Materials Science and Engineering C 43 (2014) 102–108

Contents lists available at ScienceDirect

Materials Science and Engineering C

journal homepage: www.elsevier.com/locate/msec

Activated carbon fibers/poly(lactic-co-glycolic) acid composite scaffolds: Preparation and characterizations

Yanni Shi [a,b], Hao Han [b,c], Haiyu Quan [b], Yongju Zang [b], Ning Wang [b], Guizhi Ren [b], Melcolm Xing [d], Qilin Wu [a,b,*]

[a] State Key Laboratory for Modification of Chemical Fibers and Polymer Materials, Donghua University, Shanghai 201620, PR China
[b] College of Materials Science and Engineering, Donghua University, Shanghai 201620, PR China
[c] Bayer Material Science (China) Co., Ltd, Shanghai 200120, PR China
[d] Department of Mechanical Engineering, Faculty of Engineering and Department of Biochemistry & Genetics, Faculty of Medicine P.I., Manitoba Institute of Child Health, University of Manitoba, Winnipeg, Manitoba, Canada

ARTICLE INFO

Article history:
Received 10 March 2014
Received in revised form 22 May 2014
Accepted 1 July 2014
Available online 9 July 2014

Keywords:
Activated carbon fibers
Polymer composite scaffold
Cell adhesion
Bioactivity
Histocompatibility

ABSTRACT

The present work is a first trial to introduce activated carbon fibers (ACF) with high adsorption capacity into poly(lactic-co-glycolic) acid (PLGA), resulting in a novel kind of scaffolds for tissue engineering applications. ACF, prepared via high-temperature processing of carbon fibers, are considered to possess bioactivity and biocompatibility. The ACF/PLGA composite scaffolds are prepared by solvent casting/particulate leaching method. Increments in both pore quantity and quality over the surface of ACF as well as a robust combination between ACF and PLGA matrix are observed via scanning electron microscopy (SEM). The high adsorption capacity of ACF is confirmed by methylene blue solution absorbency test. The surfaces of ACF are affiliated with many hydrophilic groups and characterized by Fourier transform infrared spectroscopy. Furthermore, the SEM images show that cells possess a favorable spreading morphology on the ACF/PLGA scaffolds. Besides, vivo experiments are also carried out to evaluate the histocompatibility of the composite scaffolds. The results show that ACF have the potential to become one of the most promising materials in biological fields.

1. Introduction

Recently, synthetic polymers have become an intense interest because of their tailored structural design for tissue engineering [1]. Poly(lactide-co-glycolide) acid (PLGA) is widely utilized due to good biocompatibility and biodegradation [2,3]. PLGA presents high hydrophilic but weak cell affinity. However, the resulting lactic acid to glycolic acid from degradation of PLGA has been a clinical concern in its lasting inflammatory response [4,5]. Quite a few strategies have been adopted to improve PLGA performance including composite with other bioactive materials and incorporating nano-scale features to increase surface/volume ratio and to promote cell attachment and osteogenic differentiation [6–10].

Carbon fibers (CF) and carbon nanotubes are already proved toxic-free and biocompatible and utilized as scaffold mechanical support, and drug vehicles and biosensors in biomedical fields [11–18]. Magdalena et al. found that the addition of CF was friendly to cells or tissues by comparing PLGA co-polymer containing CF and hydroxyapatite [19]. CF implants were also used for repairing osteochondral defects created on the articular surface of the patella of rabbits, and it was found that the defect sites were covered transparent cartilage tissue twelve months later [20]. As support additives, CF are considered to guide the proliferation of fibrous tissue following the orientation of the filaments of CF, and thus promote tissue regeneration process [21]. Besides, scanning electron microscopy (SEM) images revealed excellent integration between CF and polymer matrix, which was benefit for forming a strong mechanical interlock between cells and composites [22].

