Carbohydrate Polymers 96 (2013) 156–162

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

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

Preparation, characterization and in vitro release of microparticles based on dextran–rosuvastatin conjugate

Chaoyu Zhou, Wenxia Gao*, Guangmin Lu, Jinchang Ding, Xiuming Wu, Xiaobo Huang, Jiuxi Chen, Miaochang Liu, Jun Jiang, Huayue Wu*

College of Chemistry and Materials Engineering, Wenzhou University, Wenzhou 325027, China

ARTICLE INFO

Article history:
Received 11 March 2013
Accepted 29 March 2013
Available online 6 April 2013

Keywords:
Rosuvastatin
Dextran–rosuvastatin conjugate
Microparticles
Drug release

ABSTRACT

A novel conjugate for rosuvastatin has been prepared by a coupling reaction between rosuvastatin and dextran. The dextran–rosuvastatin conjugates (DRC) were characterized by FT-IR, NMR, and XRD. And the resulting DRC self-assembled into microparticles by controlled slow exchange of solvents applying dialysis. The size and morphology of DRC microparticles could be controlled through the solvents and pH selection. Particles with different shape and size of microspheres, micro-blocks, micro-flakes or three-dimensional network structure were obtained by adjusting the pH from 9.0 to 13.0. Multiple morphologies of nano- or microparticles were collected in different organic solvents. In vitro release studies suggested that these DRC microparticles had a sustained release manner and the release behavior could be controlled by adjusting their morphology.

1. Introduction

Rosuvastatin is one kind of cholesterol-lowering statin drugs developed by Astrazeneca, used for high cholesterol, blood lipid metabolic disorder and pure high triglyceride blood disease treatment. It is an efficient prevention of coronary heart disease of statin drugs, approved by FDA in 2003. Rosuvastatin has achieved satisfactory results in the efficacy and safety aspects, known as "super statin" (Ballantyne, Miller, & Chitra, 2004; Kostapanos et al., 2008).

Although current statin drugs have made a great contribution to lower low-density lipoprotein (LDL) and reduce cardiovascular disease, the oral bioavailability of rosuvastatin is only about 20% (Martin, Warwick, Dane, Brindley, & Short, 2003; Martin, Warwick, Dane, Hill, et al., 2003). There is still a demand for even higher efficiency and even less side effects (Sarver et al., 2008). Therefore, finding a new drug released system that has outstanding drug absorption, distribution, and elimination is necessary (Zhang, Strong, Qiu, Lesko, & Huang, 2005).

In order to enrich our knowledge of new drug released system of rosuvastatin, polymer conjugation of rosuvastatin was synthesized and characterized. Conjugation of therapeutic compounds to polymeric carriers can result in improved bioavailability, prolonged circulation half-life, increased concentration at the site of action and decreased non-specific toxicity (Sadekar & Ghandehari, 2012; Veronese & Morpurgo, 1999). When these polymer conjugates are made to be nano- or microparticles, which will further optimize drug therapy because of their potential for site-specific drug delivery, enhancement of drug solubility, improvement of the circulation life time and the therapeutic index, and reduction of side-effects (Hornig, Bunjes, & Heinze, 2009).

Dextran was chosen in this paper as polymer backbone which has been used clinically for plasma volume expansion, peripheral flow promotion, antithrombolytic agents, and as excipient in drug delivery systems (Liebert, Hornig, Hesse, & Heinze, 2005). In particular, dextran has been widely used as potential macromolecular carriers for polymer-drug conjugates in combination with bioactive molecules to increase the bioavailability and the longevity of therapeutic agents in the circulation, such as enzymes, proteins, peptides, anti-cancer agents, anti-inflammatory drugs, and antibiotics (Mehvar, 2000; Shahnaz, Perera, Sakloetsakun, Rahmat, & Bernkop-Schnürch, 2010; Tang, Dou, & Sun, 2006).

In this paper, dextran–rosuvastatin conjugates were synthesized to self-assemble into microparticles by controlled slow exchange of organic solvent against the polymer solution applying dialysis. The influence of organic solvents and pH on the morphology and size of DRC microparticles was also investigated in this paper. In vitro drug release of these DRC microparticles were examined.

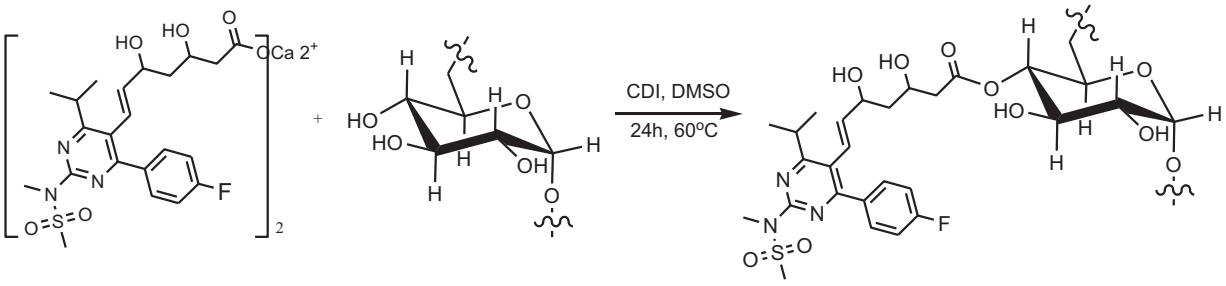
2. Experimental part

2.1. Materials

Dextran (Mw, 33,666 $g\,mol^{-1}$) and *N*,*N*-carbonyldiimidazole (CDI) were purchased from Shanghai Huamao Pharmaceutical Co.,

* Corresponding authors. Tel.: +86 577 88368280; fax: +86 577 88368280.
E-mail addresses: wenxiag@wzu.edu.cn (W. Gao), huayuewu@wzu.edu.cn (H. Wu).

