Acta Biomaterialia xxx (2013) xxx–xxx

Contents lists available at SciVerse ScienceDirect

Acta Biomaterialia

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

Polymeric topology and composition constrained polyether–polyester micelles for directional antitumor drug delivery

Di Li [a,1], Hai Sun [a,1], Jianxun Ding [a,*], Zhaohui Tang [a,*], Ying Zhang [a], Weiguo Xu [a], Xiuli Zhuang [a,b], Xuesi Chen [a,b]

[a] Key Laboratory of Polymer Ecomaterials, Changchun Institute of Applied Chemistry, Chinese Academy of Sciences, Changchun 130022, People's Republic of China
[b] Changchun SinoBiomaterials Co. Ltd, Changchun 130103, People's Republic of China

ARTICLE INFO

Article history:
Received 21 March 2013
Received in revised form 22 June 2013
Accepted 24 June 2013
Available online xxxx

Keywords:
Composition
Controlled release
Malignancy therapy
Micelle
Topology

ABSTRACT

Amphiphilic linear and dumbbell-shaped poly(ethylene glycol)–poly(lactide-*co*-glycolide) (PEG–PLGA) copolymers were simply synthesized by the ring-opening polymerization of lactide and glycolide using PEG or tetrahydroxyl-functionalized PEG as the macroinitiator and stannous octoate as the catalyst. The copolymers spontaneously self-assembled into spherical micelles in phosphate-buffered saline at pH 7.4. The self-assembly behavior was dependent on both the polymeric topology and composition. Doxorubicin (DOX), an anthracycline antitumor drug, was loaded into micelles through nanoprecipitation. The in vitro release behavior could be adjusted by regulating the topology or composition of the copolymer, or the pH of the release medium. The effective intracellular DOX release from DOX-loaded micelles was confirmed by confocal laser scanning microscopy and flow cytometry in vitro. DOX-loaded micelles displayed great cellular proliferation inhibition efficacies after incubation for 24, 48 or 72 h. The hemolysis ratio of DOX was significantly reduced by the presence of copolymer. These properties indicated that the micelles derived from linear or dumbbell-shaped copolymers were promising candidates as smart antitumor drug carriers for malignancy therapy.

1. Introduction

Malignancy (i.e. cancer) is one of the most serious worldwide diseases, threatening human health and longevity [1,2]. In spite of the vigorous exploitations of various antitumor drugs, their clinical efficacy is unsatisfactory due to their life-threatening side effects, such as leukemia and cardiotoxicity [3]. To reduce the severe side effects, many kinds of nanocarriers, such as micelles [4–7], vesicles [8,9] and nanogels [2,10], have been developed as vehicles to transport antitumor drugs.

It is well known that the amphiphilic copolymers can self-assemble into various nanoscale aggregations, such as micelles [11–15] and vesicles [16–18], in the aqueous environment dominated by the topological structures, proportions, compositions and physicochemical properties of both hydrophilic and hydrophobic moieties. For intravenous drug delivery applications, the hydrophilic segments of amphiphilic copolymers are composed of zwitterionic materials or polyethylene glycol (PEG), which can resist nonspecific protein adsorptions (i.e. nonfouling properties) and prolong the circulation times of nanoparticles in the complex in vivo circumstances [19,20]. Aliphatic polyesters, such as poly(ε-caprolactone) (PCL) [21], polylactide (PLA) [22] and poly(lactide-*co*-glycolide) (PLGA) [23], are the most commonly chosen hydrophobic moieties that work as the sustained release reservoirs of bioactive agents benefiting from their good biocompatibilities and biodegradabilities. Of all the aforementioned nanovehicles, micelles have emerged as one of the most promising nanocarriers for various antitumor drugs. The micelles are usually associated with several merits as polymeric drug carriers, such as improved drug solubility in water, prolonged circulation time, enhanced accumulation in tumor sites, decreased side effects, and elevated drug bioavailability and efficacy [24–26].

