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**PREPARATION AND ANTIBACTERIAL ACTIVITY OF  
QUATERNARY AMMONIUM-CONTAINING CHITOSAN  
SURFACE**

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A Thesis Submitted in Partial Fulfillment of the Requirements  
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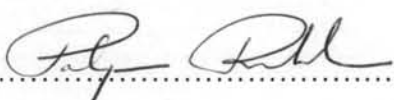
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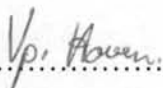


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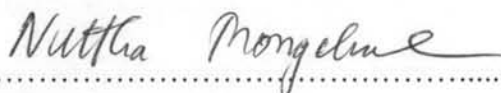
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การทำปฏิกิริยาควอเทอร์ไนเซชันของหมู่อะมิโนของไคโตซานเป็นแนวทางที่ได้รับการยอมรับว่ามีประสิทธิภาพในการเพิ่มฤทธิ์ต้านแบคทีเรียของไคโตซานในช่วงความเป็นกรด-เบสที่กว้างขึ้น ปฏิกิริยาดังกล่าวสามารถเพิ่มประจุบวกและความไม่ชอบน้ำซึ่งเป็น 2 ตัวแปรที่เชื่อว่ามีผลต่อการออกฤทธิ์ต้านแบคทีเรีย งานวิจัยนี้จึงมีเป้าหมายที่จะเพิ่มฤทธิ์ต้านแบคทีเรียของพื้นผิวไคโตซาน โดยการติดหมู่ควอเทอร์นารีแอมโมเนียมผ่านปฏิกิริยาแบบวิวิธพันธ์ ที่แบ่งเป็น 2 ชั้น คือ ปฏิกิริยารีดักทีฟอัลคิลเลชัน โดยแปรเปลี่ยนชนิดของอัลดีไฮด์ตามด้วย ปฏิกิริยาเมทิลเลชันด้วยเมทิลไอโอไดด์ ผลจากการวิเคราะห์ด้วยเอทีอาร์-เอฟทีไออาร์, การวัดมุมสัมผัสของน้ำ และ การหาค่าประจุบนพื้นผิว แสดงให้เห็นถึงความสำเร็จในการทำปฏิกิริยาควอเทอไนเซชันของผิวไคโตซาน จากผลการศึกษาและเปรียบเทียบฤทธิ์ต้าน *Staphylococcus aureus* (แบคทีเรียแกรมบวก) และ *Escherichia coli* (แบคทีเรียแกรมลบ) ซึ่งพิจารณาจากการวัดค่าออปติคัลเดนซิตีที่ความยาวคลื่น 600 นาโนเมตร ( $OD_{600}$ ), ภาพถ่ายสแกนนิ่งอิเล็กตรอนไมโครสโกปี และการนับจำนวนแบคทีเรียที่ยังคงมีชีวิต พบว่าผิวฟิล์มไคโตซานที่ผ่านการดัดแปรแล้วมีฤทธิ์ต้านแบคทีเรียที่เหนือกว่าผิวฟิล์มไคโตซานธรรมชาติ แสดงให้เห็นว่าประจุบวกและหมู่อัลคิลที่เพิ่มขึ้นบนผิวฟิล์มไคโตซานหลังจากการทำปฏิกิริยาควอเทอร์ไนเซชันของพื้นผิวทำให้เกิดหมู่ควอเทอร์นารีแอมโมเนียมบนผิวไคโตซานที่สามารถเกิดอันตรกิริยากับประจุลบของผนังเซลล์แบคทีเรีย

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NAPANPORN VALLAPA: PREPERATION AND ANTIBACTERIAL  
 ACTIVITY OF QUATERNARY AMMONIUM-CONTANINIG CHITOSAN  
 SURFACE. THESIS ADVISOR: ASST. PROF. VORAVEE P. HOVEN,  
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Quaternization of amino groups of chitosan has been recognized as a potential way to enhance the antibacterial activity of chitosan in a broader pH range. The reaction can simultaneously introduce both positive charge and hydrophobicity, the two parameters that are believed to affect the antibacterial activity. This research aims to increase the antibacterial activity of chitosan surface by introducing quaternary ammonium groups via a heterogeneous two-step process: reductive alkylation using selected aldehydes followed by methylation with methyl iodide. Results from ATR-FTIR analysis, water contact angle and zeta potential measurements confirmed the success of surface quaternization. As determined from an optical density ( $OD_{600}$ ), scanning electron microscopy (SEM) image and viable cell counting method, the antibacterial activity of the surface-modified chitosan film against *Staphylococcus aureus* (gram positive bacteria) and *Escherichia coli* (gram negative bacteria) were superior to that of the virgin chitosan film. The additional positive charge and hydrophobicity introduced to the chitosan film after surface quaternization apparently make the quaternary ammonium-containing chitosan film a more favorable substrate for interacting with the negatively-charged membrane of the bacteria.

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## LIST OF ABBREVIATION

ATR-FTIR	: Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy
BuI	: Butyl iodide
DBuM	: <i>N</i> -methyl- <i>N,N</i> -dibutyl chitosan
DEM	: <i>N</i> -methyl- <i>N,N</i> -diethyl chitosan
DOM	: <i>N</i> -methyl- <i>N,N</i> -dioctyl chitosan
DMBu	: <i>N</i> -butyl- <i>N,N</i> -dimethyl chitosan
DMBz	: <i>N</i> -benzyl- <i>N,N</i> -dimethyl chitosan
DME	: <i>N</i> -ethyl- <i>N,N</i> -dimethyl chitosan
DMP	: <i>N</i> -propyl- <i>N,N</i> -dimethyl chitosan
<i>E.coli</i>	: <i>Escherichia coli</i>
EtI	: Ethyl iodide
MeI	: Methyl iodide
OcI	: Octyl iodide
OD	: Optical Density
PBS	: Phosphate buffer saline
<i>S.aureus</i>	: <i>Staphylococcus aureus</i>
SEM	: Scanning Electron Microscopy