0144-8617/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.carbpol.2013.03.094



Scheme 1. Synthesis of dextran–rosuvastatin conjugate (DRC).

Ltd. Rosuvastatin was obtained from Aifeimu Chemical Co., Ltd. All solvents and reagents were commercially available and analytical-reagent-grade. They were directly used without further treatment. Dialysis bag was obtained from Xinhui Zeao Science and Technology Co., Ltd.

2.2. Measurement

NMR spectra were acquired using a 500-Bruker spectrometer and reported as parts per million (ppm) from the internal standard TMS. Powder X-ray diffraction measurement (XRD) was performed on a Bruker D8 Advance diffractometer using pressed pellets as samples with Cu Kα radiation (λ = 1.5418 Å) at a voltage of 40 kV and current of 200 mA. FTIR spectra were recorded on a Bruker EQUINOX 55 spectrometer with the KBr-technique. Scanning electronic microscopy (SEM) images were taken on a Nova NanoSEM 200 scanning electron microscope.

2.3. Synthesis of dextran–rosuvastatin conjugate (DRC)

The synthesis of dextran–rosuvastatin conjugate was carried out by adding 0.31 mmol *N*,*N*-carbonyldiimidazole (CDI) and 0.31 mmol rosuvastatin calcium to a solution of 0.62 mmol dextran in 6.0 mL DMSO, as shown in Scheme 1. The mixture was allowed to react at 60 °C under stirring for 24 h and then precipitated in 100 mL isopropanol, washed twice with ethanol and dried to get the product.

2.4. Microparticles preparation of dextran–rosuvastatin conjugate (DRC)

Preparation of microparticles was carried out by a dialysis process. DRC (40 mg) was dissolved in 10 mL organic solvent and then dialyzed against 200 mL anhydrous ethanol with different pH value for two days. Dimethyl sulfoxide (DMSO), *N*,*N*-dimethylformamide (DMF) and *N*,*N*-dimethylacetamide (DMAC) were selected as organic solvents. For SEM images, the dialyzate was centrifugated and placed on a mica surface. The system was lyophilized for 6 h and sputtered with gold.

2.5. In vitro release studies

In vitro release studies were performed in triplicate according to the Ch.P 2010 (paddle method). Formulations of the dextran–rosuvastatin conjugate and control were added to phosphate buffer (pH 7.4, 900 mL). The stirring speed was 50 rpm, and the temperature was maintained at 37 ± 0.5 °C. At selected time intervals for a period of 25 h, 5 mL solution was withdrawn from the dissolution medium through a 0.22 μm membrane filter and assayed spectrophotometrically at 242 nm. Formulations contained 10 mg pure rosuvastatin or equivalent to 10 mg rosuvastatin of the dextran–rosuvastatin conjugate, 35 mg maize starch, and 5 mg carboxymethylcellulose sodium were compressed into tablets with constant pressure.

3. Results and discussion

3.1. Synthesis and characterization of dextran–rosuvastatin conjugate (DRC)

The infrared spectras of dextran, rosuvastatin, and dextran–rosuvastatin conjugate were presented in Fig. 1. The characteristic absorption peaks of rosuvastatin were at 3409, 2967, 1601 and 1548, 1381 and 775–964 cm^{-1}. The band at 3409 cm^{-1} could be assigned to asymmetric and symmetric stretching vibrations of COO^- groups, the absence of —COOH absorption peaks is due to most carboxyl groups exists in the form of $—COO^-$ at neutral medium. The peak at 1601 cm^{-1} was the absorption of the —CO— groups in rosuvastatin. Furthermore, the band at 775–964 cm^{-1} typical for phenyl group of rosuvastatin could also be found in the spectrum of rosuvastatin (Astarita et al., 2007). Fig. 1(b) showed the FTIR spectra of dextran, there were four characteristic absorption peaks at 3420, 2926, 1648, and 1438–900 cm^{-1}. 3420 cm^{-1} could be assigned to hydroxyl groups (—OH), the peak at 2926 cm^{-1} was assigned to CH_2 symmetrical stretching vibrations, and the band at 1648 cm^{-1} was assigned to scissoring of two O—H bonds of absorbed water molecules. The band at 1438–900 cm^{-1} was typically of polysaccharide (Li, Niu, Yang, Nie, & Yang, 2011; Varshosaz et al., 2009; Wondraczek, Elschner, & Heinze, 2011). The dextran–rosuvastatin conjugate showed new bands at 1801 cm^{-1} and 1019 cm^{-1}. Those new bands confirmed that rosuvastatin' carboxymethylation took place on the dextran molecules.

