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A new supramolecular gel via host–guest complexation with cucurbit[8]uril and *N*-(4-diethylaminobenzyl)chitosan

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ABSTRACT

A novel supramolecular gel has been prepared via host–guest interaction between cucurbit[8]uril (Q[8]) and *N*-(4-diethylaminobenzyl)chitosan (EBCS). The structure of supramolecular gel has been characterized. The spectrum of ^{1}H NMR demonstrated the benzene ring of EBCS is reside inside the hydrophobic cavity of Q[8] and the host–guest interaction between Q[8] and EBCS was the main driving force for the formation of the supramolecular gel. The network structure of the xerogel of Q[8]/EBCS gel was observed by SEM. The Q[8]/EBCS gel system showed thermosensitive and pH-sensitive properties. The physical characterization by SEM, DSC, TG demonstrated the distinguished characters, which proved the formation of supramolecular gel instead of physical blending. The *in vitro* release study of the 5-fluorouracil-loaded supramolecular gel showed that sustained release profile in acidic condition, suggesting that Q[8]/EBCS gel could be a potential carrier for pH-sensitive drug controlled release system.

1. Introduction

With the introduction of supramolecular chemistry by Lehn (1988), supramolecular chemistry had gained great interests to scientists. The recent studies of supramolecular chemistry facilitated the development of functional materials with supramolecular structure, such as self-assembly nanocapsules, self-assembly monolayer, supramolecular gels, and so on (Kim, 2002; Kim et al., 2007; Liu, Xu, Wu, & Feng, 2004; Rauwald, Del Barrio, Loh, & Scherman, 2011; Sangeetha & Maitra, 2005; Steed, 2011; Yang et al., 2005). Especially supramolecular gels based on host macrocyclic compounds have attracted attention due to their biocompatibility, biodegradability and stimuli-responsive characteristic (Charlot & Auzly-Velty, 2007; Choi, Yamamoto, Ooya, & Yui, 2005; Ge, Hu, Huang, & Liu, 2009; Guo, Jiang, Pispas, Yu, & Zhou, 2008; Van De Manakker, Vermonden, El Morabit, Van Nostrum, & Hennink, 2008; Yang & Kim, 2010).

The new family of macrocyclic host molecules, cucurbit[*n*]urils (Q[*n*], *n* = 5–10) with its unique structure of center hydrophobic cavity and two identical cavity portals lined by the carbonyl groups of the glycoluril units, can selectively interact with guest molecules to form the clathrate compounds (Corma et al., 2007; Lagona, Mukhopadhyay, Chakrabarti, & Isaacs, 2005; Zhang, Wu, Li, Wang, & Dou, 2006). This host–guest interaction is sensitive to external environment, such as temperature and pH, which lead to different supramolecular structures and properties (Angelos et al., 2009; Hennig, Bakirci, & Nau, 2007). In addition, cucurbit[*n*]urils have other characteristics unmatched by other families of macrocyclic compounds, such as highly symmetrical structure, good thermal stability and low toxicity (Hettiarachchi et al., 2010; Uzunova, Cullinane, Brix, Nau, & Day, 2010). Thereby, cucurbit[*n*]urils are considered as new functional material compounds with extensive applications in various fields, such as biomedicine, materials science and pharmacology (Kim, Agasti, Zhu, Isaacs, & Rotello, 2010). Furthermore, the supramolecular gel with 3D network structure based on host–guest interaction of Q[n] shows excellent biological properties and great potential applications in drug delivery system (Appel et al., 2010; Hwang et al., 2007; Yang, Tang, & Wang, 2009).

Chitosan (CS) is an ideal candidate for hydrogel formation due to its non-toxic, biocompatible, and biodegradable characteristics. Chitosan based materials have been widely used in the drug delivery of therapeutic agents for sustained release (Chenite, Buschmann, Wang, Chaput, & Kandani, 2001; Ravi Kumar, Muzzarelli, Muzzarelli, Sashiwa, & Domb, 2004; Tien, Lacroix, Ispas-Szabo, & Mateescu, 2003). In particular, thermal and pH sensitive chitosan gels have been used in the target delivery of some drug compounds (Lin, Chen, & Luo, 2007; Ta et al., 2008; Zhou et al., 2008). However, preparation of the chitosan hydrogels usually

Abbreviations: Q[8], cucurbit[8]uril; EBCS, *N*-(4-diethylaminobenzyl)chitosan; CS, chitosan; SEM, scanning electron microscope; XRD, X-ray powder diffraction; TG, thermogravimetric method; DSC, differential scanning calorimetry; 5-Fu, 5-fluorouracil.

