

**STRUCTURALLY CONTROLLED CHITIN DERIVATIVES BY  
CHEMICAL MODIFICATION APPROACH : AN APPLICATION FOR  
INSECTICIDE CONTROLLED RELEASE**

Ms. Amornrat Lertworasirikul

A Thesis Submitted in Partial Fulfillment of the Requirements  
for the Degree of Master of Science  
The Petroleum and Petrochemical College, Chulalongkorn University  
in Academic Partnership with  
The University of Michigan, The University of Oklahoma  
and Case Western Reserve University

1999

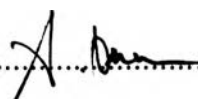
ISBN 974-331-915-8

119337401

**Thesis Title** : Structurally Controlled Chitin Derivatives by Chemical  
Modification Approach : An Application for Insecticide  
Controlled Release  
**By** : Ms. Amornrat Lertworasirikul  
**Program** : Polymer Science  
**Thesis Advisors** : Assoc. Prof. David C. Martin  
Dr. Suwabun Chirachanchai

---

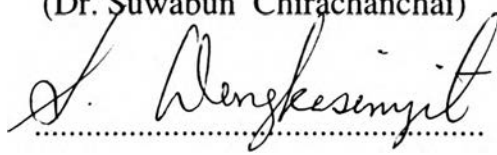
Accepted by the Petroleum and Petrochemical College, Chulalongkorn  
University, in partial fulfillment of the requirements for the Degree of Master  
of Science.

  
..... College Director  
(Prof. Somchai Osuwan)

**Thesis Committee:**

  
.....  
(Assoc. Prof. David C. Martin)

  
.....  
(Dr. Suwabun Chirachanchai)

  
.....  
(Asst. Prof. Sujitra Wongkasemjit)

## บทคัดย่อ

อมรรัตน์ เกศวรสิริกุล : การควบคุมโครงสร้างของอนุพันธ์ไคตินโดยวิธีการทางเคมี : การใช้งานด้านควบคุมการปลดปล่อยยาปราบศัตรูพืช (Structurally Controlled Chitin Derivatives by Chemical Modification Approach : An Application for Insecticide Controlled Release) อ. ที่ปรึกษา : ดร. สุวบุญ จิรชาญชัย และ รศ. ดร. เดวิด ซี มาร์ติน (Assoc. Prof. David C. Martin) 58 หน้า ISBN 974-331-915-8

อนุพันธ์ของไคโตแซนประเภท 1-แนพทิลเมทิลคาร์บาเมตได้ถูกสังเคราะห์ขึ้นโดยการออกแบบโมเลกุล 2 รูปแบบ คือ แบบการเชื่อมเข้าโดยตรงโดยปราศจากตัวเชื่อมต่อ (Spacer) ได้แก่ ไคโตแซน-คาร์บาริล (Chitosan-Carbaryl, CHI-CBR) ซึ่งเตรียมผ่านอนุพันธ์ไอโอดิไคโตแซน (Iodochitosan) และ แบบมีตัวเชื่อมต่อ ได้แก่ ไคโตแซนอะซีเตตคาร์บอนิลอิมิดาโซลิด-คาร์บาริล (Chitosan Acetate-Carbonyl Imidazolid-Carbaryl, CA-CDI-CBR) โดยเตรียมได้จากอนุพันธ์ไคโตแซน-คาร์บอนิลอิมิดาโซลิด (Chitosan-Carbonyl-imidazolid) สารประกอบที่ได้ปรากฏฟีกของคาร์บาเมตเอสเตอร์ที่ 1707-1708 เลขคลื่น การศึกษาด้วยเครื่องมือวิเคราะห์ธาตุ (Elemental Analysis) พบว่าประสิทธิภาพของปฏิกิริยาการเข้าแทนที่อยู่ที่ระดับ 50-60 เปอร์เซ็นต์ ผลการวิเคราะห์โดยเครื่องเทอร์โมกราวิเมตริก (Thermogravimetric Analysis) พบว่า CHI-CBR มีจุดหลอมเหลวที่ 221 องศาเซลเซียส ในขณะที่ CA-CDI-CBR ปรากฏจุดหลอมเหลวในช่วงอุณหภูมิกว้างจาก 158 ถึง 376 องศาเซลเซียส การศึกษาโดยเอกซเรย์ดิฟแฟรกชันสเปกโตรสโกปี (X-ray Diffraction Spectroscopy) พบว่าความยาวคลอริเรชัน (Correlation Length) ของสารประกอบลดลงเมื่อเทียบกับไคโตแซน การศึกษา CHI-CBR ในสารละลายต่างที่ความเป็นเบสที่พีเอช (pH) 12 ยืนยันเสถียรภาพของคาร์บาริลในสายโซ่โพลิเมอร์ ในขณะที่เดียวกันการศึกษาด้วยเทคนิคของยูวี-วิสิเบิลสเปกโตรโฟโตเมตรี (UV-VIS Spectrophotometry) พบว่าการปลดปล่อย CBR จาก CA-CDI-CBR เกิดขึ้นได้จากกระบวนการไฮโดรไลซิส (Hydrolysis) ของพันธะเอสเทอร์ในตัวเชื่อม

