

**SURFACE-MODIFIED ELECTROSPUN POLYCAPROLACTONE FIBROUS  
MEMBRANES MODIFIED WITH GELATIN, BOVINE SERUM ALBUMIN  
OR CRUDE BONE PROTEIN EXTRACT AND THEIR POTENTIAL FOR  
USE AS BONE SCAFFOLDS**



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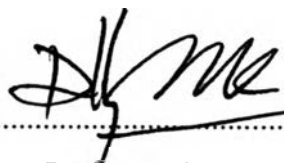
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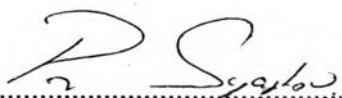
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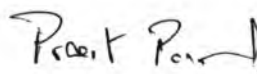


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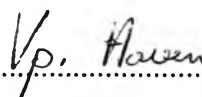
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## ABSTRACT

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Immobilization of biomolecules; i.e. gelatin type-A, gelatin type-B, bovine serum albumin (BSA) and crude bone protein (CBP), making polycaprolactone (PCL) fibrous scaffolds that have been fabricated by electrospinning more suitable for bone tissue engineering. PCL scaffolds were first covalently introduced with amino groups on their surfaces through the aminolysis reaction using 1,6-hexamethylenediamine (HMD) and later immobilized with the above mentioned biomolecules using disuccinimidyl carbonate (DSC) as the coupling agent. Various techniques; ATR-FTIR, XPS, SEM, and water contact angle measurement were used to monitor the scaffold surfaces after each modification step. The potential use of the modified materials as bone scaffolds was evaluated with a murine pre-osteoblastic cell line (MC3T3-E1). MC3T3-E1 proliferation was improved remarkably on the modified surface, especially the BSA-immobilized PCL fibrous scaffolds which showed the greatest proliferation after cell culture as well as the highest ALP activity. In mineralization, the deposited minerals was highest on the CBP-immobilized PCL scaffolds. All the obtained results suggested that immobilization of BSA and CBP is an attractive method for fabricating of fibrous scaffolds for bone tissue engineering.

## บทคัดย่อ

สุทธิลักษณ์ ไชยชำนาญ : การปรับปรุงพื้นผิวแผ่นเส้นใยพอลิคาโพรแลคโตนอิเล็กโตรสปินด้วยเจลาติน, โบวีน เซรัม อัลบูมิน หรือโปรตีนสกัดจากกระดูก และความสามารถในการใช้เป็นวัสดุโครงร่างสำหรับเซลล์กระดูก (Surface-Modified Electrospun Polycaprolactone Fibrous Membranes Modified with Gelatin, Bovine Serum Albumin or Crude Bone Protein Extract and Their Potential for Use as Bone Scaffolds) อ. ที่ปรึกษา : ศ. ดร. พิชญ์ สุภผล และ รศ. ดร. ประสิทธิ์ ภาสันต์ 86 หน้า