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requires the use of chemical crosslinker and chemical reactions which may be potentially deleterious to the drug loaded. The formation of hydrogels by supramolecular interactions (noncovalent crosslinking referring to Vander Waals force, electrostatic action, hydrogen binding, $\pi-\pi$ stacking and hydrophobic action based on synergistic stereochemistry effect, etc.) is considered to be a useful method of preparing hydrogels for drug delivery since these gels are likely to be more biocompatible (Ravi Kumar et al., 2004).

Accordingly, it is expected that the supramolecular gel based on chitosan compounds with Q[8] as a cross-linker for noncovalent crosslinking, should be more biocompatible and 'smart' (i.e. pH and temperature sensitivities) for drug controlled release because of the above-mentioned excellent properties of Q[8] and chitosan derivatives. Thus, in this paper, a chitosan derivative, *N*-(4-diethylaminobenzyl)chitosan (EBCS), was synthesized to fabricate supramolecular gel system based on modified chitosan and cucurbit[8]uril for pH sensitive drug controlled release. It is the first time to report the formation of this type of gel based modified chitosan and Q[8]. The supramolecular gel structures and its physical characteristics, such as morphology, crystallinity and thermal properties of the xerogel have been characterized through SEM, X-ray powder diffraction, thermogravimetric method and differential scanning calorimetry, respectively. The thermosensitive and pH-sensitive properties of Q[8]/EBCS gel system were then investigated. The release profiles of 5-fluorouracil (5-Fu) embedded into Q[8]/EBCS gel were also studied in different simulated pH media.

2. Materials and methods

2.1. Materials

Cucurbit[8]uril (Q[8]) was from Fluka Company(Switzerland). Chitosan (CS) with deacetylation degree of 95% and the molecular weight range of 50,000–100,000 was supplied by Zhejiang Aoxing Biotechnology Co., Ltd. (China). 4-(Diethylamino)benzaldehyde (98%), sodium borohydride ($NaBH_4$) and 5-Fu were purchased from Shanghai Jingchun Reagent Co., Ltd. (China). Dialysis bags with molecular weight cutoff (MWCO) of 12–14 kDa were obtained from Sino-American Biotechnology Co., Ltd. (China). Other chemicals and reagents used in the study were of analytical grade and used without further purification.

2.2. Synthesis of EBCS

Chitosan powder (0.8 g) was dissolved in 2% (w/v) aq HOAc (80 mL). Then a solution of 4-(diethylamino)benzaldehyde (0.9 g) in EtOH (20 mL) was added to the chitosan solution. After stirring at 50 °C for 5 h, an aqueous solution of $NaBH_4$ (1.0 g, 10 mL) was added dropwise to the reaction mixture which was maintained at room temperature. After the reactive fluid was stirred consecutively at room temperature for 5 h, a large amount of acetone was poured into the reaction mixture to produce the precipitate, following by filtration, wash with acetone, and then vacuum drying at 25 °C to constant weight to obtain a yellow powder of EBCS.

2.3. Preparation of Q[8]/EBCS gel and drug-loaded Q[8]/EBCS gel

EBCS (0.2 g) was dissolved in 1 mol/L HCl. Q[8] powder (0.05 g) was then slowly added to the EBCS solution, while gentle stirring for 5 min at room temperature until fully mixed. Then the mixture was put at 25 °C for more than 30 min to convert into a solid-like material, the Q[8]/EBCS supramolecular gel was formed.

To prepare the drug-loaded Q[8]/EBCS gel, 5-Fu (5 mg) was added to the above Q[8]/EBCS mixture while it was in solution prior to Q[8]/EBCS gel formation. The mixture was stirred continuously for 5 min, and was set for more than 30 min at 25 °C till the solidified gel was formed.

2.4. Characterization of Q[8]-EBCS gel

The Q[8]/EBCS gel was vacuum dried for 24 h by an FD-1 freeze-dry machine (Boyikang Tech. Co., Beijing, China) to prepare xerogel. Cross section of the xerogel of Q[8]/EBCS was sputtered with gold in order to make the sample conductive, and visualized using an environmental scanning electron microscope (XL30 ESEM-TMP, Philips-FEI Co., Holland) at 20 kV accelerating voltage. The photographs were taken at 2000× and 500× magnification.

