

**NANOMATERIALIZATION OF CHITOSAN: APPROACHES VIA  
SUPRAMOLECULAR STRUCTURE AND CONTROLLED MORPHOLOGY**

Sasiprapha Phongying

A Dissertation Submitted in Partial Fulfilment of the Requirements  
for the Degree of Doctor of Philosophy  
The Petroleum and Petrochemical College, Chulalongkorn University  
in Academic Partnership with  
The University of Michigan, The University of Oklahoma,  
and Case Western Reserve University

2006

ISBN 974-9990-16-1

492219

**Thesis Title:** Nanomaterialization of Chitosan: Approaches via  
Supramolecular Structure and Controlled Morphology  
**By:** Sasiprapha Phongying  
**Program:** Polymer Science  
**Thesis Advisors:** Assoc. Prof. Suwabun Chirachanchai  
Dr. Sei-ichi Aiba

---

Accepted by the Petroleum and Petrochemical College, Chulalongkorn  
University, in partial fulfilment of the requirements for the Degree of Doctor of  
Philosophy.

*Nantaya Yanumet*  
..... College Director  
(Assoc. Prof. Nantaya Yanumet)

**Thesis Committee:**

*Nantaya Yanumet*  
.....  
(Assoc. Prof. Nantaya Yanumet)

*S. Chirachanchai*  
.....  
(Assoc. Prof. Suwabun Chirachanchai)

*胡有斌*  
.....  
(Dr. Sei-ichi Aiba)

*Hathaikarn Manuspiya*  
.....  
(Dr. Hathaikarn Manuspiya)

*Paitip T.*  
.....  
(Assoc. Prof. Paitip Thiravetyan)

**ABSTRACT**

4692004063: Polymer Science Program

Sasiprapha Phongying: Nanomaterialization of Chitosan: Approaches via Supramolecular Structure and Controlled Morphology.

Thesis Advisors: Assoc. Prof. Suwabun Chirachanchai and Dr. Seiichi Aiba 95 pp. ISBN 974-9990-16-1

Keywords: Chitosan/ Chitin/ Crown ethers/ Ion adsorbent/ Ion adsorption/ Ion extraction/ Ion selectivity/ Metal/ Pseudocyclic/ Chitin whisker/ Nanomaterial/ Scaffold/ Whisker/ Nanoscaffold/ Sugar/ Lactose/ Maltose/ Poly(ethylene glycol)/ PEG/ Carboxylation

Two approaches to develop nanomaterial of chitosan, i.e. (i) the supramolecular structured chitosan, and (ii) the changing in morphology of chitosan, are proposed. In the case (i), the functionalization carried out on the low molecular weight chitosan is focused. A series of aza-alkyl chains and oxy-alkyl chains conjugated onto chitosan is succeeded by using the coupling reaction. The studies on metal ion interaction clarify that the derivatives exhibit their ion adsorption via the supramolecular structure to result a comparable adsorption capacity to the chitosan with crown ether conjugated. For (ii), the nano-structured chitosan is obtained from the changing in morphology from chitin whisker to chitosan nanoscaffold via deacetylation. The surface functionalizations of chitin whisker with poly(ethylene glycol), and chitosan nanoscaffold with lactose and maltose are originally proposed.

## บทคัดย่อ

ศศิประภา ผ่องยิ่ง: การทำวัสดุไคโตซานให้มีขนาดระดับนาโนเมตร: โดยอาศัยหลักการ  
โครงสร้างแบบซูปราโมเลกุลและการควบคุมโครงสร้างอสัณฐาน (Nanomaterialization of  
Chitosan: Approaches via Supramolecular Structure and Controlled Morphology) อ.  
ที่ปรึกษา: รองศาสตราจารย์ ดร. สุวบุญ จิราญชัย และ ดร. เซอิชิ ไอบะ (Dr. Sei-ichi Aiba),  
95 หน้า ISBN 974-9990-16-1

