



UNIVERSITI PUTRA MALAYSIA

***NANO-ENCAPSULATION OF PARACETAMOL IN L-POLY LACTIC
ACID USING SUPERCRITICAL ANTI-SOLVENT METHOD***

MAHSHID KALANI

FK 2011 19

**NANO ENCAPSULATION OF PARACETAMOL IN L-
POLY LACTIC ACID USING SUPERCRITICAL ANTI-
SOLVENT METHOD**



MAHSHID KALANI

**MASTER OF SCIENCE
UNIVERSITI PUTRA MALAYSIA**

2011

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

**NANO-ENCAPSULATION OF PARACETAMOL IN L-POLY LACTIC ACID
USING SUPERCRITICAL ANTI-SOLVENT METHOD**

By

MAHSHID KALANI

March 2011

Chairman: Associate Professor Robiah Yunus, PhD

Faculty: Faculty of Engineering

The present work demonstrates and discusses the encapsulation of a model drug using a supercritical anti-solvent method (SAS). In this study, paracetamol was chosen as a model drug and was encapsulated in the poly L lactic acid (L-PLA), a semi crystalline polymer, under different process parameters namely pressure, temperature, and polymer concentration. The produced nanoparticles were completely spherical with very small size (300 nm) and narrow distribution. Also, the optimum process parameters to produce the smaller particle size were studied by response surface methodology (RSM)

statistical software. The nano encapsulated paracetamol release profile demonstrated a long and sustained release of drug in which 70% of paracetamol release was recorded in 4 weeks. The drug release profile of paracetamol inside the PBS buffer solution was fitted with Korsmeyer Peppas kinetic model based on the R^2 value equal to 0.987. The first burst happened after 1 week. The size and morphology of the encapsulated nano-particles were characterized by scanning electron microscopy. Transmission electron microscopy (TEM) revealed the internal structure of nano-encapsulated paracetamol and verified the full coating of the drug particle with biodegradable polymer. The results demonstrated that increasing the pressure and decreasing the temperature reduce the mean particle size. These results also showed that the particle size is influenced by the degree of super-saturation and initial polymer concentration, simultaneously. Thus, it is crucial to balance the rate of crystallization and the rate of growth. The optimum process parameters to produce minimum mean particle size (301nm) were obtained at 120 bar, 30°C, and 16 ppm polymer concentration based on both SEM images and statistical analysis. The thermal characteristics of nano-particles were investigated via differential scanning calorimetry (DSC) and thermo gravimetric analysis (TGA). The TGA characteristics of nanoparticles were similar to the TGA characteristic of pure polymer due to the higher ratio of polymer in solute. Conversely, the DSC characteristics of nano-particles were similar to paracetamol characteristic due to the higher heat capacity of paracetamol. Based on the DSC thermograms, the intensity of the endothermal melting peak of pure paracetamol was considerably reduced during SAS process due to the changing of nano-particles structure with respect to the pure L-PLA. This issue was also confirmed by the X-ray diffraction pattern as well. All peaks related to both polymer and drug crystallographs were exhibited in the nano-particles crystallography. Fourier transform infrared spectroscopy (FTIR) investigated the

chemical composition of nano-encapsulated paracetamol inside L-PLA. The positions of spectra peaks in FTIR for the encapsulated paracetamol were similar to the absorption peaks of pure polymer due to the high ratio of polymer over drug. The stability of nanoparticles demonstrated by high negative electric charge (-33 ± 3 mV) on the surface of nano particles was confirmed by means of zeta potential characteristic. This high negative surface charge may be due to the presence of carboxyl end groups of L- PLA chain.



Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

**PENKAPSULAN NANO PARASETAMOL DI DALAM ASID L POLI LAKTIK
MENGUNAKAN KAEDAH GENTING ANTI PELARUT**

Oleh

MAHSHID KALANI

Mac 2011

Pengerusi: Profesor Madya Robiah Yunus, PhD

Fakulti: Kejuruteraan

Kajian ini menunjukkan dan membincangkan pengkapsulan model ubat menggunakan kaedah genting anti-pelarut (SAS). Dalam kajian ini, parasetamol dipilih sebagai model ubat yang dikapsulkan di dalam L-PLA (asid L-poli laktik), suatu polimer semi kristal, di bawah parameter proses yang berbeza iaitu tekanan, suhu dan kepekatan polimer.