^{1}H NMR spectroscopy. ^{1}H NMR spectrum was performed using a Bruker Advance III spectrometer at 400 MHz at room temperature with DCl/D_2O as the solvent for the samples of Q[8]-EBCS system. The chemical shifts were reported as ppm and D_2O signal (4.80 ppm) for ^{1}H NMR experiments was as reference.

X-ray powder diffraction (XRD). X-ray powder diffraction patterns were recorded on a X'Pert Pro MPD X diffractometer. The voltage and current were set to 40 kV and 30 mA, respectively. The scanning rate was 1° min^{-1} over a 2θ range of 5–50°.

Simultaneous thermal analysis (STA). Thermal analyses were performed on Netzsch STA 449C equipment. Samples of CS, EBCS, Q[8] and Q[8]/EBCS xerogel were sealed into alumina pans, respectively. Samples were heated over a temperature range of 25–700 °C at a heating rate of 10 °C min^{-1} under nitrogen gas flow.

2.5. Release profiles of 5-Fu from Q[8]/EBCS gel

In *in vitro* release study, Q[8]/EBCS gel loaded with 5-Fu was studied in phosphate buffer solution (pH 6.8), citric acid–disodium hydrogen phosphate buffer solution (pH 4.0) and 0.1 mol/L HCl solution (pH 1.2), respectively. Raw 5-Fu (5 mg), Q[8]/EBCS gel and Q[8]/EBCS drug-loaded gel (with 5 mg 5-Fu) were sealed in three dialysis bags, respectively, following by lowering the bags into three plastic tubes filled with 20 mL of the tested buffer solution. The tubes were set in a water bath at 37 ± 0.5 °C, and a constant shaking speed of 100 rpm. At regular time interval, 1 mL of the medium was removed (then replaced with 1 mL of fresh buffer solution to keep the volume constant) and diluted at 1:25 ratio for UV absorbance measurement at 265 nm on a 752PC Spectrum ultraviolet–vis spectrophotometer (Shanghai Spectrum Instrument Co., China). The experiments were replicated three times. The cumulative 5-Fu release percentage at various time points was obtained through 5-Fu standard calibration curves.

3. Results and discussion

3.1. Characteristics and structure morphology of Q[8]/EBCS gel

After mixing light yellow EBCS solution with Q[8], the formation of yellow gel could be observed after 30 min mixing at 25 °C. The gelation process of the supramolecular gel based on the interaction between Q[8] and EBCS was shown as Fig. 1. When this gel was heated at 50 °C, it could quickly turn into the yellow solution. When the warm solution was cooled at room temperature, the solution was converted to the gel state again. This solution–gel transformation could be repeated several times under heat–cool cycles, indicating that this supramolecular gel was sensitive to the temperature change.

The structure morphology of the xerogel prepared by freeze drying was investigated by scanning electron microscopy. The SEM images of Q[8]/EBCS (Fig. 2) showed cross-linked network structure with many cavities for Q[8]/EBCS gel, and the pore sizes of Q[8]/EBCS xerogel are in the range of 5–10 μm.

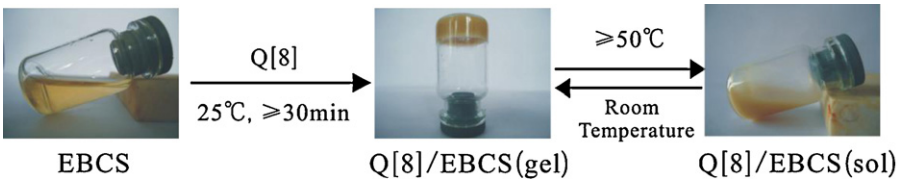

Fig. 1. The pictures of Q[8]/EBCS gel.

