



Cell adhesion behavior of poly(ϵ -caprolactone)/poly(L-lactic acid) nanofibers scaffold

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ABSTRACT

Biodegradable nanofibrous tubes are being investigated and developed for nerve tissue regeneration. The poly (ϵ -caprolactone) and poly (L-lactic acid) offer competitive candidacy due to higher stability by former and better biodegradability of the latter. Exploiting these characteristics of both the polymers, we present our study on generation of nanofiber tubes from pure PCL, pure PLLA and their blends (PCL/PLLA). The nanofibers were electrospun and collected on 2 cm diameter rotating collector. The samples were analyzed for cell adhesion, tensile strength, homogeneity, chemical analysis by FTIR. The results show comparatively enhanced cell adhesion in PLLA samples and blends with higher PLLA proportion. Contrary, samples with higher PCL proportion depicted better tensile strength. FTIR results demonstrated PCL and PLLA characteristic peaks in their blends. The results confirm suitability of PCL/PLLA nanofibrous tubes for nerve tissue regeneration and tissue growth.

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1. Introduction

Biodegradability of the polymers is a well desired characteristic for fabrication of nerve tissue regeneration scaffolds [1,2] offering advantage of elimination of post operative surgery necessary for removal of the conduits in case of non-biodegradable materials [3]. However, selection of a suitable polymer or polymer blend is a fundamental consideration for success of the implant. The commonly used polymers used so far for tissue regeneration applications are PCL [4,5], PLLA [6,7], PLGA [8], PLCL [9], PLLA-PDLA [10], PLGA-PCL [3], SF [11] and P(LLA-CL) [12]. Synthesis of implants composed of polymer blends offers a suitable choice to obtain tailored properties and improved performances in terms of physical and biological characteristics [13].

PCL in general offers mechanical strength to the structure, higher permeability and suitable cell adherence characteristics [14,15]. However, the PLLA on the other hand endow exceptional biodegradation [16] and has been referred for faster biodegradation

but slow permeation potential [17]. To exploit these differing characteristics of both these polymers we attempted to carry out comparative analysis of these polymers and fabricated their electrospun nanofibers in order to investigate their combined cell culture potential and mechanical stability of the nanofibers. The PCL, PLLA and their three blends were electrospun in to tubular form in order to simulate nerve conduit by collecting the nanofibers on 2 cm diameter collector. The nano-fibrous structure of the polymer conduits offers higher surface area and porous structure for cell adherence and growth [18]. The results exhibited substantial cell growth on the samples hence confirmation of suitability of the nanofiber conduits for nerve tissue regeneration.

2. Materials and methods

2.1. Materials

Poly(ϵ -caprolactone) (PCL: Mw 80,000), poly(L-Lactic Acid) (PLLA: Mw 143,000), DMF, chloroform and acetone were purchased from Sigma Aldrich, Japan. PCL solution 12% (w/w) was prepared using DMF: chloroform (1:9) while PLLA 8% (w/w) using chloroform: acetone (3:1) solvents. Five formulations such as pure PCL and PLLA, PCL/PLLA 1:1, PCL/PLLA 1:2 and PCL/PLLA 2:1 were prepared and stirred for 24 h at 50 °C prior to electrospinning.

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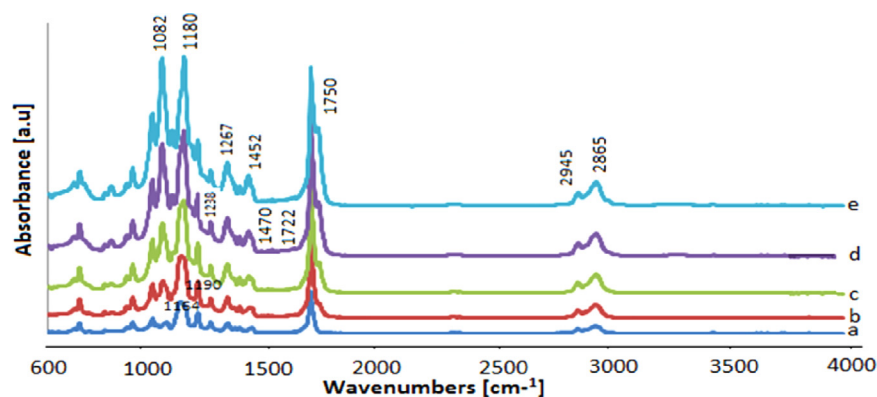


Fig. 1. FTIR spectrum of (a) PCL (b) PCL/PLLA 2:1 (c) PCL/PLLA 1:1 (d) PCL/PLLA 1:2 (e) PLLA.