So far, the major research works have been focused on the micelles based on amphiphilic linear di/triblock copolymers in the realm of drug delivery [27,28]. As a typical example, the micelles from diblock copolymers of PEG and PLA for delivery of paclitaxel have been approved in Korea (Genexol®-PM) for the treatments of ovarian and metastatic breast cancers, and are in phase IV clinical trials in the USA as a safer alternative to Cremophor® EL and Taxol® [29,30]. Prostate-specific membrane antigen-targeted PEG–PLA and PEG–PLGA mixed micelles containing docetaxel for the treatment of patients with solid tumors are in ongoing phase II clinical trials [31]. However, the relatively poor stabilities and low drug loading capabilities of micelles based on linearcopolymers affect

* Corresponding authors. Tel./fax: +86 431 85262110 (J. Ding), +86 431 85262116 (Z. Tang).
E-mail addresses: jxding@ciac.ac.cn (J. Ding), ztang@ciac.ac.cn (Z. Tang).
[1] These authors contributed equally to this work.

1742-7061/$ - see front matter © 2013 Acta Materialia Inc. Published by Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.actbio.2013.06.041

their wide application as drug carriers [32]. Amphiphilic miktoarm star-shaped copolymers, composing of three or more hydrophilic or hydrophobic arms linked to the same junction point, have attracted much attention because the diverse topologies may improve the properties of micelles [21,22]. Although many studies on micelles originating from linear or star-shaped copolymers have been undertaken [15,21,33], systematic comparisons of the properties of micellar drug carriers made from copolymers with different topologies have rarely been reported [22].

In this study, amphiphilic linear and dumbbell-shaped copolymers composed of hydrophilic PEG and hydrophobic PLGA were efficiently synthesized by the ring-opening polymerization (ROP) of lactide (LA) and glycolide (GA) with PEG or tetrahydroxyl-functionalized PEG ($(OH)_2$–PEG–$(OH)_2$) as the macroinitiator and stannous octoate ($Sn(Oct)_2$) as the catalyst [21,22]. The obtained copolymers were employed as novel polymeric surfactants, and spontaneously self-assembled into micelles in phosphate-buffered saline (PBS) at pH 7.4. Doxorubicin (DOX), an anthracycline antitumor drug, was loaded into the cores of micelles with tunable drug loading efficiency (DLE) associated with the topologies and compositions of copolymers [15,22]. In vitro DOX release from DOX-loaded micelles in PBS was revealed to beaccelerated with the linear polymeric topology, the decrease of PLGA content, or in tumor tissular or intracellular acidic condition, which indicated their potential usages as the smart tissular and intracellular targeting drug carriers [34]. The cytocompatibilities and hemocompatibilities of copolymers, and the cellular proliferation inhibitions of DOX-loaded micelles were also revealed to be desirable. These properties indicated that the amphiphilic linear or dumbbell-shaped copolymers were promising vehicle matrices in antitumor drug delivery.

2. Materials and methods

2.1. Materials

Poly(ethylene glycol) (PEG_{90}, M_n = 4000) was purchased from Sigma–Aldrich (Steinheim, Germany) without further purification. Dowex 50 W-X2 ion exchange resins were obtained from Sigma–Aldrich (Steinheim, Germany) and used after methanol rinse. 2,2-Dimethoxypropane (DMP; Sinopharm Chemical Reagent Co. Ltd, Shanghai, China), 2,2-bis(hydroxymethyl) propionic acid (BHPA; Sinopharm Chemical Reagent Co. Ltd, Shanghai, China), 4-dimethylamiopryidine (DMAP; GL, Biochem Co. Ltd, Shanghai, China) and $Sn(Oct)_2$ (>95%, Sigma–Aldrich, Steinheim, Germany) were used as obtained. Acetonide-2,2-bis(hydroxymethyl) propionic anhydride (ABHPA) was prepared from DMP and BHPA according to the literature procedure [35]. LA and GA were recrystallized with ethyl acetate before use. Doxorubicin hydrochloride (DOX·HCl) was purchased from Zhejiang Hisun Pharmaceutical Co. Ltd (Zhejiang, China) and used as obtained. Toluene was stored over calcium hydride and purified by vacuum distillation. All the other reagents and solvents were purchased from Sinopharm Chemical Reagent Co. Ltd (Shanghai, China) and used without further purification.