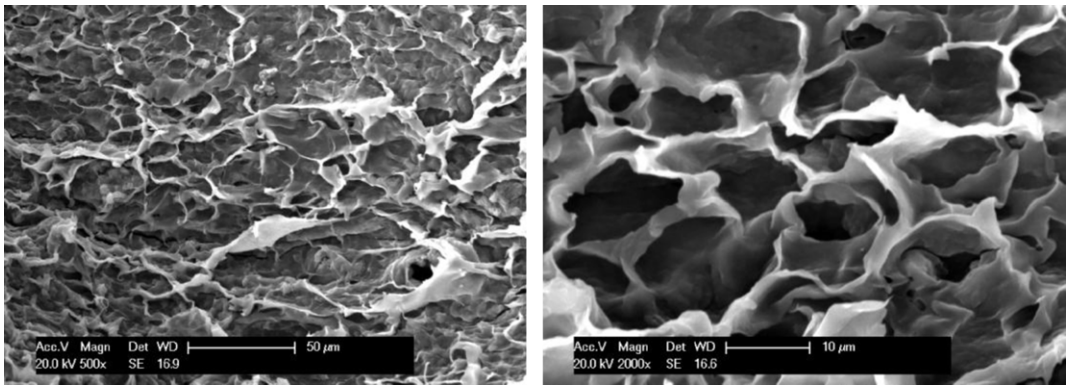

Fig. 2. The SEM images of Q[8]/EBCS xerogel.

The proposed structural model of Q[8]/EBCS supramolecular gel is shown in Fig. 3. Q[8] has a larger cavity volume of 479 Å^3 and can simultaneously accommodate two guests (Appel et al., 2010). It was observed that the gelation phenomenon did not appear between Q[8] and chitosan. When chitosan was modified with the *N*,*N*-diethylaminobenzyl groups, the obtained EBCS possessed the aromatic moiety for the interaction with Q[8] to form supramolecular gel, meanwhile basic diethylamino groups were also helpful to bring into the property of pH sensitivity for gel. In acidic conditions, diethylamino groups of EBCS were charged and could bind to Q[8] portals through electrostatic interaction, which promote supramolecular assembling, and two aromatic rings at different sites of EBCS could then enter the hydrophobic cavity of Q[8] and the network structure was formed favorably because of the inclusion of Q[8] and EBCS. In the formation of the supramolecular gel process, Q[8] has played the role of physical cross-linking agent. A sharp gel-to-sol transition was observed upon heating to 50 °C, which is presumably due to the non-covalent host–guest interaction being weak, benzene ring moving out from the Q[8] cavity when gel heated, thus the loss of cross-linking leads to disassemble and turns into a fluid. Q[8] and EBCS can be re-assembled to form a gel upon cooling at room temperature.

The gelation is also largely affected by the pH value of the solution. As we know that the pK_a of amines in chitosan is around 6.3–6.4 (Ravi Kumar et al., 2004). It could be reasonably speculated that the pK_a of EBCS should be in the vicinity of 6 due to that the amino groups of chitosan were partly substituted with the *N*,*N*-diethylaminobenzyl groups. The protonation of diethylamino groups on side chain of EBCS in strong acidic media results in ion–dipole interactions between ammonium positive ions and Q[8] portals, which promotes the supramolecular assembly of Q[8]/EBCS gel. In contrast, a higher pH value (i.e. 6.8) led to a remarkable decrease of the diethylamino group protonation. Hence, electrostatic repulsion between dissociative diethylamino groups and the

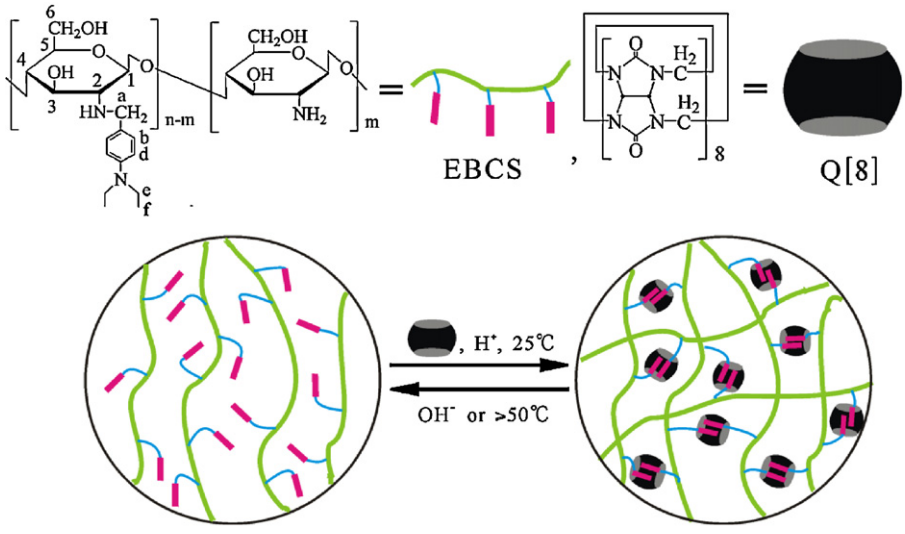

Fig. 3. Schematic diagrams for the formation of Q[8]/EBCS supramolecular gel.

