Electrospinning preparation and surface hydrophilicity investigation of poly(lactic acid), poly(3-hydroxybutyrate-o-4-hydroxybutyrate), and poly(propylene carbonate) nanometer fiber membranes*☆

Li Nan[1], Wang Xue-ming[1], Qi Hong-xu[2], Zhai Jun-shan[1], Wang Yan[2], Hu Ping[2], Zhu Jian-hua[1]

Abstract

BACKGROUND: Recently, poly (lactic acid)/hydroxyapatite composite scaffolds have been widely used due to good biodegradation and biocompatibility. But poly (lactic acid)/hydroxyapatite composite material can not satisfy the requirements of ideal tissue-engineered scaffold materials owing to drawbacks in some aspects including enhancing material surface binding, adjusting material degradation velocity, and improving material intensity.

OBJECTIVE: To investigate the structural morphology and surface hydrophilicity of nanometer fiber membrane prepared by electrospinning.

METHODS: Poly(lactic acid), poly (3-hydroxybutyrate-co-4-hydroxybutyrate), and poly(propylene carbonate) nanometer fiber membranes were respectively prepared by electrospinning. The structural morphology of these three nanometer fiber membranes, as well as the surface hydrophilicity after soaking in phosphate buffered saline (37 ℃, pH 7.4) similar to human body environment for different time periods, was observed through the use of scanning electron microscope.

RESULTS AND CONCLUSION: Poly(lactic acid), poly(3-hydroxybutyrate-co-4-hydroxybutyrate), and poly(propylene carbonate) could be prepared into micro- and nano-sized fiber structure by electrospinning. Different fiber diameters of nanometer fiber membranes could be produced by controlling preparation parameters. With the prolonged soaking time in culture medium, the contact angle of three fiber members was reduced greatly and surface hydrophilicity was gradually enhanced.

[1]Department of Gastroenterology, PLA General Staff Headquarter General Hospital (309th Hospital of Chinese PLA), Beijing 100091, China; [2]Institute of Polymer Science and Technology, Department of Chemical Engineering, Tsinghua University, Beijing 100084, China

Li Nan☆, Doctor, Professor, Chief physician, Doctoral Supervisor, Department of Gastroenterology, PLA General Staff Headquarter General Hospital (309th Hospital of Chinese PLA), Beijing 100091, China
linan957@sina.com

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INTRODUCTION

Preparation of scaffold materials is one of core techniques in tissue engineering research. Homogenous material can hardly satisfy the requirements of tissue-engineered scaffold materials, so a hot spot of current scaffold material research is to compound materials with different properties to harvest composite scaffold materials with new properties[1-2].

Recently, poly (lactic acid) (PLA)/hydroxyapatite composite scaffolds have been widely used due to good biodegradation and biocompatibility. The commonly used preparation methods primarily include cold compression, particle leaching, and thermally induced phase separation. But PLA/hydroxyapatite composite material can not satisfy the requirements of ideal tissue-engineered scaffold materials owing to drawbacks in some aspects including enhancing material surface binding, adjusting material degradation velocity, and improving material intensity[3-4]. For this reason, novel preparation technology is needed. Nanometer fiber membrane prepared by electrospinning has become an increasing research area in tissue engineering scaffold materials owing to high porosity, adjustable pore diameter, and good biocompatibility[3]. Cells exhibit better adhesive capacity on fibers with fiber diameter (micrometer-sized) less than cell diameter. Therefore, the porous three-dimensional cell scaffold prepared by electrospinning can better imitate the structural characteristics of natural extracellular matrix, hopefully becoming an ideal tissue engineering scaffold[5-13].

The present study prepared PLA, poly(3-hydroxybutyrate-co-4-hydroxybutyrate) [P(3HB-co-4HB)], and poly(propylene carbonate) (PPC) nanometer fiber membranes by electrospinning and studied the structural morphology and surface hydrophilicity of nanometer fiber membranes, hopefully harvesting ideal cell scaffold materials, fully exerting the structure and property of PLA, P(3HB-co-4HB), and PPC composites, and embodying the nanometer effect characterization of PLA, P(3HB-co-4HB), and PPC nanometer fibers in tissue-engineered scaffold materials and during the process of cell attachment, growth, and proliferation.