เพื่อเพิ่มความสามารถในการเป็นวัสดุโครงร่างสำหรับเซลล์กระดูกของแผ่นเส้นใยพอลิคาโพรแลคโตน ซึ่งเตรียมได้โดยวิธีการปั่นเส้นใยด้วยไฟฟ้าสถิต ทำโดยการเพิ่มหมู่อะมิโนไปบนพื้นผิวของแผ่นเส้นใยก่อน ด้วยการทำปฏิกิริยาอะมิโนไลซิสกับ 1,6-เฮกซะเมทิลีน ไดเอมีน (HMD) หลังจากนั้นสารชีวโมเลกุลขนาดใหญ่ เช่น เจลาตินชนิดเอ, เจลาตินชนิดบี, โบวีน เซรัม อัลบูมิน (bovine serum albumin) หรือ โปรตีนสกัดจากกระดูก (crude bone protein) ได้ถูกตรึงโดยใช้ ไคซึกซิมิติดิล คาร์บอเนต (DSC) เป็นสารคู่ควบ หาความหนาแน่น ความมีรูพรุน และปริมาตรของรูพรุน เพื่อ พิสูจน์เอกลักษณ์ของแผ่นเส้นใยพอลิคาโพรแลคโตนอิเล็กโตรสปิน เทคนิค เอทีอาร์เอฟ ทีไออาร์ สเปกโทรสโกปี (ATR-FTIR), เอกซเรย์โฟโตอิเล็กตรอน สเปกโทรสโกปี (XPS), การส่องกล้องจุลทรรศน์อิเล็กตรอนแบบสแกนนิ่ง (SEM), และการวัดมุมสัมผัสกับน้ำ ถูกนำมาใช้เพื่อตรวจสอบพื้นผิวของแผ่นเส้นใยหลังจากได้รับการปรับปรุงพื้นผิวแล้วในแต่ละขั้นตอน แผ่นเส้นใยพอลิคาโพรแลคโตนอิเล็กโตรสปิน ถูกนำมาทดสอบความสามารถในการเป็นวัสดุโครงร่างสำหรับเซลล์กระดูก โดยใช้เซลล์กระดูกของหนู (MC3T3-E1) ผลการทดสอบพบว่าเซลล์กระดูกของหนูที่ถูกเลี้ยงบนพื้นผิวแผ่นเส้นใยที่ได้รับการปรับปรุงมีการเจริญเติบโตที่ดียิ่งขึ้นได้ชัดเจนเมื่อเทียบกับแผ่นเส้นใยที่ไม่ได้รับการปรับปรุงพื้นผิวและตัวควบคุม (จานเลี้ยงเซลล์) โดยที่เซลล์ที่เลี้ยงบนแผ่นเส้นใยที่ได้รับการปรับปรุงด้วยโบวีน เซรัม อัลบูมิน จะเจริญเติบโตได้มากที่สุดและเอแอลพีเอกติวิตี (ความสามารถในการเปลี่ยนแปลงหน้าทีไปเป็นเซลล์กระดูก) มากที่สุดเช่นกัน ในการทดลองหาปริมาณแร่ธาตุที่เซลล์เปลี่ยนแปลงไปเป็นในระยะเวลา 21 วัน พบมากที่สุดใแผ่นเส้นใยพอลิคาโพรแลคโตนอิเล็กโตรสปินที่ได้รับการปรับปรุงด้วยโปรตีนสกัดจากกระดูก ผลการทดสอบแสดงให้เห็นว่า แผ่นเส้นใยพอลิคาโพรแลคโตนอิเล็กโตรสปิน ที่ได้รับการปรับปรุงด้วยโบวีน เซรัม อัลบูมินและโปรตีนสกัดจากกระดูก เป็นวัสดุที่น่าสนใจในการนำไปทำวัสดุโครงร่างสำหรับเซลล์กระดูกและเพิ่มการทำงานของเซลล์กระดูกได้ดี

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## TABLE OF CONTENTS

	<b>PAGE</b>
Title Page	i
Abstract (in English)	iii
Abstract (in Thai)	iv
Acknowledgements	v
Table of Contents	vi
List of Tables	xi
List of Figures	xiii
 <b>CHAPTER</b>	
<b>I INTRODUCTION</b>	<b>1</b>
<b>II LITERATURE REVIEW</b>	<b>3</b>
2.1 Tissue Engineering	3
2.2 Basic Bone Biology	7
2.2.1 Bone Cells	7
2.2.2 Extracellular Matrix	8
2.2.2.1 Organic Substance contains 25% of ECM	8
2.2.2.2 Inorganic Substance contains 70 % of ECM	8
2.3 Bone Remodelling	10
2.3.1 Process of Bone Remodeling	10
2.3.2 Bone Remodeling and the Osteoclast	11
2.3.3 Bone Remodeling and the Osteoblast	11
2.4 Ossification	12
2.5 Scaffold Preparation Methods	14
2.5.1 Solvent Casing	14
2.5.2 Solvent-Casting Particulate-Leaching	14
2.5.3 Gas Foaming	15

<b>CHAPTER</b>	<b>PAGE</b>
<b>II</b>	<b>LITERATURE REVIEW</b>
2.5.4	Gas Foaming/Salt Leaching Method 15
2.5.5	Freeze Drying 15
2.5.6	Emulsification/Freeze-drying 16
2.5.7	Thermally Induced Phase Separation (TIPS) 16
2.5.8	Phase Separation/Emulsification 17
2.5.9	Solid Freeform Fabrication 17
2.6	Electrospinning 18
2.7	Tissue Engineering Scaffold Materials 20
2.8	Polycaprolactone (PCL) 22
2.9	Surface Modification 23
2.10	General Description of Proteins Immobilized onto PCL Scaffolds 25
2.10.1	Gelatin 25
2.10.2	Bovine Serum Albumin (BSA) 27
2.11	Immobilization of Biomolecules onto Polyester Surface 28
<b>III</b>	<b>EXPERIMENTAL</b> 34
3.1	Materials 34
3.1.1	Materials used in the Fibrous Scaffolds Preparation and Surface Modification 34
3.1.2	Materials used for cell culture 34
3.1.2.1	Model Cells 34
3.1.2.2	Medium for MC3T3-E1 cells 34
3.1.2.3	Material for Cell Culture Study 35
3.2	Equipment 35
3.2.1	Equipment for Electrospinning Process 35
3.2.2	Equipment for Characterization of Materials 36

