

# Antimicrobial activity of chitosan and its derivatives exhausted cotton fabrics as ecofriendly antimicrobial agents

## Abstract

Development of the antimicrobial activity of cotton fabric using chitosan, N-(2-Hydroxy) propyl-3-trimethyl-ammonium chitosan chloride (HTACC), and N-methylol-acrylamide-N-(2-hydroxy) propyl-3-trimethyl-ammonium chitosan chloride (NMA-HTACC) was investigated. Moisture absorbency is an important property of cotton fabric due to which bacteria can easily affect the cotton fabric and lowers the quality of it. Chitosan, HTACC, and NMA-HTACC were applied on cotton fabric by exhaustion method and observed antibacterial activity against *Escherichia coli* and *Staphylococcus aureus*. Among the finished fabrics, NMA-HTCC treated cotton fabric showed over 93% of bacterial reduction against *E. coli* and over 97% of bacterial reduction against *S. aureus* even after 30 home launderings. This is due to the strong attachment ability of NMA-HTACC to cotton fabric and better solubility in aqueous solution and the presence of positive charge of the nitrogen atom in its structure in all conditions. For this reason, NMA-HTACC can be applied to develop cotton fabric as anti-bacterially active which makes it suitable for medical purposes by minimizing the chances of growth and multiplication of bacteria.

**Keywords:** chitosan, antimicrobial activity, cotton fabric, chitosan derivatives

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## Introduction

Biodegradability, renewability, moisture absorbency, comfortability etc of natural cellulose-based cotton fabric make it more suitable for different purposes especially healthcare as well as medical purposes than any other fabrics. Also due to having no environmental impact its demand is increasing day by day. Besides these cellulose-based cotton fabric possess an important inherent limitation is microbial attack which limits the direct use of it in medical sector.<sup>1</sup> To minimize the microbial degradation and to improve the textile properties many researchers finished the cotton fabric with different chemical agents such as triclosan, metal and their salts, phenols and organo metallics etc. which show good antimicrobial activity on cotton fabric but have a great concern for both environment and human health. That's why an ideal anti-microbial textile finishing agent need to be safe, non-toxic, environmentally benign and durable to wash, so textile researchers should prefer to apply eco-friendly finishing agents on cotton fabrics. Chitosan and its water soluble derivatives such as N-(2-Hydroxy) propyl-3-trimethylammonium chitosan chloride (HTACC) and N-methylolacryl amide-N-(2-hydroxy) propyl-3-trimethylammonium chitosan chloride (NMA-HTACC) have a positive charged nitrogen atom in its structure which has a possibility to damage the negatively charged bacterial cell wall.<sup>2</sup> Hence, the major objectives of this study are to:

- 1) Develop the antibacterial activity of the finished cotton fabric with chitosan and its derivatives (HTACC and NMA-HTACC)
- 2) Compare the antibacterial activity of Chitosan, HTACC and NMA-HTACC finished cotton fabrics
- 3) Manage the prawn shell waste through a value-added product in textile finishing.

## Experimental

### Materials

Prawn shell was collected from Mongla (near Sundarban forest), Khulna, Bangladesh. Cotton fabrics were collected from Keya Spinning Mills Ltd., Bangladesh. *Escherichia coli* and *Staphylococcus aureus* bacteria are collected from "Biochemistry and Molecular Biology" department of Rajshahi University. Hydrochloric acid, sodium hydroxide, acetone, methanol etc. were purchased from Sigma Aldrich, Merck, BDH etc and used without further purification.

### Methods

**Fabric preparation:** Cotton fabrics were washed with 0.2%  $\text{Na}_2\text{CO}_3$  solution at 75°C for 30 min in a beaker in the fabric to liquor ratio of 1:50. After washing thoroughly with distilled water and drying at 60°C, the fabric was stored in a desiccators.<sup>3</sup>

**Finishing process:** First, HTACC, NMA-HTACC were dissolved in distilled water and whereas chitosan was dissolved in 2% (v/v) aqueous acetic acid solution (maintaining pH-5.5) by stirring for 2-3 h and sonicated for 2h. Then 50 $\mu\text{L}$  of Triton-X was added to the liquor to improve the wettability.<sup>4,5</sup>