Partikel nano yang dihasilkan adalah berbentuk sfera sepenuhnya bersaiz sangat kecil (300 nm) dan pengagihan sempit. Parameter proses yang optimum untuk menghasilkan saiz partikel yang lebih kecil dikaji menggunakan perisian komputer statistik RSM. Profil pembebasan partikel nano parasetamol menunjukkan pembebasan ubat yang lama dan tertahan di mana 70% pembebasan parasetamol dicatatkan dalam tempoh 4 minggu.

Penghasilan parasetamol di dalam larutan penimbal PBS dipadankan bersesuaian dengan model kinetik Korsmeyer Peppas berdasarkan nilai R^2 bersamaan dengan 0.987. Pemecahan pertama telah berlaku selepas 1 minggu. Saiz dan morfologi partikel nano yang dikapsulkan telah dicirikan menggunakan SEM. TEM menunjukkan struktur dalaman partikel nano parasetamol dan membuktikan litupan penuh partikel ubat dengan polimer terbiodegradasi. Keputusan menunjukkan peningkatan tekanan dan penurunan suhu mengurangkan saiz partikel purata. Tambahan pula, keputusan ini menunjukkan bahawa saiz partikel dipengaruhi secara serentak oleh penepuan lampau dan kepekatan awal polimer. Jadi, adalah kritikal untuk mengimbangkan antara kadar penghabluran dan kadar pertumbuhan.

Parameter proses yang optimum untuk menghasilkan purata saiz partikel minimum (301 nm) diperolehi pada 120 bar, 30°C dan kepekatan polimer sebanyak 16 ppm berdasarkan imej SEM dan analisis statistik. Pencirian terma partikel nano telah dikaji dengan DSC dan TGA. Ciri-ciri TGA partikel nano adalah serupa dengan ciri-ciri TGA polimer tulen disebabkan nisbah polimer yang lebih tinggi di dalam zat terlarut. Ciri-ciri DSC partikel nano adalah serupa dengan ciri-ciri parasetamol disebabkan muatan haba yang tinggi pada parasetamol. Berdasarkan thermogram DSC, takat lebur keamatan endotermik parasetamol disifatkan berkurangan semasa proses SAS disebabkan perubahan struktur partikel nano terhadap L-PLA tulen. Isu ini dipastikan dengan pembelauan X-ray. Semua puncak di dalam kristalgraf kedua-dua polimer dan ubat telah ditunjukkan dalam kristalografi partikel nano. Komposisi kimia parasetamol yang dinanokapsulkan dalam L-PLA telah dikaji dengan FTIR. Posisi puncak spektra parasetamol yang dikapsulkan adalah serupa dengan puncak penyerapan polimer tulen disebabkan nisbah polimer yang lebih tinggi berbanding ubat. Kestabilan partikel nano parasetamol –L-PLA yang diproses menggunakan SCF mengandungi potensi zeta

negatif yang tinggi (-33 ± 3 mV), telah dipastikan dengan ciri-ciri potensi zeta. Cas permukaan negatif yang tinggi ini boleh diterangkan dengan kehadiran kumpulan berfungsi karboksil pada hujung rantaian L-PLA.



ACKNOWLEDGEMENT

All praises and thanks be to Allah (S.W.T), who has guided me to complete this study, never could I have found guidance, were it not that Allah had guided me!

Words cannot express my gratitude towards my supervisor Associate Professor Dr. Robiah Yunus for the patience, humble supervision and friendly advices that I received from her in both the course of this project and problems in life. May the sky be your limits in all your future endeavors and may jannatul-firdaus be your abode in the hereafter.

Furthermore, I appreciate a lot from my co-supervisor Associate Professor Dr. Norhafizah for her helpful advices in my thesis.

Also, I would like to thank the technician in the analytical lab, Mr. Adli and all of my friends for constantly providing helping hands during my laboratory sessions and of course to the rest of the staff, academic and non-academic of the faculty of engineering, UPM.

Finally, my acknowledgment will be incomplete without my husband, my son, and my parents on support and help throughout my study.