MATERIALS AND METHODS

Design

Material property characterization, observational experiment.

Time and setting

This study was performed at the Department of Bioengineering, Gastroenerology Center, PLA General Staff Headquarter General Hospital, and Institute of Polymer Science and Technology, Department of Chemical Engineering, Tsinghua University, as well as the National Food Quality Supervision and Inspection Center between June 2007 and October 2009.

Materials

The primary materials and instruments used in this study are as follows:

Material/instrument	Source
PLA (relative molecular mass 500 000)	Shandong Medical Equipment Quality Surveillance Detection Center, China
P(3HB-co-4HB)(relative molecular mass 350 000, copolymer segment ratio 9/1)	Tianjin Guorun Biological Materials Co., Ltd., China
PPC(relative molecular mass 200 000)	Inner Mongolia Melic Sea High-Tech Co.,Ltd., China
Electrospinning instrument	Beijing Research Institute of Mechanical and Electrical Technology, China
Scanning electron microscope (S-3400)	Hitachi, Japan

Methods

Electrospinning preparation of PLA fiber

1 g PLA was fully dissolved in 10 mL methylene chloride. Then the mixture was prepared into fibers through the use of electrospinning instrument, with the precise parameters: voltage 10 kV, distance from spout to receiver 15 cm, and 9# flat needle as spout.

Electrospinning preparation of P(3HB-co-4HB) fiber

1 g P(3HB-co-4HB) was fully dissolved in 10 mL methylene chloride. Then the mixture was prepared into fibers through the use of electrospinning instrument, with the precise parameters: voltage 13 kV, distance from spout to receiver 20 cm, and 12# flat needle as spout.

Electrospinning preparation of PPC fiber

1 g PPC was fully dissolved in 10 mL methylene chloride. Then the mixture was prepared into fibers through the use of electrospinning instrument, with the precise parameters: voltage 8 kV, distance from spout to receiver 10 cm, and 12# flat needle as spout.

In vitro degradation of fiber membrane materials

A certain amount of prepared PLA, P(3HB-co-4HB), and PPC fibers were placed in the phosphate buffered saline-containing centrifuge tube and incubated at temperature-constant (37 ℃) water bath. After different periods of time, fibers were taken for contact angle measurements.

Main outcome measures

Microscopic appearance of fibers and surface contact angle of fiber membrane.

Design, enforcement and evaluation

Authors of this study were responsible for experimental design, procedures enforcement and data evaluation.

RESULTS

Morphology of electrospun PLA, P(3HB-co-4HB), and PPC nanofibers

Microscopic morphology and structure analysis of electrospun PLA nanofibers are shown in Figure 1.

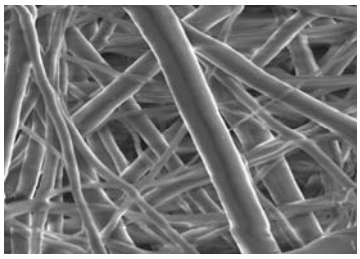

Figure 1 Scanning electron micrograph of electrospun poly (lactic acid) nanofiber membrane (× 5 000), with an average fiber diameter of approximately 800 nm (parameters: voltage 10 kV, distance from spout to receiver 10.5 cm, concentration 6 *wt*%, and feeding rate 0.8 mL/h)

Microscopic morphology and structure analysis of electrospun P(3HB-co-4HB) nanofibers are shown in Figure 2.

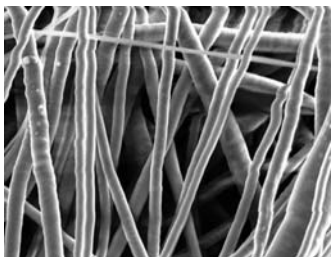

Figure 2 Scanning electron micrographs of electrospun poly(3-hydroxybutyrate-co-4-hydroxybutyrate) nanofiber membrane, with an average fiber diameter of approximately 1 μm (parameters: voltage10 kV, distance from spout to receiver 10.5 cm, concentration 6 *wt*%, and feeding rate 0.8 mL/h)

Microscopic morphology and structure analysis of electrospun PPC nanofibers are shown in Figure 3.