In Exhaustion method, the fabrics were immersed in the prepared solution of various concentrations and were sonicated for 1hr at 70°C. The samples were then dried at 100°C for 10 min and cured at 170°C for 5 min. These treated cotton fabrics were dried at an ambient condition.<sup>6</sup>

### Characterization of finished fabrics

**Moisture absorption study:** The finished and washed cotton fabrics were placed on a humidity chamber at room temperature (70°C)

for a week where humidity was maintained at the saturation level.<sup>7</sup> After that the fabrics were analyzed by moisture analyzer using the following formula:

$$\text{Moisture Content, } (\%) = \frac{wf-wi}{wi} \times 100$$

Where  $W_f$  is the weight of wet sample and  $W_i$  is the weight of dry sample.

**Swelling behavior study:** Swelling behaviour of the finished and washed cotton fabric were determined by dipping into water at room temperature for 24 h. The percent swelling was calculated according to following equation.<sup>8</sup>

$$\text{Percent Swelling, } (Ps) = \frac{wf-wi}{wi} \times 100$$

Where  $W_i$  is the initial weight of fabric and  $W_f$  is the final weight of fabric.

### Water vapor permeability test

The water vapour permeability of the samples was measured using the cup method, according to ASTM E96 (procedure B) testing standard. The water vapour permeability of the finished and non-finished fabrics can be calculated by the following equation:

$$WVP = \frac{24 \times M}{A \times T} \text{ g / m}^2 / 24h$$

Where, M is the loss in mass of water (g); T, the time interval (h); and A, the internal area of the cup ( $\text{m}^2$ ). A was calculated using the following relationship:<sup>8</sup>

$$A = \frac{\pi d^2}{4} \times 10^{-4}$$

Where d is the internal diameter of the cup (mm).

### Assessment of antibacterial activity (quantitative test)

Bacterial percent reduction was calculated using the following formula.

$$R = (A - B) / A \times 100\%$$

Where R is percentage reduction, A is the number of bacteria in the broth inoculated with treated test fabric sample immediately after

inoculation i.e., at zero contact time and B is the number of bacteria recovered from the broth inoculated with treated test fabric sample after the desire contact period (18 hours).<sup>9</sup>

### Fourier transform infrared (FTIR) analysis

FTIR spectroscopic measurements were carried out in Central Science Laboratory, University of Rajshahi, Bangladesh (Shimadzu-8900, FTIR Spectrum, Kyoto, Japan). Samples were placed in the path of infrared beam of wave number in the range of 400-4000  $\text{cm}^{-1}$ .

### Scanning electron microscopy (sem) analysis

Scanning electron microscopic (SEM) studies of raw and finished cotton fabrics were carried out on an Electron Microscopy Machine (FEI Quanta Inspect, Model:S50).

### Thermal analysis

Thermo gravimetric Analysis (TGA) was performed using a Perkin Elmer simultaneous thermal analyzer (STA 8000, Germany). The tests were conducted between 30-600°C under an inert atmosphere (Nitrogen). The heating rate and the airflow rate were 20°C/min and 200ml/min respectively.