<b>CHAPTER</b>	<b>PAGE</b>
<b>III EXPERIMENTAL</b>	
3.2.3 Equipment for Study of Cell Culture	36
3.3 Experimental Procedures	36
3.3.1 Preparation of Polycaprolactone Scaffolds	36
3.3.1.1 Preparation and Characterization of Fibrous Scaffolds	36
3.3.1.2 Preparation of Crude Bone Protein.	37
3.3.1.3 Surface Modification of PCL Scaffold via Aminolysis and Immobilization of Proteins	37
3.3.2 Characterization of Fibrous Scaffolds	38
3.3.2.1 Density, Porosity and Pore Volume	38
3.3.2.2 Water Retention Capacity	39
3.3.2.3 Degradation Study of Fibrous Scaffolds	39
3.3.3 Determination of the Amino Groups on PCL Surface after Aminolysis and Protein Immobilization	40
3.3.4 Surface Characterization	40
3.3.4.1 Water Contact Angle Measurements	40
3.3.4.2 UV-Vis Spectrophotometer	40
3.3.4.3 Attenuated Total Reflectance-Fourier Transform Infrared Spectrometer (ATR-FTIR)	40
3.3.4.4 X-ray Photoelectron Spectrometer (XPS)	41
3.3.4.5 Scanning Electron Microscope	41
3.3.5 Cell Culture Studies	41
3.3.5.1 Cytotoxicity Evaluation	41
3.3.5.1.1 Direct Cytotoxicity	41
3.3.5.1.2 Indirect Cytotoxicity	42
3.3.5.2 Cell Adhesion and Proliferation	42
3.3.5.3 MTT Assay	43



<b>CHAPTER</b>	<b>PAGE</b>
<b>III</b>	<b>EXPERIMENTAL</b>
3.3.5.4	Morphological Observation of Cultured Cells 43
3.3.5.5	Production of Alkaline Phosphatase of Cultured Cells 43
3.3.5.6	Mineralization 44
3.3.5.7	Statistical analysis 45
<b>IV</b>	<b>RESULTS AND DISCUSSION</b> 46
4.1	Preparation of Poly( $\epsilon$ -caprolactone) Electrospun Fibrous Scaffold 46
4.2	Characterization of Fibrous Scaffolds 47
4.2.1	Density, Porosity, and Pore Volume 47
4.2.2	Water Retention Capability 48
4.2.3	Degradation Study of PCL Fibrous Scaffolds 48
4.3	Surface Characterization 50
4.3.1	Quantification of Amino Groups 50
4.3.2	Surface Wettability 51
4.3.3	Chemical Analysis of Surface 54
4.3.4	Elemental Composition of the Surface 56
4.4	Biological Characterizations 57
4.4.1	Cytotoxicity 57
4.4.1.1	Direct Cytotoxicity 57
4.4.1.2	Indirect Cytotoxicity 57
4.4.2	Cell Attachment and Proliferation 59
4.4.3	Cell Morphology 61
4.4.4	Alkaline Phosphatase Activity (ALP) 65
4.4.5	Mineralization 66

<b>CHAPTER</b>		<b>PAGE</b>
<b>V</b>	<b>CONCLUSIONS AND RECOMMENDATIONS</b>	<b>68</b>
	4.5 Conclusions	68
	4.6 Recommendations	69
	<b>REFERENCES</b>	<b>70</b>
<b>APPENDICES</b>		
	<b>Appendix A</b> Experimental Data of Density, Porosity and Pore Volume	<b>75</b>
	<b>Appendix B</b> Experimental Data of Water Retention Capacity and Weight Loss of Polycaprolactone Electrospun Fibrous Scaffolds	<b>77</b>
	<b>Appendix C</b> Experimental Data of NH <sub>2</sub> Density, Elemental Composition of the Surface and Water Contact Angle Measurement	<b>79</b>
	<b>Appendix D</b> Experimental Data of Cell Culture Studies	<b>82</b>
	<b>CURRICULUM VITAE</b>	<b>86</b>

