Biomaterials xxx (2013) 1–10

Contents lists available at SciVerse ScienceDirect

Biomaterials

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

Shell-core bi-layered scaffolds for engineering of vascularized osteon-like structures

Xuening Chen [a], Asli Ergun [b], Halil Gevgilili [b], Seher Ozkan [b], Dilhan M. Kalyon [a,b], Hongjun Wang [a,*]

[a] Department of Chemistry, Chemical Biology and Biomedical Engineering, Stevens Institute of Technology, Hoboken, NJ 07030, USA
[b] Department of Chemical Engineering and Materials Science, Stevens Institute of Technology, Hoboken, NJ 07030, USA

ARTICLE INFO

Article history:
Received 12 June 2013
Accepted 9 July 2013
Available online xxx

Keywords:
Osteon
Electrospinning
Twin screw extrusion
Vascularization
Cortical bone

ABSTRACT

Bottom-up assembly of osteon-like structures into large tissue constructs represents a promising and practical strategy toward the formation of hierarchical cortical bone. Here, a unique two-step approach, *i.e.*, the combination of electrospinning and twin screw extrusion (TSE) techniques was used to fabricate a microfilament/nanofiber shell–core scaffold that could precisely control the spatial distribution of different types of cells to form vascularized osteon-like structures. The scaffold contained a helical outer shell consisting of porous microfilament coils of polycaprolactone (PCL) and biphasic calcium phosphates (BCP) that wound around a hollow electrospun PCL nanofibrous tube (the core). The porous helical shell supported the formation of bone-like tissues, while the luminal surface of nanofibrous core enabled endothelialization to mimic the function of Haversian canal. Culture of mouse pre-osteoblasts (POBs, MC 3T3-E1) onto the coil shells revealed that coils with pitch sizes greater than 135 μm, in the presence of BCP, favored the proliferation and osteogenic differentiation of POBs. The luminal surface of PCL nanofibrous core supported the adhesion and spreading of mouse endothelial cells (ECs, MS-1) to form a continuous endothelial lining with the function similar to blood vessels. Taken together, the shell–core bi-layered scaffolds with porous, coil-like shell and nanofibrous tubular cores represent a new scaffolding technology base for the creation of osteon analogs.

1. Introduction

Bone tissue engineering is promising to provide bone substitutes that can potentially eliminate donor site scarcity associated with autografts, and immune rejection and pathogen transfer associated with allografts [1,2]. However, very limited progress has been made to engineer large bone grafts [3], especially those cortical bone segments with complex hierarchical structures, which are difficult to recapture *via* conventional tissue engineering. Considering the fact that cortical bone consists of compacted repeating units called osteons [4,5], one possible strategy to recapitulate the hierarchical complexity of cortical bone is to generate osteon-like units that can be assembled and fused into large and integral constructs (Fig. 1).

Effective creation of "osteons" is a critical step toward the generation of "cortical bone". Initial attempts involving the fabrication of poly-L-lactide (PLLA) or gelatin/PLLA nanofibrous rods *via* electrospinning and then seeding with osteoblasts [6] have been made. However, the tightly packed nanofiber layers of the resulting rods and the lack of vascular systems prevented cell infiltration and led to rapid nutrition depletion and hypoxia [7,8]. These initial investigations have pointed out the importance in more closely mimicking the key functions of osteons. In each osteon, the concentric bone-cell-containing layers (called lamellae) surround a narrow central channel (called Haversian canal), which contains small blood vessel to facilitate the exchange of nutrients and metabolic waste [9]. The unique structure and cellular organization of osteon give rise to its functionality. In order to effectively regenerate osteon-like structures with proper functions, we have developed a shell–core bi-layered scaffold that contains a tubular core for creating functional vessel-like structure to mimic the Haversian canal, and surrounding porous shell to support bone tissue formation (Fig. 1a).

* Corresponding author. Department of Chemistry, Chemical Biology and Biomedical Engineering, Stevens Institute of Technology, McLean Building Room 416, Castle Point on Hudson, Hoboken, NJ 07030, USA. Tel.: +1 201 216 5556; fax: +1 201 216 8240.
E-mail address: Hongjun.Wang@stevens.edu (H. Wang).

0142-9612/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.biomaterials.2013.07.035