วิทยานิพนธ์ฉบับนี้เสนอ 2 วิธีการในการพัฒนาวัสดุไคโตซานให้มีขนาดระดับนาโน  
เมตร ซึ่งประกอบด้วย (1) ไคโตซานที่มีโครงสร้างเป็นซูปราโมเลกุล และ (2) การเปลี่ยน  
โครงสร้างอสัณฐานของไคโตซาน ในกรณี (1) ไคโตซานที่มีมวลโมเลกุลต่ำได้ถูกปรับ  
โครงสร้างให้เป็นซูปราโมเลกุลโดยการผนวกสายโซ่อะซาลคิลและออกซีอัลคิลทลงบนสายโซ่  
ไคโตซาน การศึกษาอันตรกิริยากับไอออนของโลหะ พบว่าอนุพันธ์ที่สังเคราะห์ได้สามารถดูดจับ  
ไอออนโดยผ่านโครงสร้างที่เป็นซูปราโมเลกุล นำไปสู่การเปรียบเทียบความสามารถในการดูด  
จับกับไคโตซานที่ผนวกกับคราวน์อีเทอร์ ในกรณี (2) ไคโตซานที่มีโครงสร้างในระดับนาโน  
เมตรสามารถทำได้จากการเปลี่ยนโครงสร้างอสัณฐานจากไคตินวิสเกอร์ไปเป็นไคโตซานนาโนส  
แกฟโฟลด์โดยกระบวนการเปลี่ยนหมู่อะเซทิลของไคตินให้เป็นหมู่อะมิโนของไคโตซาน. ใน  
งานวิจัยนี้เสนอการปรับโครงสร้างบนผิวหน้าของไคตินด้วยพอลิเอทรีนไกลคอล และการปรับ  
โครงสร้างบนผิวหน้าของไคโตซานนาโนสแกฟโฟลด์ด้วยแลคโตสและมอลโทส

## ACKNOWLEDGEMENTS

The present dissertation would not have been accomplished without her Thai supervisor, Associate Professor Suwabun Chirachanchai, who not only originated this work, but also provided her continuous guidance, intensive recommendation, constructive criticism, invaluable suggestions and discussion, inspiration, and the opportunity to do the research in Japan from time to time.

She would like to express her appreciation to her Japanese co-advisor, Dr. Sei-ich Aiba (National Institute of Advanced Science and Technology (AIST), Osaka, Japan), for the recommendation, meaningful advices, the supports, and the well taken-care throughout her stay in Japan.

A deeply gratitude is expressed to Professor Mitsuru Akashi for Short-term Student Exchange Promotion Program Scholarship in Japan. Her appreciation also extends to Associate Professor Toshiyuki Kida for his suggestions and concerns. She is grateful to Dr. Michiya Matsusaki, Dr. Yoshio Nakahara, Mr. Takeyoshi Fukuda, and Mr. Daisuke Ogomi for his taking care and friendship during her research period in Japan of the first-year Ph.D. She wishes to thank all members in Akashi's laboratory and entire staffs at Bio-based Polymers Group, AIST, for their helps, friendship, good memories and warm hospitality throughout her stay in Japan. She would like to thank all Thai friends in Japan for their suggestion, encouragement, and friendship, and lots of help.

She would like to thank the dissertation committee for their suggestions and comments. She wishes to thank all Professors who have tendered valuable knowledge to her at the Petroleum and Petrochemical College, Chulalongkorn University.

Special thanks are to Dr. Amornrat Lertworasirikul and Dr. Nungruethai Yoswathananont for many helps and suggestion during her stay in Japan. She wishes to thank Mr. Jirawut Junkasem and Mr. Pinit Rattanadilok na Phuket for many helps, encouragement, and friendship all the time. Thanks to all members in SWB group for giving her helps and good time and memories during her study. In addition, she also wishes to express her appreciation to the entire college staff at the Petroleum and Petrochemical College who helped her throughout this research.

She appreciates the financial support from The Thailand Research Fund through the Royal Golden Jubilee Ph.D. Program (Ph.D. Grant No. PHD/0263/2545). She also acknowledges The National Research Council of Thailand for partial support (NRC 12/2548). This thesis work is partially funded by Postgraduate Education and Research Programs in Petroleum and Petrochemical Technology (PPT consortium).

She is indebted to Seafresh Chitosan (Lab) Company Limited, Thailand for providing her chitin-chitosan starting materials. Her acknowledgement also extends to Mr. Norioki Kawasaki (National Institute of Advanced Science and Technology (AIST), Osaka, Japan) for the help in FT-IR, GPC and  $^1\text{H}$  NMR measurement during her stay in Japan.

Last but not least, she would like to express her gratitude to her family for their love, understanding, encouragement, limitless sacrifice, and for being a constant source of their inspiration throughout her study.