The NMR spectrum of DRC was exhibited in Fig. 2. ^1H NMR (500 MHz, DMSO-d_6): δ = 7.72 (2′, 6′), 7.32 (3′, 5′), 6.81 (7), 6.53 (6),

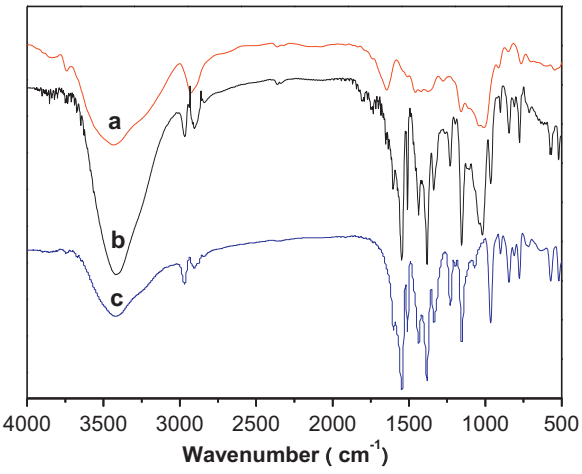


Fig. 1. FTIR spectra (a) dextran; (b) dextran-rosuvastaitin conjugate; and (c) rosuvastaitin.

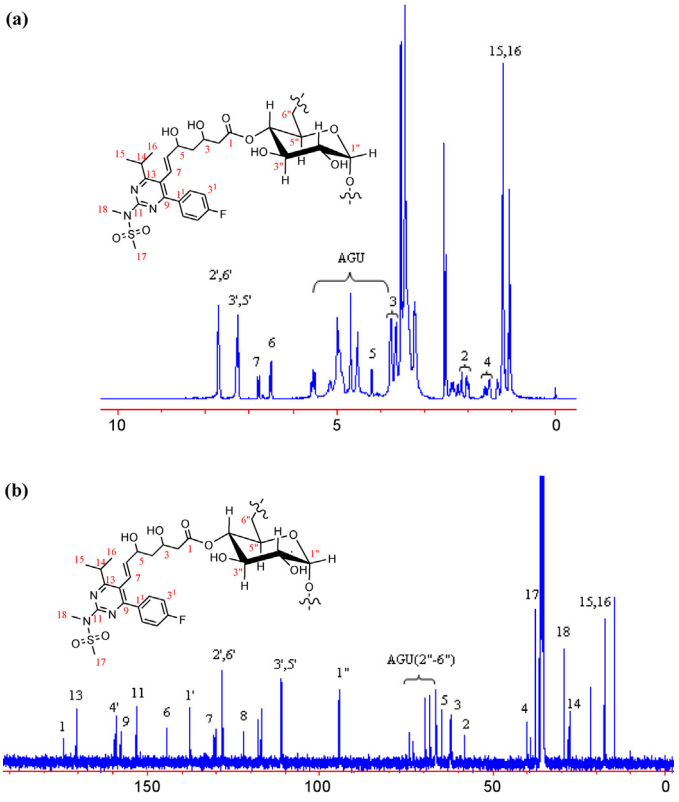


Fig. 2. (a) ^{1}H NMR spectra and (b) ^{13}C NMR of dextran–rosuvastatin conjugate.

5.4–3.5 (H-AGU), 3.78–3.5 (3, 17, 18), 2.17–1.52 (2, 4), 1.21 ppm (15, 16); ^{13}C NMR (125 MHz, DMSO-d_6): δ = 174.4 (1, O—CO), 170.5 (13), 159.2 (4′), 157.7 (9), 153.4 (11), 144.5 (6), 137.7 (1′), 130.7 (7), 128.3 (2′, 6′), 122.3 (8), 111.4 (3′, 5′), 94.4 (1″), 74.2–66.5 (AGU 2″-6″), 64.8 (5), 62.3 (3), 58.2 (2), 40.1 (4), 39.0 (17), 29.4 (18), 27.8 (14), 17.7 ppm (15, 16) (Astarita et al., 2007; Hornig et al., 2009).

The XRD spectra of DRC was demonstrated in Fig. 3(b), which showed a section microcrystal structure with three diffraction peaks at approximately 13°, 19° and 21° of 2θ. They had a great difference with the strong diffraction peaks of rosuvastatin at approximately 12.6°, 13.7°, 15°, 21.4°, 24.8°, and 27.5° of 2θ in Fig. 3(a). They were also different with the weak diffraction peaks of dextran at approximately 18° and 23° of 2θ in Fig. 3(c). This result suggested the structure change of compounds (Li et al., 2011).

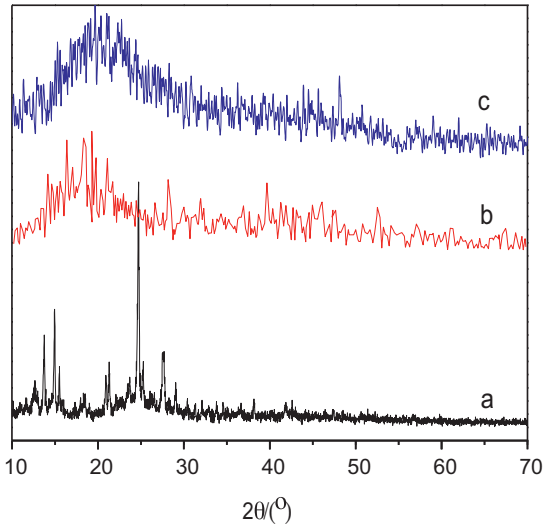


Fig. 3. XRD profile of rosuvastatin (a), dextran (b) and dextran–rosuvastatin conjugate (c).