## LIST OF TABLES

TABLE	PAGE
2.1 Polyester-type polymer in many type of forms and preparation methods used for tissue engineering application	6
4.1 The density, porosity and pore volume of the unmodified PCL scaffolds and modified PCL scaffolds	48
4.2 The water contact angle of the control and all modified PCL fibrous scaffolds measured by the sessile drop method	53
4.3 $N_{1s}/C_{1s}$ ratios as a function of 1,6-hexanediamine concentration	56
4.4 Selected SEM images of MC3T3-E1 cultured on the specimens, i.e., glass (control), neat PCL, aminolyzed PCL, activated PCL, and protein-immobilized PCL fibrous scaffolds at various time points (magnification = 2,000X; scale bar = 10 $\mu$ m)	63
4.5 Selected SEM images of MC3T3-E1 cultured on the specimens, i.e., glass (control), neat PCL, aminolyzed PCL, activated PCL, and protein-immobilized PCL fibrous scaffolds at various time points (magnification = 2,000X; scale bar = 10 $\mu$ m)	64
A1 Raw data of the density, porosity and pore volume of the unmodified PCL scaffolds and modified PCL scaffolds	76
B1 Raw data of water retention capacity of aminolyzed PCL electrospun fibrous scaffolds at various HMD treatments	78
B2 Raw data of water retention capacity of aminolyzed PCL electrospun fibrous scaffolds at various HMD treatments	78

<b>TABLE</b>	<b>PAGE</b>
C1 NH <sub>2</sub> concentration and density as a function of 1,6-hexamethylenediamine	80
C2 Elemental compositions of C <sub>1s</sub> , N <sub>1s</sub> , O <sub>1s</sub> and N <sub>1s</sub> /C <sub>1s</sub> ratio on the surface of aminolyzed PCL fibrous scaffolds as a function of 1,6-hexamethylenediamine	80
C3 Elemental compositions of C <sub>1s</sub> , N <sub>1s</sub> , O <sub>1s</sub> and N <sub>1s</sub> /C <sub>1s</sub> ratio on the surface of the neat, aminolyzed, and all types of immobilized PCL scaffolds.	81
D1 Raw data of direct cytotoxicity test of proteins, which were immobilized onto the PCL fibrous scaffolds, determined the viability of cells by MTT assay method at 570 nm	82
D2 Raw data of indirect cytotoxicity test of all types of PCL fibrous scaffolds, determined the viability of cells by MTT assay method at 570 nm	82
D3 Raw data of cell attachment of MC3T3-E1 onto all types of PCL fibrous scaffolds at 2, 4, and 6 hours, determined the viability of cells by MTT assay method at 570 nm	83
D4 Raw data of cell proliferation of MC3T3-E1 onto all types of PCL fibrous scaffolds at 1, 2, and 3 days, determined the viability of cells by MTT assay method at 570 nm	83
D5 Raw data of ALP Activity of MC3T3-E1 seeded onto all types of PCL fibrous scaffolds at 3 days	84
D6 Raw data of ALP Activity of MC3T3-E1 seeded onto all types of PCL fibrous scaffolds at 7 days	84
D7 Raw data of quantity of mineral deposition on all types of PCL fibrous scaffolds using Alizarin Red-S method at 570 nm	84

## LIST OF FIGURES

FIGURE	PAGE
2.1 Scaffold-guided tissue regeneration.	3
2.2 Bone matrix.	9
2.3 Three distinctly different bone cell types. Picture from <a href="http://cell.utmb.edu">http://cell.utmb.edu</a>	9
2.4 Bone remodeling mechanism.	11
2.5 Polymer film fabrication using solvent cast film.	14
2.6 The schematic of electrospinning process.	19
2.7 The structure of polycaprolactone.	22
2.8 Ring opening polymerization of $\epsilon$ -caprolactone to polycaprolactone.	22
2.9 The structure of gelatin.	26
2.10 The schematic representation of photo-induced grafting with PMAA and further immobilization with gelatin on PCL membrane surface (Zhu et al., 2002b).	29
2.11 Schematic representation of the process of UV introduced AAc grafting and collagen immobilization (Cheng et al., 2004).	30
2.12 Chemical pathway for the immobilization of different biomolecules, such as collagen, chitosan, or GRGDS peptide (i.e., H-Gly-Arg-Cly-Asp-Ser-OH), on the surface of the electrospun PCL fibrous scaffolds (Mattanavee et al., 2009).	32
4.1 Selected SEM image of electrospun PCL fibrous scaffolds (a), aminolysed PCL fibrous scaffolds which treated with (magnification = 2000x; scale bar = 10 $\mu$ m). 1,6- hexamethylenediamine (HMD) at concentration 0.04 g/ml (b), 0.20 g/ml (c) and 0.40 g/ml (d)	47

