

PHYSICOCHEMICAL AND ANTIBACTERIAL PROPERTIES OF COTTON
FABRIC TREATED WITH CHITOSAN–TRIPOLYPHOSPHATE NANOPARTICLES
AND LAURIC ACID

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UNIVERSITI TEKNOLOGI MALAYSIA

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To my beloved mother and father.....

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ABSTRACT

Today, antibacterial treatment has become a prerequisite for textile goods used in hospitals, hotels, sports and personal care industries. Cotton is one of the most important natural fibers that is used extensively in textile industry. In this work, properties of cotton fabric treated with chitosan-tripolyphosphate nanoparticles (CNPs) and lauric acid (LA) was studied. CNPs were prepared based on the ionic gelation of chitosan with tripolyphosphate anions. The pretreatment process of cotton fabric was performed using sodium hydroxide. Afterwards, the CNPs solution with the addition of LA was applied to the treated cotton fabric to give wider antibacterial effects. The physicochemical properties of the CNPs and treated cotton fabric were determined by size distribution, Fourier transform infrared analysis and field emission scanning electron microscopy analysis. The average size of CNPs obtained was 179.1 ± 82.45 nm at concentration of 0.1% chitosan (w/v). The morphology surface of CNPs revealed the particles were spherical and almost uniform. CNPs solution with the addition of 15% (w/v) LA was formulated to treat the cotton fabric by soaking the cotton fabric in the solution while stirring for two hours at 60 °C. The antibacterial activity of treated cotton fabric with CNPs and LA against *Bacillus subtilis* (Gram-positive bacteria) and *Escherichia coli* (Gram-negative bacteria) was evaluated by using agar diffusion and liquid culture test methods. Cotton fabric treated with CNPs and LA showed a good ability to inhibit bacteria reproduction with diameter of zone of inhibition of 2.03 ± 0.09 cm for *Escherichia coli* and 2.27 ± 0.07 cm for *Bacillus subtilis*. Results showed that CNPs incorporated with LA gave better inhibition toward tested bacteria compared to CNPs solely, with differences as much as 30% for Gram-negative bacteria and 71% for Gram-positive bacteria.

ABSTRAK

Sekarang, rawatan anti-bakteria telah menjadi pra-syarat utama bagi barangan tekstil yang digunakan di hospital, hotel, sukan dan industri penjagaan diri. Kapas adalah merupakan salah satu daripada gentian semulajadi paling penting yang digunakan secara meluas dalam industri tekstil. Dalam hasil kerja ini, sifat-sifat kain kapas yang dirawat dengan nanopartikel kitosan-tripolifosfat (CNPs) dan asid laurik (LA) telah dikaji. CNPs disediakan berasaskan gelatin ionik kitosan dengan anion tripolifosfat (TPP). Proses prarawatan kain kapas telah dilakukan menggunakan natrium hidroksida. Selepas itu, larutan CNPs dengan penambahan LA telah digunakan untuk kain kapas terawat untuk memberi kesan anti-bakteria yang lebih meluas. Sifat-sifat fizikokimia daripada CNPs dan kain kapas terawat ditentukan dengan taburan saiz, analisis inframerah jelmaan Fourier dan analisis mikroskop elektron imbasan pancaran medan. Saiz purata CNPs diperolehi ialah 179.10 ± 82.45 nm pada kepekatan 0.1% chitosan (w/v). Permukaan morfologi CNPs menunjukkan zarah yang terhasil adalah berbentuk sfera dan hampir seragam. Larutan CNPs dengan penambahan 15% (w/v) LA telah diformulasikan untuk merawat kain kapas dengan merendam kain kapas di dalam larutan tersebut dan dikacau selama dua jam pada suhu 60 °C. Aktiviti anti-bakteria kain kapas yang dirawat dengan CNPs dan LA terhadap *Bacillus subtilis* (bakteria Gram-positif) dan *Escherichia coli* (bakteria Gram-negatif) telah diukur dengan menggunakan kaedah penyebaran agar dan ujian kultur cecair. Kain kapas dirawat dengan CNPs dan LA menunjukkan keupayaan yang baik untuk merencat pembiakan bakteria dengan zon perencatan sebanyak 2.03 ± 0.09 cm untuk *Escherichia coli* dan 2.2 ± 0.07 cm untuk *Bacillus subtilis*. Keputusan menunjukkan bahawa CNPs yang digabungkan dengan LA memberikan perencatan yang lebih baik terhadap bakteria diuji berbanding dengan CNPs tanpa LA dengan perbezaan sebanyak 30% untuk bakteria Gram-negatif dan 71% untuk bakteria Gram-positif.