3.2. Preparation and characterization of DRC microparticles

3.2.1. Effect of pH on DRC microparticles forming

Microparticles of DRC with different morphology were self-assembled by dialyzing from DMSO to anhydrous ethanol with different pH value. The scanning electron micrographs of these microparticles were presented in Fig. 4. The morphology of dextran–rosuvastatin conjugate microparticles at different pH appeared great change in shape compared to native rosuvastatin. Native rosuvastatin were irregular granules, as shown in Fig. 4(a). After self-assembling, the size of most DRC particles was declined to micrometer with relatively regular shape of granule, block or flake, as presented in Fig. 4. When the DMSO solution of DRC dialyzed against to ethanol solution with pH value of 9.0, self-assembled spheroids were obtained as shown in Fig. 4(b). Parts of these dispersed microspheres had good spherical shape with particle size of 1.5–4.5 μm, while parts of those microparticles were conglutinated. With the pH value of anhydrous ethanol increasing to 11.0, the microparticles were self-assembled to be blocks with particle size of 0.9–7.3 μm. However, the flakes with particle width of 1.0–6.1 μm were self-assembled with the pH increasing to 13.0. The width and length of these flakes were even. These results indicated that pH had a great effect on the morphology and size of self-assembled microparticles. We also found that the microparticles self-assembled fasterly as the pH value increased. The roundness of self-assembled microparticles was proportional to the time of self-assembling. So the roundness of self-assembled microparticles was worse as the pH value increased and self-assembling time decreased, as showed in Fig. 4. These results further illustrated the forming reason of DRC microparticles.

3.2.2. Effect of solvents on DRC microparticles forming

Besides the effect of pH, organic solvents also had a great influence on the size and morphology of the microparticles, since the formation of the microparticles during dialysis was based upon the slow exchange of the different organic solvents in both sides of dialysis membrane (Wang, Zhang, Wang, Wang, & Yang, 2007). These effects were also investigated in our work. Kinds of good solvent with different dipole moment and viscosity including DMF (η = 0.82 cp, μ = 3.82 D), DMAC (η = 0.92 cp, μ = 3.72 D), and DMSO (η = 2.24 cp, μ = 3.96 D) were studied. SEM images of DRC microparticles prepared by two days' dialysis of one organic solvent against anhydrous ethanol with the pH value of 11.0, as shown in Fig. 5.

Particles with different shape were got in each of these organic solvents. Nanoparticles of DCR with heterogeneous appearance were prepared in DMF. Nano-needles with the width of 200–600 nm were obtained in DMAC. Microparticles were self-assembled to be blocks with particle size of 0.9–7.3 μm in DMSO. There was a tendency to smaller size of nano-particles with the decrease in viscosity of the organic solvent. This phenomenon showed that the penetration of DRC into anhydrous ethanol slowed down with the increase of solvent's viscosity, thus slowing the process of crystallization and prolonging self-assembly process. Besides the difference in particle size, organic solvents also had an influence on nanospheres' morphology (Yu & Eisenberg, 1997; Zhang et al., 2004).

In addition, the self-assembly process was also influenced by the hydrogen-bonding interaction between DRC and organic solvents. The hydrogen-bonding complementary partners were the hydroxyl groups of DRC and carbonyl or sulphoxide groups of organic solvents. This hydrogen-bonding interaction would affect self-assembling speed and pattern of DRC. For example, when