## TABLE OF CONTENTS

	<b>PAGE</b>
Title Page	i
Abstract (in English)	iii
Abstract (in Thai)	iv
Acknowledgements	v
Table of Content	vii
List of Schemes	x
List of Tables	xi
List of Figures	xii
 <b>CHAPTER</b>	
<b>I INTRODUCTION</b>	<b>1</b>
 <b>II LITERATURE REVIEW</b>	
2.1 Chitin-Chitosan: The Structure and Specific Properties	4
2.2 Materialization of Chitin-chitosan	6
2.3 Nanomaterialization	10
2.4 Chitin-chitosan Nanomaterialization	14
2.5 Points of the Present Work	17
 <b>III CHITOSAN GRAFTED WITH OXYETHYLENE AND AZAETHYLENE CHAINS AND ITS METAL ION ADSORPTION CAPACITY AND SELECTIVITY</b>	
3.1 Abstract	18
3.2 Introduction	18
3.3 Experimental	19
3.4 Results and Discussion	25

<b>CHAPTER</b>	<b>PAGE</b>
3.5 Conclusions	33
3.6 Acknowledgements	33
3.7 References	33
<b>IV DIRECT CHITOSAN NANOSCAFFOLD FORMATION VIA CHITIN WHISKERS</b>	
4.1 Abstract	35
4.2 Introduction	35
4.3 Experimental	36
4.4 Results and Discussion	39
4.5 Conclusions	51
4.6 Acknowledgements	51
4.7 References	52
<b>V A NOVEL SOFT AND COTTON-LIKE CHITOSAN-SUGAR NANOSCAFFOLD</b>	
5.1 Abstract	54
5.2 Introduction	54
5.3 Experimental	56
5.4 Results and Discussion	59
5.5 Conclusions	67
5.6 Acknowledgements	67
5.7 References and notes	68
<b>VI FUNCTIONALIZATION OF CHITIN WHISKER WITH POLY(ETHYLENE GLYCOL)</b>	
6.1 Abstract	69
6.2 Introduction	69
6.3 Experimental	71
6.4 Results and Discussion	73



<b>CHAPTER</b>	<b>PAGE</b>
6.5 Conclusions	79
6.6 Acknowledgements	79
6.7 References	79
<b>VII CONCLUSIONS AND RECOMMENDATIONS</b>	<b>82</b>
<b>REFERENCES</b>	<b>84</b>
<b>CURRICULUM VITAE</b>	<b>92</b>

**LIST OF SCHEMES**

<b>SCHEME</b>		<b>PAGE</b>
	<b>CHAPTER I</b>	
1.1		1
	<b>CHAPTER II</b>	
2.1		4
2.2		5
	<b>CHAPTER III</b>	
3.1		18
3.2		24
3.3		30
	<b>CHAPTER IV</b>	
4.1		36
4.2		38
	<b>CHAPTER V</b>	
5.1		58
	<b>CHAPTER VI</b>	
6.1		69
6.2		73

## LIST OF TABLES

TABLE	PAGE
<b>CHAPTER III</b>	
3.1 Degradation temperature ( $T_d$ ) of low molecular weight chitosan (1), chitosan with oxyethylene chain (5a), and chitosan with azaethylene chain (6 - 11)	31
3.2 Adsorption selectivity of adsorbents in aqueous at pH = 7.0 for $Cr^{3+}$ , $Cu^{2+}$ , $Ni^{2+}$ , and $Ag^+$	32
<b>CHAPTER IV</b>	
4.1 Degradation temperature ( $T_d$ ) and ash content of chitin flakes, 2, 3, 3 (without lyophilization), and chitosan flakes 95%DD	48
4.2 Surface area, pore volume, and pore size of chitin flakes, 2, 3, and chitosan flakes 95%DD	49
4.3 Particle size of 3 in water, methanol, DMSO, and <i>iso</i> -propanol	51
<b>CHAPTER V</b>	
5.1 Degradation temperature ( $T_d$ ) and surface area of 2, 3, 4, and 5	66
<b>CHAPTER VI</b>	
6.1 Degradation temperature ( $T_d$ ) and ash content of 2, 3, and 4	78