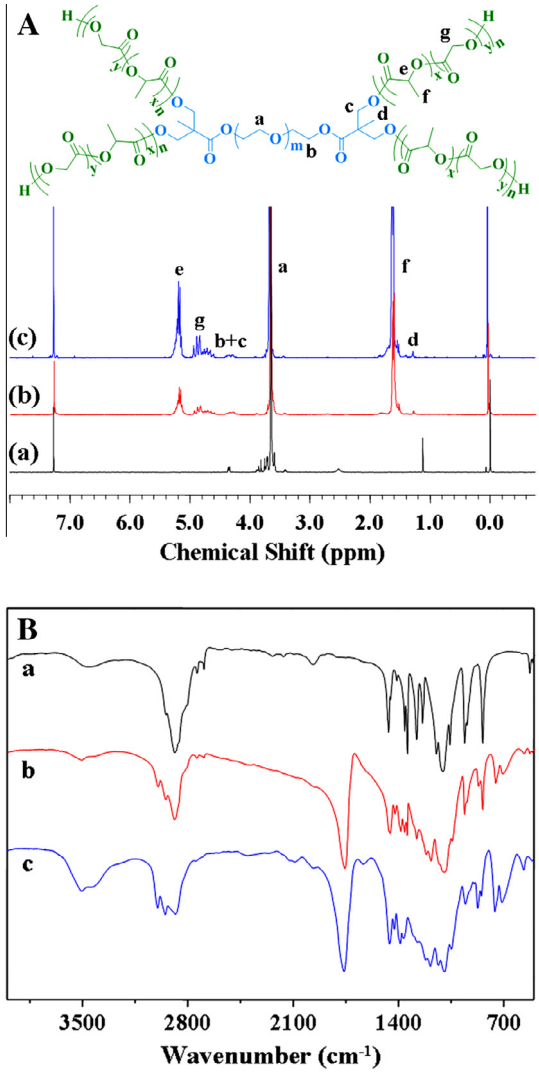
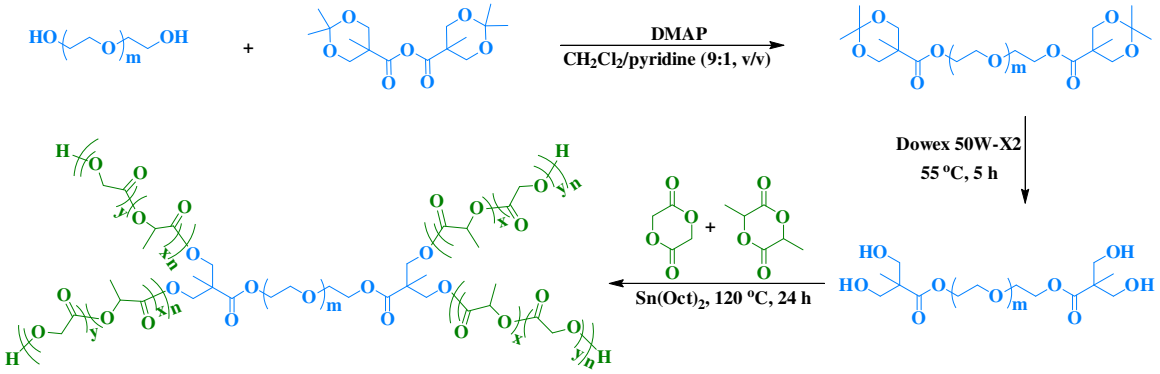


Fig. 1. ^1H NMR (A) and FTIR spectra (B) of $(OH)_2$–PEG_{90}–$(OH)_2$ (a), dsP1 (b) and dsP2 (c).



Scheme 1. Synthesis pathway for dumbbell-shaped PEG–PLGA copolymer.

Please cite this article in press as: Li D et al. Polymeric topology and composition constrained polyether–polyester micelles for directional antitumor drug delivery. Acta Biomater (2013), http://dx.doi.org/10.1016/j.actbio.2013.06.041