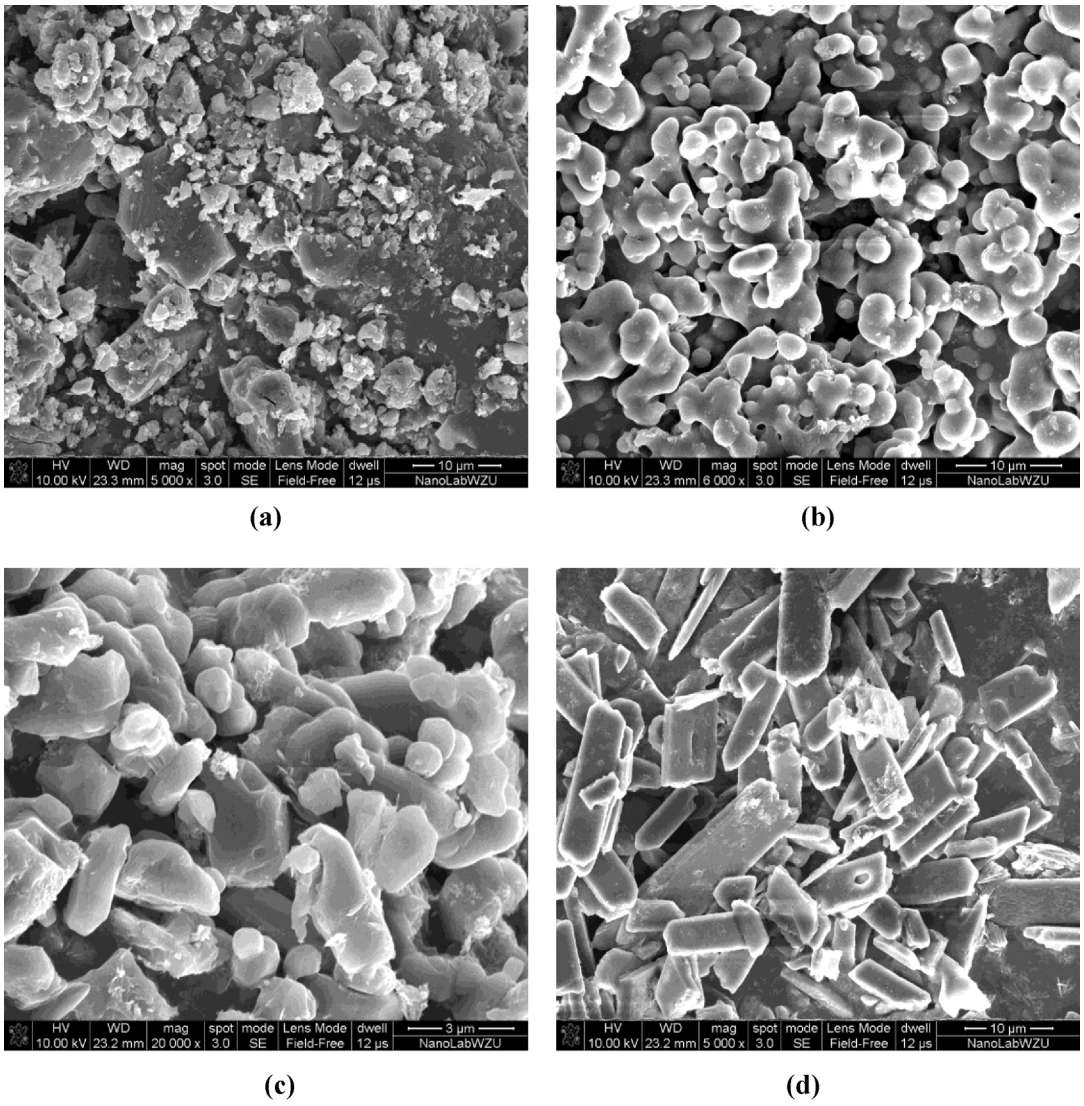


Fig. 4. SEM images of native rosuvastatin (a), and the self-assembled microparticles of dextran–rosuvastatin conjugate in anhydrous ethanol with different pH value of 9.0 (b), pH value of 11.0 (c), and pH value of 13.0 (d).

the DMSO solution of DRC penetrated into ethanol solvent, the intervention of ethanol moleculars would destroy the hydrogen-bonding between sulfoxide groups and the hydroxyl groups of DRC, with the result of increasing molecular force of DRC to gather into micron blocks. However, the destruction of the hydrogen bonds were difficult, which resulted in slow aggregation and self-assembling of DRC to become micron bulks, for DMSO had the maximum dipole moment in three different selected organic solvents and the hydrogen-bonding interaction between DRC and DMSO was strong. This result showed organic solvents had a great influence on DRC self-assembly process (Chang & Hou, 2008; Liu, Wang, & Yang, 2012). Therefore, we can control the size and morphology of the microparticles through the solvents selection.

3.2.3. *Three-dimensional network structure of DRC microparticles*

Besides different shapes of granule, block or flake were self-assembled, microparticles of DRC with three-dimensional network structure were also prepared. Representative SEM image of dextran–rosuvastatin conjugate was depicted in Fig. 6. Interpenetrated porous and network structures were observed in self-assembled formulation of DRC, by the way of dropping anhydrous ethanol into the DMSO solution of DRC inside the dialysis membrane. The pore diameters ranged from 70 to 1000 nm. The porous structural properties may be inclined to offer a larger specific surface area, which can provide a better interaction between solution and materials for easier solution uptake. Furthermore, the porous structural materials may have larger bulk for drugs loading and release, which is important for controlling the drugs loading and release (Wu et al., 2011).

The three dimensional networks porous structure of self-assembled DRC was significantly affected by the dropping speed of anhydrous ethanol. The formation of porous-mesh-structure was attributed to crowing out effect of ethanol molecules. DRC molecules presented to chains in DMSO solution inside the dialysis membrane. And these DRC chains would further wind to be membrane or mesh structure. When ethanol was dropped into the solution as a poor solvent for DRC, the mesh structure of DRC chains was crowded to porous morphology, therefore the formation of three-dimensional mesh structure by ethanol leaving after dryness (Chen, Loe, Blocki, Peng, & Raghunath, 2011; Morelli, Allen, & Ten Wolde, 2011).