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LIST OF SYMBOLS

ml	Mililiter
μl	Microliter
g	Gram
°C	Degree celcius
hr	Hour
kV	Kilo volt
wt	Weight
rpm	Revolution per minute
v/v	Volume per volume
w/v	Weight per volume

LIST OF ABBREVIATIONS

AATCC	American Association of Textile Chemists and Colorists
AM	Antimicrobial
AgNPs	Silver nanoparticles
CAGR	Compound Annual Growth Rate
CDC	Centers for Disease Control and Prevention
CFU	Colony Forming Unit
CNPs	Chitosan nanoparticles
CNTs	Carbon nanotubes
DMDMH	Dimethylol-5-5-dimethylhydantoin
DNA	Deoxyribonucleic acid
FESEM	Field Emission Scanning Electron Microscopy
FTIR	Fourier Transform Infrared Spectroscopy
HAI	Healthcare Associated Infections
HHS	Health and Human Services
KBr	Potassium Bromide
LA	Lauric Acid
MDMH	Monomethylol-5-5-dimethylhydantoin
MRSA	Methicillin-resistant <i>Staphylococcus aureus</i>
NMA-HTCC	O-acrylamidomethyl-N-[(2-hydroxy-3-trimethylammonium)propyl] chitosan derivative
NPs	Nanoparticles
TPP	Pentasodium triphosphate/ tripolyphosphate
TNTs	Titania nanotubes
UV-vis	Ultraviolet visible
WHO	World Health Organization

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Over the last twenty years, intensive research into new material and procedures, that would assure permanent bioactive effects together with complete safety for the people has been done due to the increase in the number of microbially caused diseases and hospital infections (Chung *et al.*, 1998; Liu *et al.*, 2000; Lee *at al.*, 1999). There are commonly found microorganism in textile materials likes mould, germs, Gram-positive and Gram-negative bacteria that have occupied every habitat on earth from geothermal vents to the coldest Arctic ice and play both beneficial and harmful roles in human lives.

According to the Centers for Disease Control and Prevention (CDC, USA, 2011), there are approximately 722,000 Healthcare Associated Infections (HAIs) and 75,000 associated deaths occur each year on account of infection-causing bacteria. About 85% of all invasive methicillin-resistant *Staphylococcus aureus* (MRSA) infections were associated with health care (Klevens *et al.*, 2007). In 2005, there were about 94,360 people infected with serious MRSA infection in the United States and 18,650 people died.

Hence, the control of infections has been identified as the most important target by the United States Department of Health and Human Services (HHS). In 2011, World Health Organization (WHO) has reported the HAIs from 1995 to 2010 worldwide including high and low/ middle-income countries. According to the report, the rates is higher in developed countries including Asia and Malaysia especially. Comparing to other developed countries, the prevalence of HAIs in Malaysia is around 14%. This percentage is quite high compared to neighbour country such as Indonesia at 7.1% and Thailand at 6.5%. HAIs can be controlled by inhibiting the various routes of transmission that causes an infection to spread from an infected person to healthy person. Infection can spread through various routes for example direct contact with infected individuals, infected water and food, and also contact with inanimate objects such as textiles used in daily life products (eg: bed-sheet, curtains, doctor's coat). Therefore, the antibacterial properties of a fibrous material are most important biomedical characteristics for external use (Simona *et al.*, 2000).

The control of the spread of infections through infected individuals, water and food can be achieved by developing hygienic practices. One of it by the use of antibacterial textiles that kill pathogens on contact or hinder their ability to reproduce prior to being transferred on to another material or person. Gao and Craston (2008) has stated that other than the requirements of the healthcare facilities, the increase in consumer's demand for comfort, hygiene and well-being has created a large and rapidly increasing market for antibacterial textiles. As an example, the market for antimicrobial (AM) coatings was about \$1.5 billion in 2012 by value and from 2013 to 2018, it is estimated to grow with Compound Annual Growth Rate (CAGR) of about 11.8% (MarketsandMarkets, 2013).

Antibacterial textiles are made by treating textile substrate with antibacterial agents which bound to textiles by different methods depending on the chemistry between the antibacterial agent and the textile (Gao and Cranston, 2008). Various types of chemicals have been employed to produce an antibacterial activity in textile. The antibacterial agents most widely used for textile applications are inorganic salts, organometalics, iodo-phor, phenol and thiophenols, onium salts, antibiotics,

heterocyclics with anionic groups, nitro compounds, ureas and related compounds, formaldehyde derivatives and amines (Lim and Hudson, 2004a; Lim and Hudson, 2004b). Most of these chemicals are toxic to human and cannot easily degrade in nature. These antibacterial agents have been studied independently and have been proven to possess effective antibacterial ability.

The above factors have led to high interest in researching and developing technology of natural antibacterial agent. One of the commonly used natural AM agent is chitosan, a β -(1,4)-linked polysaccharide of D-glucosamine, is a deacetylated form of chitin, the second most abundant natural polymer in the world (Ökem, 2003; Sashiwa *et al*, 2004). Furthermore, in textile technology application, it is important to adjoin some additional features in the natural biopolymer. Thus, various additives such as lauric acid are incorporated into the chitosan solution to enhanced the antibacterial activity of the treated fabric.

