culture was transferred on to the agar plate and spreaded throughout the surface using a L-shaped spreader. Specimen was imprinted directly on to the inoculated agar plate, using sterile forceps, in three test replication per concentrations. Petri plates were closed, sealed and incubated for 18-24 hrs at 37^0C.

The level of antibacterial activity was assessed by examining the extent of bacterial growth in the contact zone between the agar and the test specimen. Total inhibition zone across the specimen was measured using a scale.

AATCC 147: 2004- Parallel Streak Method

Using a 4 mm inoculating loop, one loopful of the inoculum was transferred to the surface of agar plates, making five parallel streaks on the central area of a plate without a refilling of loop. Test specimens (25×50) were cut with a rectangular die and placed onto inoculated petri plate transversely across the five inoculum streaks. Petri plates were incubated for 18-24 hr at 37^0C. Incubated plates were examined for interruption of growth along the streaks of inoculum beneath the specimen and for a clean zone of inhibition beyond its edge. Zone diameter along a streak on either side of the test specimen was measured using a scale.

Evaluation of Antifungal activity

AATCC 30-1993: Antifungal activity, Assessment of textile material: mildew and rot resistance of textile material (Humidity jar method)

500ml conical flask containing of Potato Dextrose Agar was prepared and sterilized at 121^0C for standard time. It was then allowed to cool. The samples were transferred aseptically into the conical flasks respectively. These were kept at room temperature for 3 days. Then the growth of fungi in the conical flask was observed after 3 days.

RESULTS AND DISCUSSION

EN ISO 20645:2004 - Agar Diffusion Method

Inhibition zones were calculated using the following expression:

$H = (D - d) \div 2$, where:

H- Inhibition zone in mm
D- Total diameter of specimen and inhibition zone in mm
d- Diameter of specimen in mm.

Table 1 Mean H values

S.No.	%	Mean H Value (S. Aureus)	Mean H Value (E. Coli)	Control
1	100	7.25	7.3	
2	90	6.75	5.9	
3	80	6.625	5.375	
4	70	4.875	5.125	
5	60	4.475	4.875	
6	50	3.625	3.8	
7	40	3.075	2.45	0.00
8	30	2.225	1.45	
9	20	2.675	0.875	
10	10	0	0.75	

According to test method, >1-0 mm inhibition zones and no growth under specimen were accepted as effective, whereas 0 mm inhibition zone and slight growth were evaluated as limited effect

AATCC147:2004 – Parallel Streak Method

Evaluation of antibacterial activity (AATTC Test Method 147-2004). The average width of a zone of inhibition along a streak on either side of the test specimen was calculated using the following equation:

W= (T - D)/2 where:
W is width of clear zone of inhibition in mm
T is total diameter test specimen and clear zone in mm
D is diameter of the test specimen in mm.

Table2 Mean W Values

S.No.	%	Mean W Value (S. Aureus)	Mean W Value (E. Coli)	Control
1	100	**7.625**	**6.875**	
2	90	**6.375**	**5.975**	
3	80	**5.375**	**5.1**	
4	70	**4**	**5**	
5	60	**3.25**	**4.625**	
6	50	**3**	**3.6**	0.00
7	40	**2.25**	**2.4**	
8	30	**2**	**1.325**	
9	20	**0.625**	**0.875**	
10	10	**0**	**0.75**	

Absence of bacterial colonies under the specimen in the contact area is considered as an acceptable antibacterial activity for parallel streak method.

Assessment of Antifungal Activity- AATCC30:1993 – (Antifungal activity, Assessment of textile material: mildew and rot resistance of textile material: Humidity Jar Method)

No growth of fungi was found after three days in the flask containing gel treated specimens. This indicates that the gel treated samples possess desirable anti fungal activity but growth of fungi was found in the flasks of test control.

Fig. 1 Presence of zone of inhibition on treated Samples (ENISO 20645)

Fig. 2 Presence of zone of inhibition on treated Samples (AATCC 147)

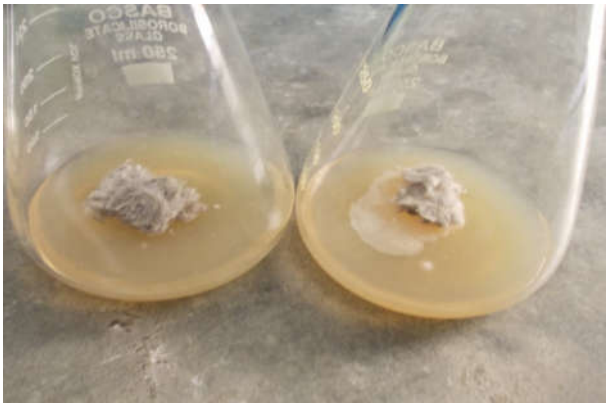

Fig. 3 Treated sample showing no fungal growth and test control (ENISO 20645)

Treated samples of aloe vera gel were tested against gram negative and gram positive bacteria and fungi. A visible zone of inhibition in each sample was found which was within the desired limits of sufficient antimicrobial activity. No fungal growth was noticed in humidity jars after 3 days which shows that the samples were found to have effective antimicrobial potential against gram positive and gram negative bacteria viz. Staphylococcus Aureus and E. Coli and against fungi as well.

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