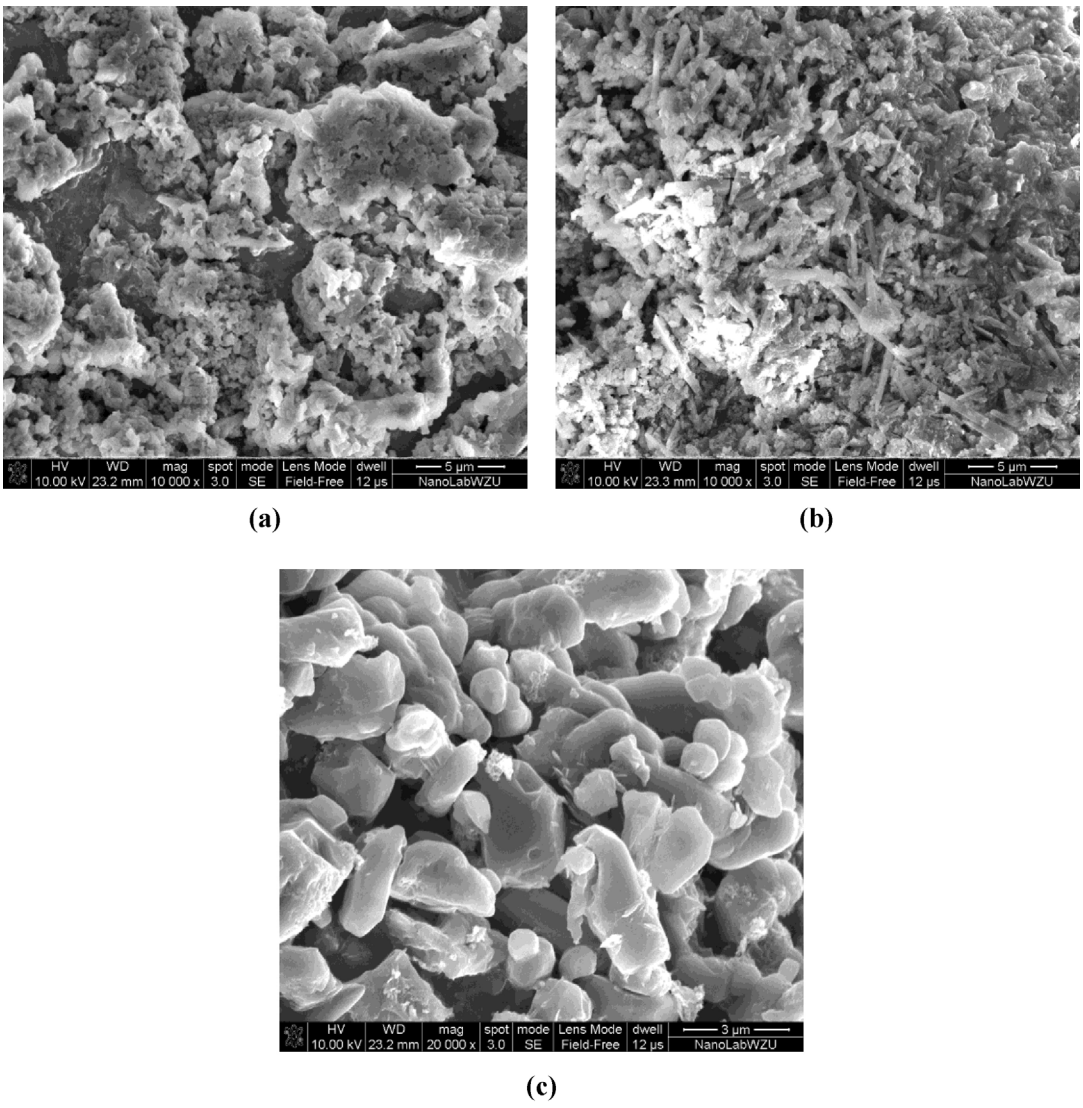


Fig. 5. SEM images of dextran–rosuvastatin conjugate microparticles in different organic solvents against anhydrous ethanol with pH value of 11.0. (a) DMF, pH = 11.0; (b) DMAC, pH = 11.0; and (c) DMSO, pH = 11.0.

3.3. *In vitro release studies*

In vitro release studies of DRC microparticles were carried out in phosphate buffer pH 7.4, as showed in Fig. 7. The tablet formulation containing pure rosovuastatin in Fig. 7A showed a burst release of 39.6% at 5 min and a release of 94.6% at 180 min. The maximum dissolution of this formulation maintained at about 97%. Compared with the pure rosovuastatin formulation, the tablets containing DRC microparticles exhibited slower release behavior. The initial release of rosovuastatin from these tablets was very low and had no burst release phenomenon. For instance, there were only 2.5% drug release in 5 min and 4.2% release in 60 min for the tablets containing spherical microparticles as shown in Fig. 7E. The slow-release of rosovuastatin from the formulations containing DRC microparticles was because the hydrolysis of DRC needed a long time, thus slowed down the rate of dissolution. Therefore, these formulations containing DRC acted as a slow-release delivery system.

In addition, the morphology of DRC microparticles had a great effect on the release behavior. The formulation containing microparticles with network structure showed the most rapid dissolution, compared with the other three microparticles of DRC, as shown in Fig. 7B. The loose reticulate structure was conducive to

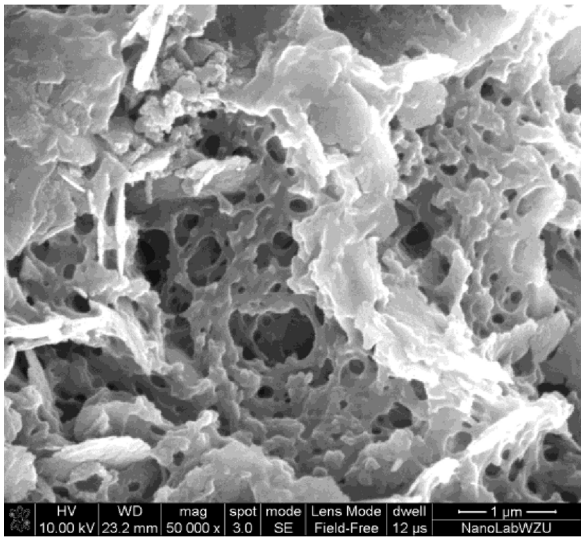


Fig. 6. SEM images of three-dimensional network structure of dextran–rosuvastatin conjugate in DMSO against anhydrous ethanol with pH value of 13.0.