## ABSTRACT

## 972001 : POLYMER SCIENCE PROGRAM

KEY WORDS: Chitin Derivatives/ Controlled Release System/

Chemical Conjugation

Amornrat Lertworasirikul: Structurally Controlled Chitin Derivatives by Chemical Modification Approach: An Application for Insecticide Controlled Release. Thesis Advisors: Dr. Suwabun Chirachanchai and Assoc. Prof. David C. Martin, 58 pp. ISBN 974-331-915-8

Chitosan derivatives with 1-naphthyl methylcarbamate were prepared by two molecular designs, i.e., without spacer via iodochitosan to obtain chitosan-carbaryl (CHI-CBR), and with spacer via chitosan-carbonyl imidazolide to obtain chitosan acetate carbonyl imidazolide-carbaryl (CA-CDI-CBR). The products show carbamate ester peak at  $1707-1708\text{ cm}^{-1}$ . The substitution is accomplished for 50-60 % as clarified by elemental analysis. TGA reveals that CHI-CBR shows a degradation temperature at  $221\text{ }^{\circ}\text{C}$  while CA-CDI-CBR exhibits wide range of degradation from  $158\text{ }^{\circ}\text{C}$  to  $345\text{ }^{\circ}\text{C}$ . XRD patterns show a decrease of the correlation length of the products as compared to that of chitosan. The treatment of CHI-CBR in alkali solution of pH 12 confirms the stability of the carbaryl in the polymer chain. The release of CBR from CA-CDI-CBR is achieved by the hydrolysis of the ester spacer as studied by UV.

## ACKNOWLEDGEMENTS

The author would like to thank all professors who have tendered invaluable knowledge to her at the Petroleum and Petrochemical College, Chulalongkorn University. She would like to give a special thanks to her U.S. advisor, Assoc. Prof. David C. Martin who gave recommendations on the research. She is deeply indebted to her Thai advisor, Dr. Suwabun Chirachanchai, who not only originated this thesis work, but also gave her intensive suggestion, constructive advice and vital help throughout this research work.

She wishes to thank Prof. Suwalee Chandkrachang and Prof. Mitsuru Akashi, for her and his kindness on supporting chitin-chitosan in this research work. She also would like to thank AG-GRO (THAILAND) Co., Ltd. for supporting insecticide of 1-naphthyl methylcarbamate (carbaryl). She specially thanks the National Metal and Material Technology Center (MTEC) for funding this research including solid state NMR measurements.

She would like to give a special thanks to Assit. Prof. Vanida Bhavakul of the Department of Chemistry, Faculty of Science, King Mongkut's Institute of Technology Thonburi, who gave her precious advice on chemical reaction.

She would like to thank her seniors, Mr. Surakit Chunharotrit and Ms. Wanpen Tachaboonyakiat, for his and her kindness on help and suggestion since the beginning of this research work. In addition, she would like to thank Mr. Apirat Laobuthee, the Ph. D. student, for his help and suggestion. She also would like to thank the college members, staff, and all her best friends at the Petroleum and Petrochemical College.

Finally, the sincerest appreciation is dedicated to her family for the love, understanding, encouragement and financial support.