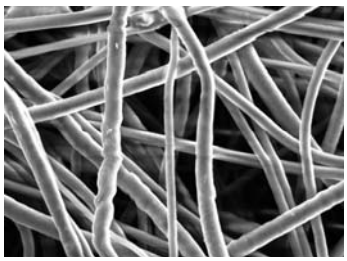

Figure 3 Scanning electron micrograph of electrospun poly(propylene carbonate) nanofiber membrane, with an average fiber diameter of approximately 1 μm (parameters: voltage 10 kV, distance from spout to receiver 10.5 cm; concentration 6 *wt*%, and feeding rate 0.8 mL/h)

Contact angle at different periods of time

Contact angle of PLA, P(3HB-co-4HB), and PPC are shown in Figure 4 and Tables 1–3.

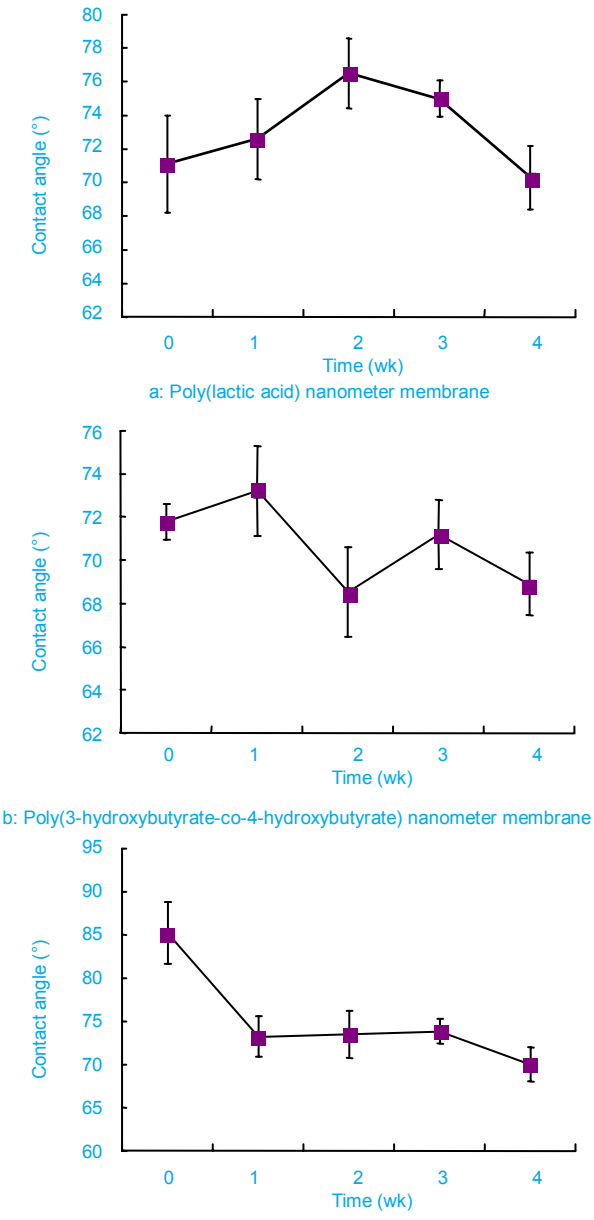

Figure 4 Changes of contact angle of poly(lactic acid) , poly(3-hydroxybutyrate-co-4-hydroxybutyrate), and poly(propylene carbonate) nanometer membranes

Table 1 Contact angle of poly(lactic acid) nanometer membrane (°)

Degradation time (wk)	Measuring point 1	Measuring point 2	Measuring point 3
0	66.3	72.8	73.2
1	74.5	70.5	71.5
2	73.5	79.3	76.2
3	73.8	74.2	75.8
4	70.8	71.5	67.3

Degradation time (wk)	Measuring point 4	Measuring point 5	Mean	Standard deviation
0	70.3	72.8	71.10	2.91
1	70.8	75.8	72.60	2.38
2	76.2	77.2	76.50	2.09
3	76.5	75.0	75.00	1.11
4	72.2	69.8	70.30	1.91

Table 2 Contact angle of poly(3-hydroxybutyrate-co-4-hydroxybutyrate) nanometer membrane (°)

Degradation time (wk)	Measuring point 1	Measuring point 2	Measuring point 3
0	72.2	71.2	72.0
1	71.2	71.3	76.2
2	71.0	66.2	67.3
3	72.8	70.3	73.0
4	70.2	69.7	69.3