The feasibility and advantages of CF applied in tissue engineering applications are well-acknowledged by many researchers, but mostly CF are used as reinforced fillers and the lack of bioactive recognition spots might restrict extensive use of CF in medical applications [23,24]. It has been demonstrated that bioactivity of CF can be improved by CF surface modification and doping other bioactive additives into CF precursors [25]. Activated carbon fibers (ACF) are obtained via CF processing in water and nitrogen streams at high temperature. Due to richly-distributed pore structures and a large specific surface area (S.S.A.), ACF possess high adsorption capacity and are expected to allow higher possibility and larger space for nutrient storage and exchange which are good for cell adhesion and proliferation. The specific performance and advantages of ACF for tissue engineering applications are rarely mentioned in previous studies. The prominent adsorption capacity and biocompatibility of ACF are taken into discussion in the paper.

The purpose of this study is to consider the use of ACF incorporation with polymer for tissue defect regeneration. We attempt to investigate

* Corresponding author at: State Key Laboratory for Modification of Chemical Fibers and Polymer Materials, Donghua University, Shanghai 201620, PR China. Tel.: +86 13917306197; fax: +86 21 6779 2855.
E-mail address: wql@dhu.edu.cn (Q. Wu).

http://dx.doi.org/10.1016/j.msec.2014.07.007
0928-4931/

Table 1
PLGA and ACF/PLGA composite scaffolds applied in this work.

Specimens	PLGA (g)	NaCl (g)	ACF (wt %)	Porosity (%)
Sample 1	1.82 ± 0.02	7.26 ± 0.01	0.00	75.00 ± 0.02
Sample 2	**1.82 ± 0.01**	**7.26 ± 0.01**	**2.75**	**75.12 ± 0.01**
Sample 3	1.82 ± 0.02	7.26 ± 0.01	8.26	79.82 ± 0.02

Bold words and numbers indicates that we applied these parameters in the following experiments.

the structure and properties of ACF/PLGA composite scaffolds for tissue engineering applications. In addition, the effects of ACF content on porosity and pore size of composite scaffolds are also discussed to provide experimental data for the use of ACF/PLGA composites.

2. Experimental

2.1. Materials

ACF with certain strength were prepared in our laboratory. Via high temperature activation treatments, ACF possess richly-distributed pore structures and a large S.S.A. Air plasma treatments and the chemical regents such as aquae hydrogenii dioxidi and ammonium hydroxide were applied afterwards to improve the hydrophilicity of ACF and enhance the integration between ACF and PLGA matrix.

PLGA (lactic acid/glycolic acid = 85:15; M_w = 69850; inherent viscosity = 1.1941 g/cm^3) was supplied by Jinan Daigang Biomaterials Co., Ltd, China. Dichloromethane (CH_2Cl_2) was used as an organic solvent for SC/PL method. All the chemical reagents used in the experiments were of analytical purity and offered by Sinopharm Co., Ltd.

2.2. Preparation

In the present work, ACF/PLGA composite scaffolds were prepared by SC/PL method with sodium chloride (NaCl) particles as the leachable component. SC/PL method, which requires neither complicated procedures nor expensive facilities, is considered to be a facile approach for the development of a highly interconnected pore structure [26]. As for scaffolds prepared by SC/PL method, the pore size and porosity can be controlled by tuning sieved salt particle size and the initial salt weight fraction, respectively [27]. Composite solution of PLGA/CH_2Cl_2 was casted into a no-covering square box, and a certain amount of NaCl particles were added aiming to fabricate pores. The masses of ACF and NaCl particles added to composite solution were adjusted so as to obtain varying porosity as depicted in Table 1. The mixture was airproofed immediately and treated by ultra-sonication. Well-dispersed composite solution was loaded in the steel-made squared mold (side length = 10 mm). Followed by solvent evaporation, the polymer disk with entrapped salt particles was removed out of the mold, and then the salt was dissolved in the deionized water for at least 48 h to remove the salt. The schematic of whole process is shown in Fig. 1.