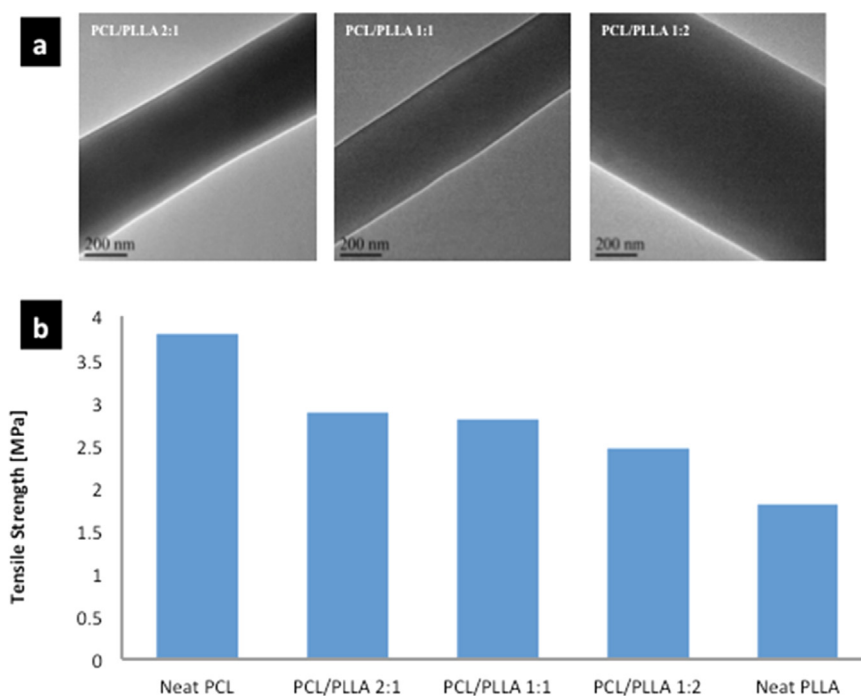


Fig. 2. TEM images (a) Tensile strength of the nanofibers (b).

2.2. Method

Electrospinning spinning apparatus (Har-100*12, Matsusada Co., Tokyo, Japan) was used for nanofiber formation. The nanofibers were collected on 2 cm rotating collector. Needle tip to collector distance was maintained 12 cm and 12 kV voltage was supplied.

2.3. Characterization

All the samples were characterized for morphology by scanning electron microscope (SEM, S-3000N by Hitachi, Japan) and transmission electron microscopy (JEOL model 2010 Fas TEM). Chemical analysis of the samples was conducted using Fourier transform infrared spectroscopy (FTIR, IR Prestige-21, Shimadzu, Japan). Testing of the mechanical property of the nonwoven nanofibers was performed according to ASTM D-638 using a universal testing machine (Tensilon RTC1250A; A&D Company Ltd, Japan). The crosshead speed was set at 5.0 mm/min.

2.4. Cell culture

The murine fibroblast L929 cells (2104 cells/well) were utilized for L929 in-vitro growth test. Fibrous RSF and RSF/TMOS electrospun-fibers were treated with CH₃OH (50%) at room temperature for 60 min followed by vacuum drying for 24 h prior to utilization. Tissue culture dishes (TCDs) were incubated at 37 °C (5% CO₂ in atmosphere) using Eagle's MEM supplemented by FBS (5%), penicillin/streptomycin (105U and 0.1 g L1MEM respectively) and 2 mM L-glutamine.

3. Results and discussion

3.1. FTIR analysis

The FTIR results of PCL, PLLA and PCL/PLLA blends are described in Fig. 1. The PCL spectrum (Fig. 1a) depicts characteristic peaks of carbonyl stretching (CO) at 1722 cm⁻¹, COC stretching (asymm.) at 1238 cm⁻¹, COC stretching (symm.) at 1164 cm⁻¹, CH₂ stretching

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