Degradation time (wk)	Measuring point 4	Measuring point 5	Mean	Standard deviation
0	70.7	72.8	71.80	0.83
1	73.8	73.3	73.20	2.06
2	70.3	67.5	68.50	2.07
3	69.8	70.0	71.20	1.58
4	66.5	68.7	68.90	1.44

Table 3 Contact angle of poly(propylene carbonate) nanometer membrane (°)

Degradation time (wk)	Measuring point 1	Measuring point 2	Measuring point 3
0	69.5[a]	80.2	86.7
1	71.3	72.8	75.5
2	77.8	72.0	74.5
3	72.8	75.2	74.3
4	71.0	66.7	70.0

Degradation time (wk)	Measuring point 4	Measuring point 5	Mean	Standard deviation
0	88.5	85.5	85.20	3.57
1	75.8	70.7	73.20	2.35
2	72.0	71.0	73.50	2.75
3	71.8	74.8	73.80	1.43
4	71.5	71.0	70.00	1.94

a: data of measuring point were greatly deviated and excluded

DISCUSSION

Ideal tissue engineering scaffold materials should provide three-dimensional structure for cell retention and guide cell adhesion, proliferation, and differentiation. Research regarding this has become a hot spot.

The guiding of materials to cells is closely related to the following factors: ① Material selection: different materials produce different stimulations and effects on cells. For example, alginate with negative charges and chitosan with positive charges were found to exhibit different effects on neural cell regeneration. In addition, chemical composition, mechanical strength, degradation velocity, and degradation products of materials have different effects on cells. ② Material surface structure: because the binding of materials and cells starts at material surface, so material surface structure directly influences cell biological behaviors. The microscopic surface appearance of materials is one of the most important surface properties. The electrospinning products exhibit better affinity because they simulate the tissue microstructure of human body. ③ Surrounding environment stimulation: this includes the effects of various factors from culture environment on cells and

the effects on the whole culture environment, such as temperature, mechanical field change, additional magnetic and electric fields, and the interaction of multiple cells.

The present study prepared PLA, P(3HB-co-4HB), and PPC nanometer membranes by electrospinning, analyzed the structural morphology of fiber membranes, and investigated the surface hydrophilicity after soaking in phosphate buffered saline (pH7.4, 37 ℃), which was close to human body environment, for different periods of time. Results revealed that chemical bonds exhibited among the PLA, P(3HB-co-4HB), and PPC nanometer membrane basal bodies, nanometer particles increased fiber diameter and surface roughness, the introduction of 4HB and PPC nanometer particles inhibited the auto-catalysis of PCL and 3HB during the process of degradation, lowered the degradation velocity of PLA and 3HB, the surface hydrophilicity of three membrane materials was gradually increased within 4 weeks of in vitro degradation. In addition, among PLA, P(3HB-co-4HB), and PPC nanometer membranes, PPC nanometer membrane exhibit the worst hydrophilicity, but PPC nanometer membrane shows the largest change of surface hydrophilicity during the process of degradation and exhibits strongest surface hydrophilicity after 4 weeks of degradation. Degradation-caused hydrophilicity change is possibly due to that all three membrane materials are polyester materials and esterfunction hydrolysis initiates firstly during the process of degradation. On one hand, esterfunction hydrolysis produces hydroxy and carboxy groups, significantly increasing the number of polar functional groups, and contributing to enhanced surface hydrophilicity. On the other hand, micromolecules produced by hydrolysis better facilitate the expose of polar functional groups than non-degraded macromolecules, contributing to better surface hydrophilicity.

In conclusion, the surface properties of PLA, P(3HB-co-4HB), and PPC nanometer fiber membranes prepared by electrospinning would change continuously. Further work is needed to perform cell culture to validate the biocompatibility of scaffold materials for final selection of proper materials and structure and optimize scaffold properties for construction of tissue-engineered esophageal stent.