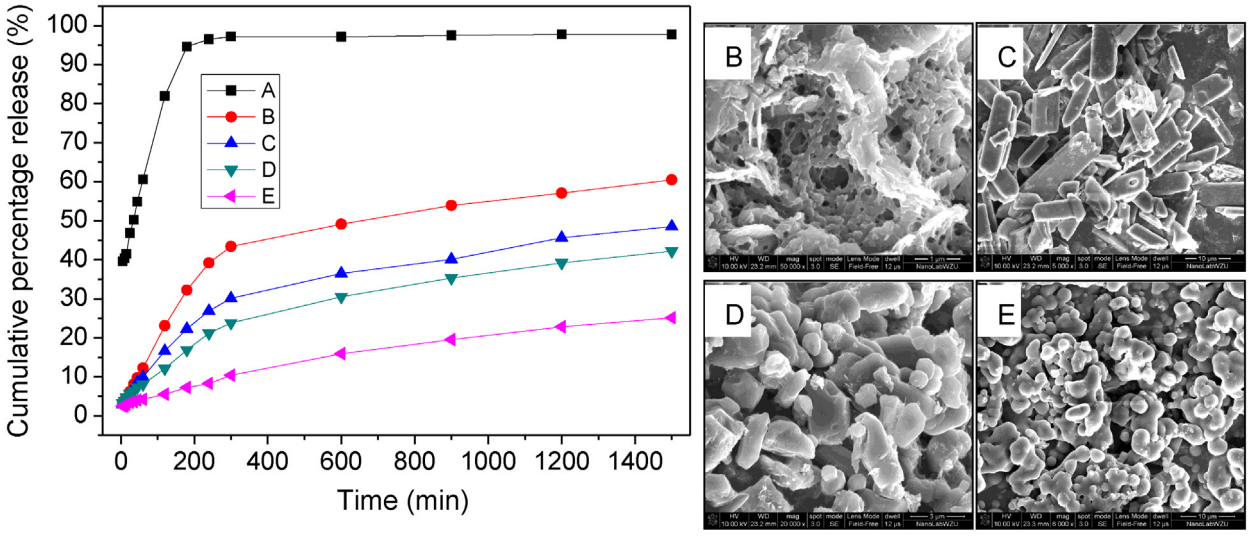


Fig. 7. In vitro release profile. (A) Pure rosovuastatin. (B) Dextran–rosuvastatin conjugate with three-dimensional network structure. (C) microparticles of DRC with the shape of flakes self-assembled at pH 9.0. (D) Microparticles of DRC with the shape of blocks self-assembled at pH 11.0. (E) Microparticles of DRC with spherical shape self-assembled at pH 13.0.

adsorption and storage of water molecules in the rich mesh. The hydrogen bond between water molecules and DRC molecules accelerated the dissolution of DRC in PBS solution, which also speeded up the hydrolysis of DRC and then accelerated the dissolution of rosovuastatin from the DRC.

On the contrary, the formulation involving the spherical microparticles showed the lowest release rate, as shown in Fig. 7E. Only 27.1% of drug was released even after 25 h. Since the intermolecular hydrogen-bond interactions, π–π stacking interactions and van der Waals force were fully formed during the long time preparation of DRC spheroids, the molecules of DRC were tightened. It was difficult for water molecules to insert DRC molecules, which slowed down the hydrolysis of DRC. Moreover, the amphiphilic character of DRC guarantied the self-assemble of microspheres with hydrophobic domains of rosuvastatin inside the spheres, which held back the dissolution of rosuvastatin. Therefore, the formulations of DRC microspheres exhibited the slowest release. It was close to linear release with the linear regression equation of $Y = 3.48374 + 0.01672 * t$ ($r = 0.99028$, SD = 1.17178, $n = 15$, $P < 0.0001$, Y: cumulative release).

4. Conclusion

Dextran–rosuvastatin conjugates were successfully synthesized and self-assembled to nano- or microparticles. It was testified that the organic solvents and pH had a great influence on the size and the morphology of the microparticles. Particles with different shape and size of microspheres, micro-blocks, micro-flakes or three-dimensional network structure were obtained by adjusting the pH value from 9.0 to 13.0. While by adjusting the pH value of 11.0, various kinds of nano- or microparticles were collected in different organic solvents. Nanoparticles were obtained in DMF, nanoneedles with the width of 200–600 nm were obtained in DMAC and micro-blocks with particle size of 0.9–7.3 μm were obtained in DMSO. These results suggested that the nano- or microparticles of dextran–rosuvastatin conjugates could be controlled through the solvents and pH selection. In vitro release studies of four kinds of DRC microparticles and control indicated that the formulations containing DRC showed a slow-release behavior. Among the four kinds selected microparticles, the formulation containing microparticles with network structure showed the most rapid dissolution, the formulation involving the spherical microparticles showed the lowest release rate. It meant that we could controlled the sustained release behavior by adjusting the morphology of DRC microparticles.

Acknowledgements

This work was supported by a grant from the National Natural Science Foundation of China (21272176, 21202122), the Fresh Talent Program of Zhejiang Province (2010R424036, 2011R424046).

References

Astarita, A., DellaGreca, M., Iesce, M. R., Montanaro, S., Previtera, L., & Temussi, F. (2007). Polycyclic compounds by sunlight exposure of the drug rosuvastatin in water. *Journal of Photochemistry and Photobiology A: Chemistry*, *187*, 263–268.

Ballantyne, M. C., Miller, E., & Chitra, R. (2004). Efficacy and safety of rosuvastatin alone and in combination with cholestyramine in patients with severe hypercholesterolemia: A randomized, open-label, plulticenter trial. *Clinical Therapeutics*, *26*, 1855–1864.

Chang, C. C., & Hou, S. S. (2008). Intercalation of poly(methyl methacrylate) into tyramine-modified layered silicates through hydrogen-bonding interaction. *European Polymer Journal*, *44*, 1337–1345.