1.2 Problem Statements

Nowadays, the protection against infection of harmful and pathogenic microorganism is strengthened. This is due to the facts that a safe, hygiene and comfortable living environment turns to be important. Hence, the demand for the healthcare textiles and medical textiles as ever increasing. Comparing to man-made fibers, natural textiles, especially made from cellulose and protein fibers are often considered to be more vulnerable towards microbe attack due to characteristics of hydrophilic porous structure and moisture transport. Therefore, to prevent the growth of bacteria using antibacterial agent becoming a standard finishing for textile goods.

In textiles these can be achieved by treating the fabrics with silver salts, quaternary ammonium chloride, metals, aromatic, halogen compounds, etc (Kenawy *et al.*, 2007; Takai *et al.*, 2002). However, most of these antibacterial agents are toxic and biocidal. Due to its non-toxicity and antibacterial properties, chitosan was selected to be incorporated into fabric fiber as AM agent. According to Simona *et al.* (2000), chitosan antibacterial activity is assigned on its amino groups, in diluted acids from ammonium salts and the manipulation of chitosan's binding strategies onto cellulose surface. Based on the previous study on the AM textiles, most of the research focused on synthesizing and evaluating uniquely distinct antibacterial agents on different textile substrates with the aim of proving their effectiveness against various microbes (Lim and Hudson, 2004; Qi *et al.*, 2004; Ye *et al.*, 2005; Demir *et al.*, 2010; Hebeish *et al.*, 2013).

Chitosan nanoparticles (CNPs) is one of the alternatives that was investigated for its AM properties in textile applications. According to Hebeish *et al.*, (2013), the nano form of chitosan is highly active due to very high surface area to volume ratio and expected to have desirable bioactivity even at very low concentration. Lauric acid (LA) is a crystalline fatty acid which has been shown to have AM effects towards Gram-positive bacteria and yeast (Beuchat and Golden, 1989; Kabara, 1993). LA will be added to widen the spectrum activity of antibacterial mechanism towards Gram-positive and Gram-negative bacteria.

In general, this study focused on the enhancement of antibacterial textiles in terms of increase the fabric safety, increase natural antibacterial effectiveness towards microbes and reducing the usage of chemical antibacterial agents by replacing natural sources that are non-toxic and safe for the public usage.

1.3 Objectives of the Study

The main objective of this study is to develop antibacterial cotton fabric incorporated with chitosan nanoparticles (CNPs) and lauric acid to enhance physical and antibacterial properties of cotton fabric. The specific objectives of this study are:

1. To investigate the effect of chitosan concentration on the formation of CNPs.
2. To determine the effect of LA concentration on antibacterial properties.
3. To characterize physical and antibacterial properties of treated cotton fabric.

1.4 Scope of the Study

In order to achieve the objectives of this study, the work performed included all the following scope.

1. The nano-sized particles of chitosan were extracted via ionotropic gelation method based on the reaction between the cationic amino groups of chitosan and Tripolyphosphate (TPP).
2. Five different concentrations of chitosan from 0.1% to 0.5% (w/v) were used to obtain the smallest nano size of particles.
3. An antibacterial solution was formulated by adding CNPs into the solution with different LA ratios (0, 7, 9, 12, and 15 wt % of chitosan weight).
4. Formulation that gave the best antibacterial properties for CNPs-LA was selected to be incorporated into fabric as an antibacterial agent.
5. Physical properties of cotton fabric treated with CNPs-LA were analyzed by surface structure and particles morphology.

6. Antibacterial study of the cotton fabric against Gram-positive (*B. subtilis*) and Gram-negative (*E. coli*) bacteria were evaluated by agar diffusion test and liquid culture test

1.5 Significance of the Study

Changing of human lifestyle nowadays, led to safe, healthy and comfortable living environment keeps demanding. It is very important to keep strengthened the protection from the infection of pathogenic microorganisms. Therefore, demand for the healthcare and medical textiles keep increasing. Until now, a number of chemical have been employed to impart the antibacterial activity of the textiles that most of these chemical are toxic to human and cannot easily degrade in the nature. The textile industry continuesly looking for the eco-friendly process that can be carried out without toxic textile chemicals. Hence, the use of natural source of AM agent like chitosan has been introduced to replace those chemical AM agent. The incorporation of CNPs will increase the AM properties of the textiles. Addition of LA will widen the AM activity of the textile.

1.6 Outline of the Thesis

This thesis consists of 5 Chapters. Chapter 1 introduces the introduction of the research, significance of the study, research problem, the objective and the scope of the study. Chapter 2 presents the literature review of the CNPs, antibacterial fabrics, antibacterial agents used in fabric treatments, nanoparticles (NPs) in textiles and ionotropic gelation method used to produced CNPs. Chapter 3 provides a detailed methodology of this research to achieve the targeted objectives. Results and

discussion shows in Chapter 4 on physical properties and antibacterial activity of CNPs and cotton fabric treated with CNPs solution. Effects of adding LA into the solution also discussed in this chapter. Finally chapter 5 summarises the findings of this study and suggestions for further work.

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