

**SURFACE MODIFICATION OF POLY(LACTIC ACID) FIBERS VIA
AMINOLYSIS AND TYPE-I COLLAGEN IMMOBILIZATION FOR
BONE TISSUE ENGINEERING**



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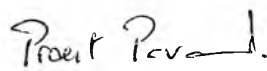
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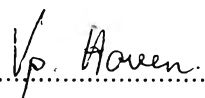
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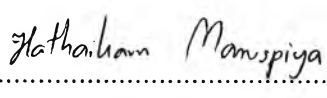

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ABSTRACT

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By the reaction of poly(lactic acid) (PLA) in the form of electrospun fibrous membranes with 1,6-hexanediamine (HMD), free amino groups were introduced onto PLA surface, through which a biocompatible macromolecule, collagen, was covalently immobilized by employing *N,N'*-disuccinimidylcarbonate (DSC) as a coupling agent. The existence of the free amino groups on the aminolyzed PLA surface was verified quantitatively by the ninhydrin analysis method, which revealed that the free amino group density were influenced by the HMD concentration and aminolyzing time. Water contact angle measurement confirmed that the hydrophilicity of the PLA fibrous membranes was enhanced with the aminolysis and the further immobilization of collagen. In addition, surface alteration of modified electrospun fibrous scaffolds was also studied by ATR-FTIR and XPS techniques. *In vitro* indirect cytotoxicity evaluation performed with mouse fibroblast (L929) and pre-osteoblastic cells (MC3T3-E1) revealed that both the neat and the modified PLA fibrous scaffolds released no substances at levels that were harmful to these cells. Scanning electron microscopy observation showed an evidence of the extension of cell cytoplasm on all types of the modified PLA fibrous surface even at 4 h after cell seeding. The culture MC3T3-E1 *in vitro* proved that the cell proliferation and cell activity of modified PLA electrospun fibers were improved compared with the neat PLA fibers. Among the various types of modified PLA scaffolds, collagen-immobilized PLA showed the greatest ability to support cell proliferation and alkaline phosphatase (ALP) activity. Therefore, it is promising material to accelerate bone regeneration.

บทคัดย่อ

ปาติดา อุประสิทธิ์วงศ์ : การดัดแปลงพื้นผิวของเส้นใยพอลิแลคติกแอซิดโดยอะมิโนไลซิสและการติดคอลลาเจนเพื่อใช้สำหรับวิศวกรรมเนื้อเยื่อสำหรับกระดูก (Surface Modification of Poly(lactic acid) fibers via Aminolysis and Type-I Collagen Immobilization for Bone Tissue Engineering) อ.ที่ปรึกษา : รศ.ดร. พิชญ์ สุภผล 66 หน้า

หมู่ะมิโนอิสระถูกเติมลงบนพื้นผิวของพอลิแลคติกแอซิด โดยการทำให้ปฏิกิริยาของแผ่นเส้นใยพอลิแลคติกแอซิดที่เตรียมจากกระบวนการปั่นเส้นใยด้วยไฟฟ้าสถิต (Electrospinning) กับเฮกซะเมทิลีนไดเอมีน (1,6-hexanediamine) ดังนั้นสารชีวโมเลกุล (เช่น คอลลาเจน) จึงสามารถเกิดพันธะโควาเลนต์กับหมู่ดังกล่าวโดยใช้ ไดซัคซินิมิดิลคาร์บอเนต (*N,N'*-disuccinimidyl-carbocnate) เป็นสารคู่ควบการวิเคราะห์หาปริมาณของหมู่ะมิโนอิสระบนผิวพอลิแลคติกแอซิดสามารถทำได้โดยใช้ไนไฮดริน (Ninhydrin) ซึ่งแสดงให้เห็นว่าความเข้มข้นของไดเอมีนที่ใช้ และเวลาในการทำปฏิกิริยาอะมิโนไลซิส มีอิทธิพลต่อความเข้มข้นของหมู่ะมิโนอิสระที่ถูกเติม จากผลของการวัดมุมสัมผัสของน้ำ สามารถยืนยันได้ว่าความชอบน้ำของแผ่นเส้นใยพอลิแลคติกแอซิดเพิ่มขึ้นหลังจากทำปฏิกิริยาอะมิโนไลซิส และการตรึงคอลลาเจน ตามลำดับ นอกจากนี้เทคนิคดังกล่าวมาแล้ว ในงานวิจัยนี้ยังใช้เทคนิค ATR-FTIR และ XPS เพื่อศึกษาการเปลี่ยนแปลงของพื้นผิวหลังทำปฏิกิริยาด้วย จากการทดสอบความเป็นพิษต่อเซลล์โดยวิธีอ้อม พบว่าแผ่นเส้นใยทุกชนิดไม่มีความเป็นพิษต่อเซลล์ การถ่ายภาพของพื้นผิววัสดุโดยกล้องจุลทรรศน์อิเล็กตรอนแบบส่องกราด แสดงให้เห็นว่าเซลล์สร้างกระดูกชนิด MC3T3-E1 ที่ทำการเพาะเลี้ยงเป็นเวลา 4 ชั่วโมง บนวัสดุที่ดัดแปลงพื้นผิวแล้ว มีการแผ่ขยายของ cytoplasm นอกจากนี้ได้ศึกษาสมบัติในการใช้เป็นวัสดุโครงสร้างสำหรับกระดูกในสภาวะนอกร่างกายด้วยเซลล์สร้างกระดูกชนิด MC3T3-E1 การเจริญเติบโต และอัลคาไลน์ฟอสฟาเตสแอคติวิตีของเซลล์ที่เพาะเลี้ยงบนแผ่นเส้นใยที่ผ่านการดัดแปลงพื้นผิวแล้วทุกชนิดดีกว่าแผ่นเส้นใยที่ยังไม่ดัดแปลงพื้นผิว ในบรรดาวัสดุที่ดัดพื้นผิวแล้วทุกชนิดพบว่า แผ่นเส้นใยพอลิแลคติกแอซิดที่ติดคอลลาเจน เกือบต่อการเจริญเติบโต และอัลคาไลน์ฟอสฟาเตสแอคติวิตีของเซลล์มากที่สุด ดังนั้นเส้นใยพอลิแลคติกแอซิดที่ติดคอลลาเจนจึงเป็นวัสดุที่น่าสนใจในการใช้เป็นวัสดุโครงสร้างสำหรับกระดูก

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