Sample 2 was applied in the following biological tests. The scaffolds were cut into disks of 14 mm in diameter and 2 mm in height and sterilized by exposing to gamma radiation (25 kGy). All the disks were rinsed with phosphate buffer solution (PBS, 0.01 mol/L, pH 7.2, supplied by Sigma, USA) three times, and pre-wet in the DMEM culture medium (Dulbecco's modified Eagle's medium, Hangzhou Gino Biomedical Technology Co., Ltd, China) overnight.

2.3. Surface texture observation of ACF and scaffolds

The morphologies of ACF as well as ACF/PLGA composite scaffolds were characterized by scanning electron microscopy (SEM, JSM-5600LV, JEOL Co., Ltd, Japan). All samples were fixed on a cupreous stub and sparked with a gold layer with the thickness of 15 nm in advance to produce a conductive surface, and their morphologies were observed at an accelerating voltage of 10 kV.

2.4. FT-IR characterization

The Fourier transform infrared spectroscopy (FT-IR, NEXUS-670, Nicolet Co., Ltd, USA) was used to identify the infrared absorption peaks of such hydrophilic groups as $-NH_3$ and $-COOH$ to confirm their presences on the surface of ACF after air plasma and chemical treatments.

2.5. Pore structure analysis of ACF

The automated surface and pore size analyzer (Autosorb-1 MP, Quantachrome, USA) was used to analyze porous texture of ACF at various activation temperatures. The S.S.A. and pore volume were calculated by the Brunauer-Emmett-Teller (BET) method and Barrett-Joyner-Halenda (BJH) model, respectively, based on the adsorption/desorption isotherms of nitrogen at 77 K.

2.6. Adsorption capacity of ACF

The adsorption capacity was measured by methylene blue solution (MBS) absorbency test at room temperature. Firstly 0.10 ± 0.01 g ACF was soaked in 1000 ml MBS (initial concentration = 400 mg/l), and then 2 ml mixture was fetched out at regular intervals to test the value of adsorption using an ultraviolet spectrophotometer (Lanbda A35, Unico. Co., Ltd, USA).

2.7. Cell culture

Mouse fibroblast cells (L929s) were offered by the cell bank of the Chinese Academy of Science. L929s were cultured in the medium comprising 89% DMEM high glucose, 10% fetal bovine serum, 1% penicillin and streptomycin at 37 °C with 5% CO_2 in a humid incubator. Unattached cells were removed on the following day and half of the culture medium was replaced by a fresh aliquot every other day. Cell colonies

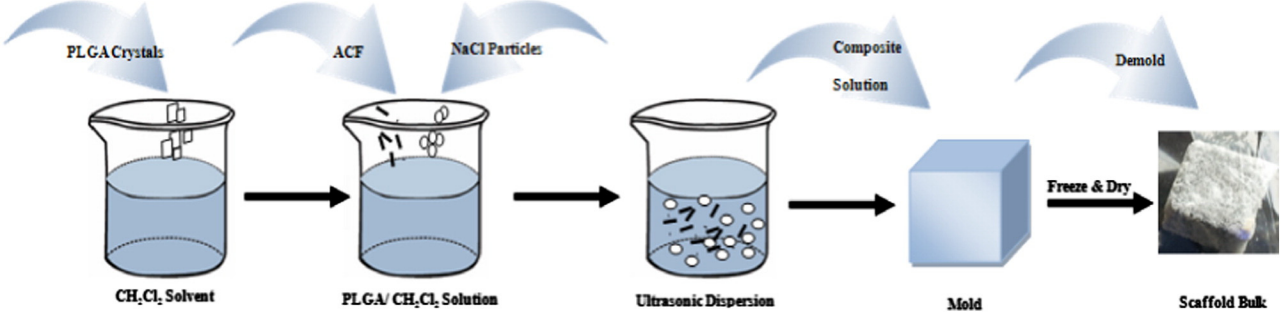

Fig. 1. The preparation process of ACF/PLGA composite scaffolds by SC/PL method.