## LIST OF FIGURES

FIGURE	PAGE
<b>CHAPTER III</b>	
3.1 FT-IR spectra of compounds: (a) <b>1</b> , (b) <b>2</b> , (c) <b>3</b> , (d) <b>4</b> , (e) <b>5a</b> , (f) <b>6</b> , and (g) <b>9</b> .	26
3.2 Performances <b>5</b> evaluated in the number of repeat unit (see structure in Scheme 2) for: potassium picrate ion extraction efficiency (●), degradation temperature before complexation (▲), and degradation temperature after complexation (■).	28
3.3 Extraction efficiency of <b>1</b> , <b>5a</b> , and <b>6 - 11</b> for metal picrates: K <sup>+</sup> (●), Na <sup>+</sup> (○), and, Cs <sup>+</sup> (▼).	30
3.4 Extraction efficiency of <b>1</b> , <b>5a</b> , <b>9</b> , and <b>11</b> for: Cr <sup>3+</sup> (●), Cu <sup>2+</sup> (○), Ni <sup>2+</sup> (▼), and, Ag <sup>+</sup> (∇).	31
<b>CHAPTER IV</b>	
4.1 FTIR spectra of (a) chitin flakes, (b) <b>2</b> , (c) <b>3</b> , and (d) chitosan flakes (95%DD).	40
4.2 <sup>1</sup> H NMR spectrum of <b>3</b> .	41
4.3 (A) FTIR spectra and the curve fitting of <b>3</b> under various alkaline treating temperatures for the alkaline treating time of 21 h: (a) 100°C, (b) 120°C, (c) 140°C, (d) 160 °C, and (e) 180°C; (B) quantitative analysis of <b>3</b> based on FTIR (●) and the degree of deacetylation of <b>3</b> resulted from <sup>1</sup> H NMR (■); (C) degree of deacetylation of <b>3</b> evaluated by <sup>1</sup> H NMR under various alkaline treating times for the alkaline treating temperature of 150°C.	42

FIGURE	PAGE
4.4 WAXD diffractograms of (a) <b>2</b> , (b) chitin flakes, (c) <b>3</b> , and (d) chitosan flakes (95%DD).	43
4.5 TEM micrographs of (a) <b>2</b> in water, (b) <b>3</b> in water, and (c) <b>3</b> in DMSO.	44
4.6 SEM micrographs at 15 kV of (a) chitin flakes ( $\times 1,500$ ), (b) chitosan flakes 95% DD ( $\times 1,500$ ), (c) <b>2</b> ( $\times 20,000$ ), and (d) <b>3</b> ( $\times 20,000$ ).	45
4.7 Relative viscosity of <b>3</b> by varying (a) alkaline treating temperature when the alkaline treating time was 21 h and (b) alkaline treating time when the reaction temperature was 150°C.	47
4.8 Turbidity of <b>3</b> in (a) distilled water ( $\circ$ ), (b) NaCl 0.05 M ( $\blacksquare$ ), (c) NaCl 0.1 M ( $\blacktriangle$ ), (d) NaCl 1.0 M ( $\square$ ), and (e) NaCl 5.0 M ( $\bullet$ ).	50

#### CHAPTER V

5.1 TEM micrographs of (A) (a) <b>2</b> and (d) <b>3</b> and SEM micrographs (B) (a) <b>2</b> ( $\times 20,000$ ) and (b) <b>3</b> ( $\times 20,000$ ).	59
5.2 $^1\text{H}$ NMR spectrum of <b>3</b> .	60
5.3 $^1\text{H}$ NMR spectrum of (a) <b>4</b> and (b) <b>5</b> .	62
5.4 WAXD patterns of (a) <b>2</b> , (b) <b>3</b> , (c) <b>4</b> , and (d) <b>5</b> .	64
5.5 TEM micrographs of (a) <b>4</b> and (b) <b>5</b> .	64
5.6 Appearances of (a) <b>3</b> and (b) <b>4</b> after lyophilization.	65
5.7 SEM micrographs at 15 kV of (a) <b>4</b> ( $\times 35$ ), (b) <b>4</b> ( $\times 500$ ), (c) <b>5</b> ( $\times 100$ ), and (d) <b>5</b> ( $\times 500$ ).	66

#### CHAPTER VI

6.1 Appearance of <b>2</b> in water.	74
6.2 FT-IR spectra of: (a) <b>2</b> , (b) <b>3</b> , and (c) <b>4</b> .	74

FIGURE	PAGE
6.3 X-ray diffractograms of: (a) <b>2</b> , (b) <b>3</b> , and (c) <b>4</b> .	76
6.4 Transmission electron microscopy (TEM) micrographs of: (a) <b>2</b> , (b) <b>3</b> , and (c) <b>4</b> .	77
6.5 (A) Transmission electron microscopy (TEM) micrographs of <b>2</b> after lyophilization and dispersion in: (a) deionized water and (b) chloroform, and (B) of <b>4</b> after lyophilization and dispersion in: (a) deionized water and (b) chloroform.	77