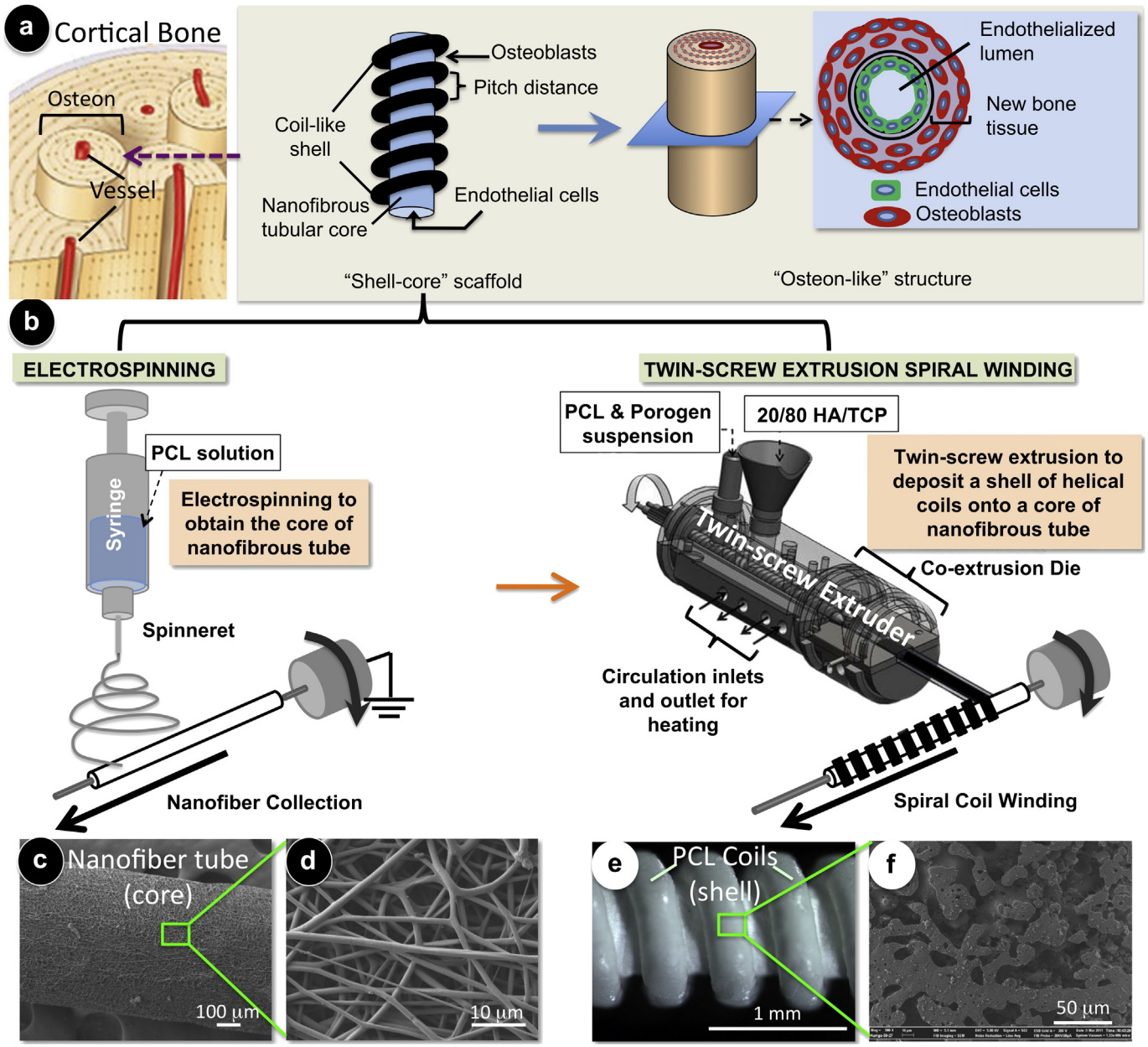

Fig. 1. (a) Schematic illustration of the approach to form "osteon-like" unit structures by respectively culturing osteoblasts onto the inter-coil space (the "shell") and endothelial cells inside the luminal surface (the "core") of the novel nanofiber/microfilament shell–core scaffolds. (b) The combination of electrospinning and twin screw extrusion (TSE) for fabricating cylindrical nanofiber/microfilament shell–core scaffolds, *i.e.*, electrospinning for preparing the hollow nanofiber tube (core) and TSE for depositing porous coils (shell) onto tubes. (c–d) SEM images of the hollow electrospun PCL nanofiber tube (core). (e) Stereomicroscopic image of the PCL coils (shell) with porous surface as visualized by SEM (f).

Previous studies have shown that electrospun nanofibrous meshes support the adhesion and spreading of endothelial cells and allow them to form continuous layers with potential utility as vascular grafts [10,11]. Thus, electrospun nanofibrous tubes would be ideal as cores to generate the vessel-like structures.

Twin screw extrusion (TSE) has shown great promise in fabrication of tissue-engineering scaffolds due to its versatile capabilities of carrying out multiple processing steps, including conveying solids, melting polymers, mixing, pressurization and devolatilization [12–23]. The ability to manipulate the feeding rates of various ingredients (*e.g.*, polymers, porogens, particles and bioactive molecules like growth factors) into the twin-screw extruder enables the fabrication of graded scaffolds with systematic changes in porosity, pore sizes, bioactive concentrations and mechanical properties [15,17,20]. Furthermore, in order to fabricate versatile structures it can be readily integrated with other processing techniques such as electrospinning [12,13], co-extrusion [16–18], and spiral winding (*i.e.*, TSESW process) to fabricate annular scaffolds [19,20]. We recognized that during the TSESW, concomitant translation and rotation of the mandrel with electrospun nanofiber tube would generate a helical trajectory to form coil-like shell structures around the tubular core and consequently yield a shell–core bi-layered scaffold (Fig. 1b).

Various materials can be used to fabricate coils, but preference would be given to those that support bone formation. Polycaprolactone (PCL) has good biocompatibility and exhibits a relatively slow biodegradation rate that can maintain mechanical stability long enough for new bone formation [24,25]. Biphasic

Please cite this article in press as: Chen X, et al., Shell-core bi-layered scaffolds for engineering of vascularized osteon-like structures, Biomaterials (2013), http://dx.doi.org/10.1016/j.biomaterials.2013.07.035