## TABLE OF CONTENTS

	PAGE
Title Page	i
Abstract (in English)	iii
Abstract (in Thai)	iv
Acknowledgements	v
Table of Contents	vi
List of Figures	x
List of Schemes	xiii
<b>CHAPTER</b>	
<b>I INTRODUCTION</b>	<b>1</b>
<b>II LITERATURE SURVEY</b>	
2.1 Chemical Structure of Chitin-Chitosan	4
2.2 Chitin-Chitosan Derivatives	5
2.2.1 Chemically Modification of Chitin-Chitosan at N-position	5
2.2.2 Chemically Modification of Chitin-Chitosan at O-position	7
2.3 Chitin-Chitosan for Drug Delivery System	8
2.3.1 Physical Conjugation System	8
2.3.2 Chemical Conjugation System	9
2.4 Chitosan for Agricultural Purpose	10
2.5 The Potential of the Present Work	10
<b>III EXPERIMENTAL SECTION</b>	
3.1 Materials	11
3.2 Instruments and Equipment	11

CHAPTER	PAGE
3.2.1 Fourier Transform Infrared Spectrophotometer (FT-IR)	11
3.2.2 Elemental Analysis (EA)	12
3.2.3 Nuclear Magnetic Resonance Spectrometer (NMR)	12
3.2.4 Thermal Gravimetric Analysis (TGA)	12
3.2.5 Ultraviolet-Visible Spectrophotometer (UV-VIS)	12
3.2.6 X-ray Diffraction (XRD)	12
3.3 Experimental Procedure	
3.3.1 Preparation of Chitosan Precursors	13
3.3.1.1 Preparation of Tosylchitosan	13
3.3.1.2 Preparation of Iodochitosan	13
3.3.1.3 Preparation of Chitosan acetate	14
3.3.1.4 Preparation of Chitosan acetate- Carbonyl imidazolide(CA-CDI)	14
3.3.2 Preparation of Chitosan Conjugated Drug	
3.3.2.1 Synthesis of Chitosan Conjugated Drug Type 1: Chitosan-Carbaryl (CHI-CBR)	15
3.3.2.2 Synthesis of Chitosan Conjugated Drug Type 2: Chitosan acetate- Carbonyl imidazolide-Carbaryl (CA-CDI-CBR)	16
3.3.3 Stability Study of Chitosan-Carbaryl (CHI-CBR) (Type 1)	17
3.3.4 Release Study of Chitosan acetate-Carbonyl- imidazolide-Carbaryl (CA-CDI-CBR) (Type 2)	17

CHAPTER	PAGE
<b>IV RESULTS AND DISCUSSION</b>	
4.1 Chemical Structure Analysis of Chitosan	18
4.2 Preparation of Chitosan Precursors	21
4.2.1 Iodochitosan Pathway	22
4.2.1.1 Preparation of Tosylchitosan	22
4.2.1.2 Preparation of Iodochitosan	25
4.2.2 Chitosan acetate-Carbonyl imidazolide (CA-CDI) Pathway	30
4.2.2.1 Preparation of Chitosan acetate (CA)	30
4.2.2.2 Synthesis of Chitosan acetate Carbonyl imidazolide (CA-CDI)	32
4.3 Preparation of Chitosan Conjugated Drug	37
4.3.1 Synthesis of Chitosan Conjugated Drug Type 1: Chitosan-Carbaryl (CHI-CBR)	37
4.3.2 Synthesis of Chitosan Conjugated Drug Type 2: Chitosan acetate-Carbonyl imidazolide-Carbaryl (CA-CDI-CBR)	40
4.4 Structural Study by XRD	44
4.4.1 Structural Analysis of Type 1: Chitosan-Carbaryl (CHI-CBR)	44
4.4.2 Structural Analysis of Type 2: Chitosan acetate- Carbonyl imidazolide (CA-CDI-CBR)	45
4.5 Stability Study of Chitosan Conjugated Drug Type 1: Chitosan-Carbaryl (CHI-CBR)	48
4.6 Release Study of Chitosan Conjugated Drug Type 2: Chitosan acetate-Carbonyl imidazolide-Carbaryl (CA-CDI-CBR)	50



<b>CHAPTER</b>		<b>PAGE</b>
<b>V</b>	<b>CONCLUSIONS</b>	<b>53</b>
	<b>REFERENCES</b>	<b>54</b>
	<b>CURRICULUM VITAE</b>	<b>58</b>