<b>FIGURE</b>	<b>PAGE</b>
4.2 Water retention capability of these unmodified and modified PCL scaffolds via aminolysis process with different 1,6-hexamethylenediamine(HMD) concentration at 37 °C within 48 hours.	49
4.3 The degradation rate of the PCL scaffolds at different HMD concentration treatments.	49
4.4 The chemical pathway for the immobilization of proteins.	50
4.5 NH <sub>2</sub> density on PCL electrospun fibrous scaffolds as a function of concentration of 1,6-hexamethylenediamine/isopropanol solution. The PCL scaffold was aminolyzed at evaluated temperature for 2 hours.	51
4.6 Water contact angles of the control and aminolyzed PCL scaffolds at different HMD concentration treatments measured by the sessile drop method.	52
4.7 Water dropped on the surface of neat PCL fibrous scaffold (a), and PCL fibrous scaffold immobilized with 3.0 mg/ml gelatin type-A (b), PCL fibrous scaffold immobilized with 3.0 mg/ml bovine serum albumin (c) and PCL fibrous scaffold immobilized with 3.0 mg/ml crude bone protein (d).	53
4.8 ATR-FTIR spectra of neat and aminolyzed PCL fibrous scaffolds with different HMD concentration treatments.	54
4.9 ATR-FTIR spectra of neat and modified PCL fibrous scaffolds.	55
4.10 The survey XPS spectra of (a) neat PCL, (b) aminolyzed PCL with 0.02 g/ml HMD treatment, (c) activated PCL, (d) PCL immobilized with 3 mg/ml gelatin type-A solution, (e) PCL immobilized with 3 mg/ml gelatin type-B solution.	56

FIGURE	PAGE
4.11 The viability of MC3T3-E1, cultured with 2% serum-containing MEM extraction media adding with all types of proteins for 1, 2 and 3 days. Statistical significance: * $p < 0.05$ compared with control and # $p < 0.05$ compared to the neat PCL fibrous scaffolds at any given time point.	58
4.12 The viability of MC3T3-E1, cultured with 2% serum-containing MEM extraction media from all type of PCL fibrous scaffolds for 1, 2, and 3 days relative to TCPS at 1 day Statistical significance: * $p < 0.05$ compared with control and # $p < 0.05$ compared to the neat PCL fibrous scaffolds at any given time point.	58
4.13 Attachment of MC3T3-E1 that had been seeded or cultured on the surfaces of TCPS and the neat and the modified PCL fibrous scaffolds for 2, 4, and 6 h. Statistical significance: * $p < 0.05$ compared with control and # $p < 0.05$ compared to the neat PCL fibrous scaffolds at any given time point.	60
4.14 Proliferation of MC3T3-E1 that had been seeded or cultured on the surfaces of TCPS and the neat and the modified PCL fibrous scaffolds for 1, 2, and 3 days. Statistical significance: * $p < 0.05$ compared with control and # $p < 0.05$ compared to the neat PCL fibrous scaffolds at any given time point.	61
4.15 Alkaline phosphatase activity (ALP) of MC3T3-E1 that were cultured on the surfaces of TCPS and the neat and the modified PCL fibrous scaffolds for 3, 5, and 7 d. Statistical significance: * $p < 0.05$ compared with control and # $p < 0.05$ compared to the neat PCL fibrous scaffolds at any given time point.	66

FIGURE	PAGE	
4.16	Quantification of mineral deposition in MC3T3-E1 at 21 days by the method of Alizarin Red-S staining measured the absorbance by UV-vis spectrometer at 570 nm. Statistical significance: * $p < 0.01$ compared with control, # $p < 0.01$ compared to the neat PCL, & $p < 0.01$ compared with gelatin type-A immobilized PCL, and + $p < 0.01$ compared with gelatin type-B immobilized PCL fibrous scaffolds at any given time point.	67
4.17	Image of Alizarin Red-S staining for the mineralization in MC3T3-E1 cells for 21 d: TCPS without (a) and with (b) cells, neat PCL without (c) and with (d) cells, aminolyzed (e) and activated PCL (f), and gelatin type-A (g), gelatin type-B (h), bovine serum albumin (i) and crude bone protein (j) immobilized PCL fibrous scaffold.	67
C1	Calibration curve of UV absorbance as a function of 1,6-hexamethylenediamine (HMD) concentration analyzed by ninhydrin assay method.	79