Chen, C., Loe, F., Blocki, A., Peng, Y. X., & Raghunath, M. (2011). Applying macromolecular crowding to enhance extracellular matrix deposition and its remodeling in vitro for tissue engineering and cell-based therapies. *Advanced Drug Delivery Reviews*, *63*, 277–290.

Hornig, S., Bunjes, H., & Heinze, T. (2009). Preparation and characterization of nanoparticles based on dextran-drug conjugates. *Journal of Colloid and Interface Science*, *338*, 56–62.

Kostapanos, S. M., Derdemezis, S. C., Filippatos, D. T., Milionis, J. H., Kiortsis, N. D., Tselepis, D. A., et al. (2008). Effect of rosuvastatin treatment on plasma visfatin levels in patients with primary hyperlipidemia. *European Journal of Pharmacology*, *578*, 249–252.

Li, H. B., Niu, R., Yang, J. L., Nie, J., & Yang, D. Z. (2011). Photocrosslinkable tissue adhesive based on dextran. *Carbohydrate Polymers*, *86*, 1578–1585.

Liebert, T., Hornig, S., Hesse, S., & Heinze, T. (2005). Nanoparticles on the basis of highly functionalized dextrans. *Journal of American Chemical Society*, *127*, 10484–10485.

Liu, H. L., Wang, D., & Yang, X. L. (2012). Preparation of polymer@titania raspberry-like core-corona composite via heterocoagulated self-assembly based on hydrogen-bonding interaction. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, *397*, 48–58.

Martin, P. D., Warwick, M. J., Dane, A. L., Brindley, C., & Short, T. (2003). Absolute oral bioavailability of rosuvastatin. *Clinical Therapeutics*, *25*, 2553–2563.

Martin, P. D., Warwick, M. J., Dane, A. L., Hill, S. J., Giles, P. B., Phillips, P. J., et al. (2003). Metabolism, excretion, and pharmacokinetics of rosuvastatin in healthy adult male volunteers. *Clinical Therapeutics*, *25*, 2822–2835.

Mehvar, R. (2000). Dextrans for targeted and sustained delivery of therapeutic and imaging agents. *Journal of Controlled Release*, *69*, 1–25.

Morelli, M. J., Allen, R. J., & Ten Wolde, P. R. (2011). Effects of macromolecular crowding on genetic networks. *Biophysical Journal*, *101*, 2882–2891.

Sadekar, S., & Ghandehari, H. (2012). Transepithelial transport and toxicity of PAMAM dendrimers: Implications for oral drug delivery. *Advanced Drug Delivery Reviews*, *64*, 571–588.

Sarver, W. R., Bills, E., Bolton, G., Bratton, D. L., Caspers, L. N., Dunbar, B. J., et al. (2008). Thermodynamic and structure guided design of statin based inhibitors of 3-hydroxy-3-methylglutaryl coenzyme a reductase. *Joural of Medicinal Chemistry*, *51*, 3804–3813.

Shahnaz, G., Perera, G., Sakloetsakun, D., Rahmat, D., & Bernkop-Schnürch, A. (2010). Synthesis, characterization, mucoadhesion and biocompatibility of thiolated carboxymethyl dextran–cysteine conjugate. *Journal of Controlled Release*, *144*, 32–38.

Tang, M. H., Dou, H. J., & Sun, K. (2006). One-step synthesis of dextran-based stable nanoparticles assisted by self-assembly. *Polymer*, *47*, 728–734.

Varshosaz, J., Emami, J., Tavakoli, N., Fassihi, A., Minaiyan, M., Ahmadi, F., et al. (2009). Synthesis and evaluation of dextran–budesonide conjugates as colon specific prodrugs for treatment of ulcerative colitis. *International Journal of Pharmaceutics*, *365*, 69–76.

Veronese, F. M., & Morpurgo, M. (1999). Bioconjugation in pharmaceutical chemistry. *Il Farmaco*, *54*, 497–516.

Wang, Y., Zhang, J. H., Wang, Z. H., Wang, Z., & Yang, B. (2007). PbS nanoparticles/polymer composite aggregates through self-assembly of amphiphilic copolymer containing cross-linked hydrophilic block. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, *292*, 159–164.

Wondraczek, H., Elschner, T., & Heinze, T. (2011). Synthesis of highly functionalized dextran alkyl carbonates showing nanosphere formation. *Carbohydrate Polymers*, *83*, 1112–1118.

Wu, W., Liu, J., Cao, S. Q., Tan, H., Li, J. S., Xu, F. J., et al. (2011). Drug release behaviors of a pH sensitive semi-interpenetrating polymer network hydrogel composed of poly (vinyl alcohol) and star poly[2-(dimethylamino) ethyl methacrylate]. *International Journal of Pharmaceutics*, *416*, 104–109.

Yu, Y., & Eisenberg, A. (1997). Control of morphology through polymer-solvent interactions in crew-cut aggregates of amphiphilic block copolymers. *Journal of American Chemical Society*, *119*, 8383–8384.

Zhang, W. Q., Shi, L. Q., An, Y. L., Gao, L. C., Wu, K., & Ma, R. J. (2004). A convenient method of tuning amphilic block copolymer micelle morphology. *Macromolecules*, *37*, 2551–2555.

Zhang, L., Strong, M., Qiu, J. W., Lesko, J., & Huang, L. S. M. (2005). Scientific perspectives on drug transporters and their role in drug interactions. *Molecular Pharmaceutics*, *3*, 62–69.