REFERENCES

[1] Langer R, Vacanti JP. Tissue engineering. Science. 1993;260(5110):920-926.
[2] Yao KD, Yin YJ. Biomaterials for Tissue Engineering. Beijing: Chemical Industry Press. 2003.
[3] Huang ZM, Zhang YZ, Kotaki M, et al. A review on polymer nanofibers by electrospinning and their applications in nanocomposites. Compos Sci Technol. 2003;63(15):2223-2253.
[4] Matthews JA, Wnek GE, Simpson DG, et al. Electrospinning of collagen nanofibers. Biomacromolecules. 2002;3(2):232-238.
[5] Cui W, Li X, Zhu X, et al. Investigation of drug release and matrix degradation of electrospun poly(DL-lactide) fibers with paracetanol inoculation. Biomacromolecules. 2006;7(5):1623-1629.
[6] Zeng J, Xu X, Chen X, et al. Biodegradable electrospun fibers for drug delivery. J Control Release. 2003;92(3):227-231.
[7] Zhang YZ, Wang X, Feng Y, et al. Coaxial electrospinning of (fluorescein isothiocyanate-conjugated bovine serum albumin)-encapsulated poly (epsilon-caprolactone) nanofibers for sustained release. Biomacromolecules. 2006;7(4):1049-1057.
[8] Huang ZM, He CL, Yang AC, et al. Encapsulation drugs in biodegradable ultrafine fibers through co-axial electrospinning. J Biomed Mater Res A. 2009;77(1):169-179.
[9] Diaz JE, Barrero A, Marquez M, et al. Controlled encapsulation of hydrophobic liquids in hydrophilic polymer nanofibers by co-electrospinning. Adv Funct Mater. 2009;16: 2110-2116.
[10] Qi H, Hu P, Xu J, et al. Encapsulation of drug reservoirs in fibers by emulsion electrospinning: morphology characterization and preliminary release assessment. Biomacromolecules. 2006;7(8):2327-2330.
[11] Xu X, Yang L, Xu X, et al. Ultrafine medicated fibers electrospun from W/O emulsions. J Control Release. 2005;108(1):33-42.
[12] Tan ST, Wendorff JH, Pietzonka C, et al. Biocompatible and biodegradable polymer nanofibers displaying superparamagnetic properties. Chemphyschem. 2005;6(8):1461-1465.
[13] Smith DJ, Reneker DH. Nitric Oxide-modified Linear Poly (ethylenimine) Fibers and Uses Therefore. United States: Patent Application Publication US. 2004/0131753, 2004.

电纺丝聚乳酸、聚 3 羟基丁酸酯共聚 4 羟基丁酸酯和聚碳酸亚丙酯纳米纤维的制备及表面亲水性*☆

李 楠[1]，王雪明[1]，齐宏旭[2]，翟俊山[1]，王 艳[2]，胡 平[2]，朱建华[1]([1]解放军总参谋部总医院(解放军第 309 医院)消化科，北京市 100091；[2]清华大学化工系高分子材料研究室，北京市 100084)

李 楠☆，男，1957 年生，山东省青岛市人，汉族，1996 年解放军第三军医大学毕业，博士，教授，主任医师，博士生导师，主要从事消化内镜新技术研究及临床应用。

摘要

背景：近年来聚乳酸、羟基磷灰石类复合材料支架具有良好的生物降解性和生物相容性而被广泛的研究，但是这类复合材料在增强材料界面的结合、调节材料的降解速率、改善材料的强度等方面仍不能满足理想的组织工程支架材料的要求。

目的：探讨电纺丝法制备纳米纤维的结构形态及表面亲水性。

方法：分别将聚乳酸、聚 3 羟基丁酸酯共聚 4 羟基丁酸酯和聚碳酸亚丙酯通过静电纺丝法制备纳米纤维膜，扫描电镜对纤维膜的结构形态进行分析，并观察在人体环境相近的磷酸盐缓冲溶液(37 ℃，pH 7.4)中浸泡不同时间的表面亲水性。

结果与结论：通过静电纺丝技术可以将聚乳酸、聚 3 羟基丁酸酯共聚 4 羟基丁酸酯和聚碳酸亚丙酯 3 种材料制备成微纳米纤维结构，控制制备参数可以获得不同直径的纤维，样品随着在培养液中的浸泡时间延长，总体显示出接触角比初始降低，亲水性增强。

关键词：聚乳酸；聚 3 羟基丁酸酯共聚 4 羟基丁酸酯；聚碳酸酯；电纺丝；亲水性

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利益冲突：无利益冲突。