### Results and discussion

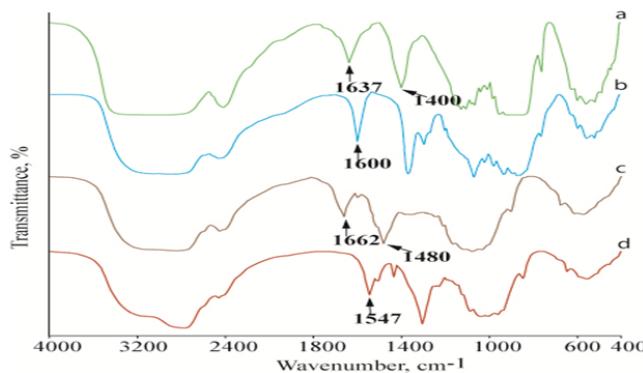
Moisture absorption, swelling test, WVP test, tensile strength, % elongation, wash fastness and biodegradation reports of washed and finished cotton fabrics are shown in Table 1. From the Table, finished cotton fabrics showed improved physicochemical properties. Due to the presence of hydroxyl groups (-OH) and channel-like structure finished fabric showed better moisture absorption. Also for having the good attachment capacity on cotton fabric, NMA-HTACC treated cotton fabric shows higher moisture absorption.<sup>7</sup> Similarly, in case of swelling test, swelling of control fabric was lower than others because of lacking the cross-linked channel structure to hold water for being swollen. In terms of WVP test, after applying Chitosan, HTACC, and NMA-HTACC on cotton fabric hydrophilic properties were aroused and channel-like structures are formed which attract water vapour and help to pass away to the outer side of the fabric to saturate the outer side. Due to more fibre reactivity, NMA-HTACC finished fabric showed higher WVP test value. Fabrics used for wound dressing should have neither higher nor lower WVP value. So for this NMA-HTACC treated cotton fabrics will be more suitable to provide moist wound condition.<sup>10</sup>

**Table 1** Physico-chemical tests of control and finished cotton fabric

Fabric type	Moisture absorption (%)	Swelling (%)	Water vapor permeability ( $\text{g}/\text{m}^2/\text{day}$ )	Tensile Strength (N)	% Elongation	Wash fastness (% reduction at 25 <sup>th</sup> wash)	Biodegradation (% wt loss at 28 <sup>th</sup> day)
Control	17.4	118.6	1371.3	290	9	Nil	78
Chitosan Finished	25.7	162.8	843.1	325	10	Nil	30
HTACC Finished	29.6	198.5	857.6	362	10.5	56	11
NMA-HTACC Finished	32.5	210.7	883.5	375	10.75	78	8

## FTIR analyses

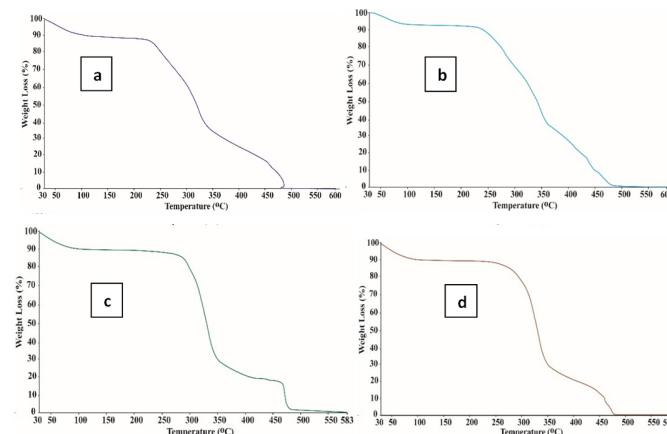
The FTIR spectra of washed and finished cotton fabrics are represented in Figure 1. FTIR spectra of washed and finished fabrics were nearly the same except the appearance of some new peaks on the finished fabric. The appearance of the peak at  $1600\text{cm}^{-1}$  for N-H bending of the primary amine (Figure 1b),  $1480\text{cm}^{-1}$  for C-H bending of trimethyl ammonium group (Figure 1c) and  $1547\text{cm}^{-1}$  for N-H bending of the secondary amide in the acrylamidomethyl group (Figure 1d) which confirm the attachment of chitosan, HTACC and NMA-HTACC on cotton fabric respectively.



**Figure 1** FTIR spectra of a) washed, b) Chitosan finished, c) HTACC finished and d) NMA-HTACC finished cotton fabrics.

## Thermal analysis

Figure 2 shows the thermal behaviour of washed and finished cotton fabrics. According to TGA thermograms, cotton fabrics undergo three stages of thermal degradation. There is a weight loss in the first stage between  $90\text{-}120^\circ\text{C}$ , due to dehydration. In the second stage between  $280\text{-}340^\circ\text{C}$ , rapid weight losses are observed for thermal degradation of the glycoside linkage and last stage, residual char is formed through weight loss which reaches to be a fixed weight. On the basis of initial decomposition temperature ( $T_i$ ), the thermal stability of washed and finished cotton fabrics show the following order: washed < Chitosan finished < HTACC finished < NMA-HTACC finished cotton fabrics.