I certify that a Thesis Examination Committee has met on 24 March 2011 to conduct the final examination of Mahshid Kalani on her thesis entitled “Nano-Encapsulation of Paracetamol in L-Polylactic Acid using Supercritical Anti-Solvent Method “in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

Members of the Thesis Examination Committee were as follows:

Tey Beng Ti, PhD

Associate Professor
Faculty of Engineering
Universiti Putra Malaysia
(Chairman)

Tey Beng Ti, PhD

Associate Professor
Faculty of Engineering
Universiti Putra Malaysia
(Internal Examiner)

Arbakariya Ariff, PhD

Professor
Faculty of Biotechnology & Biomolecular Sciences
Universiti Putra Malaysia
(Internal Examiner)

Ir. Abdul Wahab Mohammad, PhD

Professor
Faculty of Engineering
Universiti Kebangsaan Malaysia
(External Examiner)

NORITA OMAR, PhD

Associate Professor and Deputy Dean
School Of Graduate Studies
Universiti Putra Malaysia

Date: 24 May 2011

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the Degree of Master of Science. The members of the Supervisory Committee were as follows:

Robiah Yunus, PhD

Associate Professor

Faculty of Engineering

Universiti Putra Malaysia

(Chairman)

Norhafizah Abdullah, PhD

Associate Professor

Faculty of Engineering

Universiti Putra Malaysia

(Member)

HASANAH MOHD GHAZALI, PhD

Professor and Dean

School Of Graduate Studies

Universiti Putra Malaysia

Date: 9 JUNE 2011

DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.

MAHSHID KALANI

Date: 24 March 2011

TABLE OF CONTENTS

	Page
ABSTRACT	i
ABSTRAK	iv
ACKNOWLEDGEMENT	vii
APPROVAL	viii
DECLARATION	x
LIST OF TABLES	xv
LIST OF FIGURES	xv
LIST OF ABBREVIATIONS	xv
 CHAPTER	
 1. INTRODUCTION	 1
1.1 Introduction	1
1.2 Problem Statement	3
1.3 Objective	5
1.4 Scope of Study	5
1.5 Outline of Thesis	6
 2. LITERATURE REVIEW	
2.1 Introduction	8
2.2 Super Critical Fluid Properties	11
2.3 The Antisolvent Supercritical Fluid Process	13
2.3.1 Supercritical Antisolvent Process (SAS)	14
2.4 Thermodynamic of SAS	31
2.4.1 Two Component Vapor-Liquid	34
2.4.2 Three Component Phase Equilibrium	35
2.5 Crystallization Mechanisms	39
2.5.1 Nucleation Kinetics	39

2.5.2	Crystal Growth	42
2.5.3	Crystal Morphology	42
2.6	Effects of Process Parameters on Particle Size	43
2.6.1	Effects of Pressure and Temperature	43
2.6.2	Effects of Concentration	48
2.6.3	Effects of Chemical Composition of the Organic Solvent	49
2.6.4	Effects of Chemical Composition of the Solute (Drug and Biodegradable Polymer)	50
2.6.5	Effects of the Nozzle Geometry	52
2.6.6	Effects of Flow Rates of CO ₂ and Liquid Phase	54
2.7	Conclusion	59
3.	MATERIALS AND METHODS	
3.1	Material	60
3.2	Apparatus and Procedures	61
3.3	Experimental Design Parameters	65
3.4	Characterization Methods	69
3.4.1	Scanning Electron Microscopy (SEM)	69
3.4.2	Transmission Electron Microscopy	69
3.4.3	Thermo Gravimetric Analysis	69
3.4.4	Differential Scanning Calorimetry	70
3.4.5	X-ray Diffraction Pattern	71
3.4.6	Fourier Transform Infrared (FTIR) Spectroscopy	71
3.4.7	Zeta Potential	71
3.4.8	In Vitro Drug Release	72
4.	RESULTS AND DISCUSSIONS	
4.1	Basis Selection of Materials and Method	73
4.2	Characterization	74
4.2.2	Transmission Electron Microscopy	90
4.2.3	Thermo Gravimetric Analysis	91
4.2.4	Differential Scanning Calorimetry	94

4.2.5	X-ray Diffraction	98
4.2.6	Fourier Transform Infrared Spectroscopy	101
4.2.7	Zeta Potential	103
4.2.8	In Vitro Drug Release	104
5. CONCLUSIONS AND RECOMMENDATIONS		
5.1	General discussion	109
5.2	Conclusions	109
5.3	Recommendations	113
REFERENCES		115
Appendix A		125
Appendix B		128
Appendix C		131
Appendix D		134
Appendix E		137
Appendix F		139
BIODATA OF STUDENT		143
LIST OF PUBLICATIONS		144