## LIST OF FIGURES

FIGURE	PAGE
1.1 Chemical structure of chitin-chitosan copolymer	1
1.2 Chemical structure of 1-naphthyl methylcarbamate (carbaryl)	2
1.3 Molecular design for insecticide conjugated chitosan, (a) Type 1, polymeric drug molecule without spacer, and (b) Type 2, polymeric drug molecule with spacer	3
2.1 Chemical structures of (a) cellulose, (b) chitin, and (c) chitosan	5
2.2 Alkylation of chitosan	7
2.3 N-Phthaloylation of chitosan	7
4.1 FT-IR spectrum of chitosan (DD = 85.9%)	18
4.2 FT-IR spectrum of chitosan (DD = 75.8%)	19
4.3 TGA diagram of chitosan with a degree of deacetylation 85.9%	20
4.4 TGA diagram of chitosan with a degree of deacetylation 75.8%	21
4.5 FT-IR spectra of (a) chitosan (DD = 85.9%), and (b) tosylchitosan	23
4.6 Ideal structure of tosylchitosan when tosylation is completed to make the S/N ratio be 4.25	24
4.7 TGA diagram of tosylchitosan	24
4.8 Solid state <sup>13</sup> C-NMR spectrum of tosylchitosan	25
4.9 FT-IR spectra of (a) tosylchitosan, and (b) iodochitosan	26
4.10 Qualitative curve fitting of iodochitosan	28
4.11 TGA diagram of iodochitosan	29

FIGURE	PAGE
4.12 FT-IR spectra of (a) chitosan (DD = 75.8%), and (b) CA	30
4.13 TGA diagram of chitosan acetate	31
4.14 Structures of chitosan acetate from chitosan %DD = 85.9 (a) ideal structure for completion of acetate group, and (b) 80% acetate salt formation structure	32
4.15 FT-IR spectra of (a) chitosan acetate, and (b) CA-CDI	33
4.16 Qualitative curve fitting of CA-CDI	34
4.17 Ideal structure of chitosan acetate-carbonyl imidazolide of 80% chitosan acetate formation	35
4.18 TGA diagram of CA-CDI	35
4.19 Solid state <sup>13</sup> C-NMR spectrum of CA-CDI	36
4.20 Chemical structure of carbaryl	37
4.21 FT-IR spectra of (a) iodochitosan, (b) CHI-CBR, and (c) carbaryl	39
4.22 Qualitative curve fitting of CHI-CBR	39
4.23 TGA diagram of CHI-CBR	40
4.24 FT-IR spectra of (a) CA-CDI, and (b) CA-CDI-CBR	42
4.25 Qualitative curve fitting of CA-CDI-CBR	43
4.26 TGA diagram of CA-CDI-CBR	43
4.27 XRD patterns of chitosans (a) DD = 85.9 %, and (b) DD = 75.8 %	46
4.28 XRD patterns of (a) chitosan (DD = 85.9 %), (b) tosylchitosan, (c) iodochitosan, (d) CHI-CBR, and (e) carbaryl	46
4.29 XRD patterns of (a) chitosan (DD = 75.8 %), (b) CA, (c) CA-CDI, (d) CA-CDI-CBR, and (e) carbaryl	47
4.30 UV spectra of (a) carbaryl in methanol, (b) carbaryl in 0.01 N NaOH at 60°C for 2 h, (c) CHI-CBR in 0.01N NaOH at 60°C for 2 h, and (d) NaI in 0.01 N NaOH	49

FIGURE	PAGE
4.31 Polymer chain as a protector for polymer drug as a stable drug	50
4.32 Release mechanism of CA-CDI-CBR via the basic reaction for basic condition at (a) CDI ester position, and (b) carbaryl ester position	51
4.33 UV spectrum of CA-CDI-CBR in 0.01 N NaOH at 60°C for 0.5 h	52

**LIST OF SCHEMES**

SCHEME	PAGE
3.1 Preparation of tosylchitosan	13
3.2 Preparation of iodochitosan	13
3.3 Preparation of CA-CDI	14
3.4 Preparation of CHI-CBR (Type1)	15
3.5 Preparation of CA-CDI-CBR (Type2)	16
4.1 Iodination for tosylated chitosan by (a) $S_N1$ mechanism, and (b) $S_N2$ mechanism	27
4.2 Mechanism of acetyl migration on chitin unit	29
4.3 Alkylation of N-substituted amides by using NaH	38
4.4 Mechanism of carbaryl coupling onto chitosan acetate-carbonyl imidazolide	41