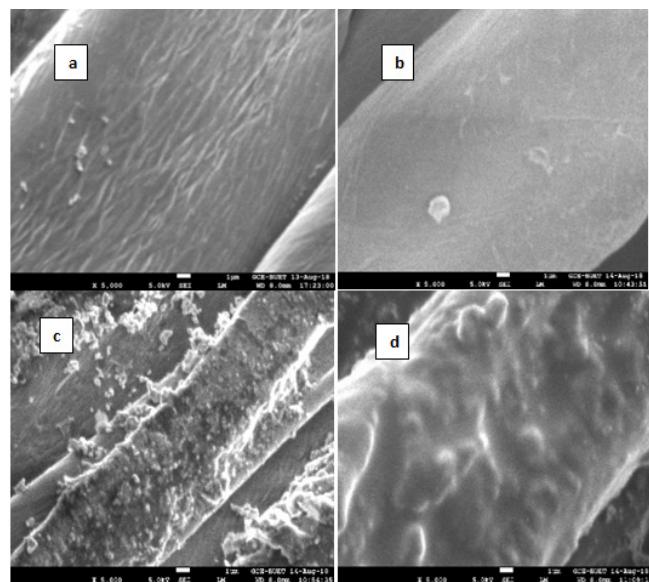


**Figure 2** TGA of a) Control, b) Chitosan finished, c) HTACC finished and d) NMA-HTACC finished cotton fabrics.

## Surface morphology

Surface morphology of washed and finished cotton fabrics was studied by Scanning Electron Micrographs. Figures 3b–3c) confirm

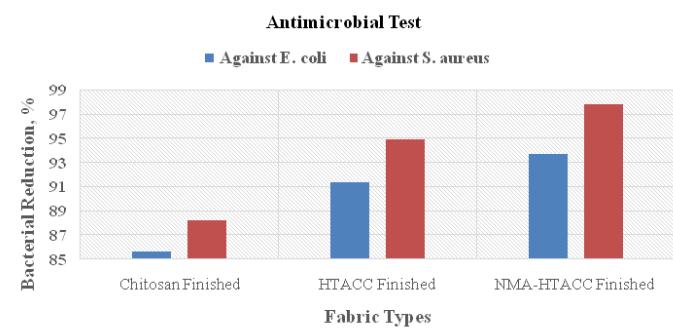
the attachment of chitosan, HTACC and NMA-HTACC on cotton fabric in comparison with washed fabric (Figure 3a). Among the finished fabrics, SEM of NMA-HTACC finished cotton fabric shows the better attachment of it on cotton fabrics, this is due to having good fibre reactivity.



**Figure 3** SEM of (a) Control, (b) Chitosan finished, (c) HTACC finished and (d) NMA-HTACC finished cotton fabrics.

## Anti-bacterial activity measurement

Figure 4 represents the anti-bacterial activity test (Quantitative analysis) of finished cotton fabrics. From this Figure, it can be seen that finished cotton fabrics have enough percentage of bacterial reduction against *E. coli* and *S. aureus*. But among these, NMA-HTACC treated cotton fabric shows higher anti-bacterial activity with respect to the percentage of bacterial reduction (97.8% against *S. aureus* and 93.7% against *E. coli*) than HTACC and Chitosan treated cotton fabrics. This is due to the better crosslinking activity of NMA-HTACC with cotton fabric than HTACC and Chitosan.



**Figure 4** Bacterial reduction (%) of the finished cotton fabric against *E. coli* and *S. aureus*.

## Conclusion

In this study Chitosan, HTACC and NMA-HTACC were exhausted on cotton fabric to develop antibacterial property including other textile properties. The presence of the finishing agents on cotton fabrics was confirmed by FTIR and SEM analysis of finished cotton fabrics. The experimental data support that HTACC and NMA-

HTACC can be used for cotton fabric finishing as an eco-friendly antibacterial agent to make it suitable in medical as well as personal care textiles for both gram-positive and gram-negative bacteria.

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## Conflicts of interest

The authors declare there are no conflicts of interest.

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