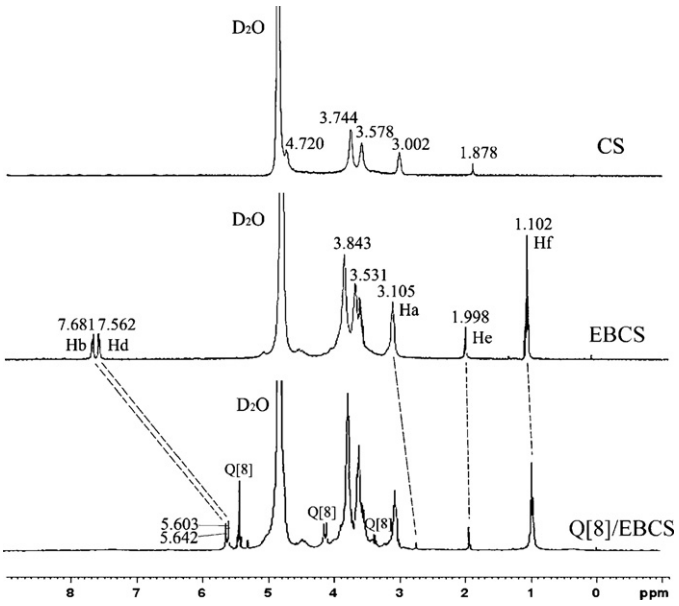

Fig. 4. ^{1}H NMR spectra of CS, EBCS and Q[8]/EBCS gel.

oxygen atoms of Q[8] portals could hamper supramolecular assembling. Therefore, this gel has an interesting property of pH and thermo-sensitivity.

3.2. ^{1}H NMR spectra of chitosan, EBCS and Q[8]/EBCS gel

The ^{1}H NMR spectra of chitosan, EBCS and Q[8]/EBCS gel was shown in Fig. 4. The peak at 4.80 ppm due to the protons in C1 of chitosan was covered by the D_2O peak and was unable to be distinguished. The chemical shift of 1.878 ppm was signed to methyl protons in the residual *N*-acetyl group of chitosan, and the signals at 3.002 ppm, 3.578 ppm and 3.744 ppm were all for the protons of C2–C6 on pyranose ring (Singh & Dutta, 2009). In the spectrum of EBCS, ^{1}H peaks at 7.562–7.681 ppm were assigned to the H_b and H_d of the benzene ring. The signals at 1.102 ppm were for H_f of diethylamino group of EBCS. As shown in the spectrum of Q[8]/EBCS, there were some new signals at 3.554 ppm, 4.153 ppm and 5.438 ppm which were for the protons of Q[8] (Buschmann, Wego, Zielesny, & Schollmeyer, 2006). With addition of Q[8], the H_b and H_d chemical shifts of benzene ring in EBCS moved to 5.603–5.642 ppm. It was also found that the peaks of H_a, H_e and H_f were all shifted to high magnetic field. The occurrence of these changes was caused by the clathrate interaction between EBCS and Q[8]. The aromatic rings of side chains of EBCS could enter the hydrophobic cavity of Q[8] and were shielded by Q[8], so that the chemical shifts were changed.

3.3. XRD analysis

The XRD diagrams of chitosan, EBCS, Q[8] and Q[8]/EBCS gel were illustrated in Fig. 5. The XRD diffractogram of chitosan consisted of two major crystalline peaks around 10.7° (2θ) and 23.2° (2θ), which was in agreement with Samuels's report (Samuels, 1981). There were several sharp diffraction peaks at 14.7°, 20.7° and 29.3° in the diffractogram of Q[8], which demonstrated high degree of crystalline for Q[8]. Compared with chitosan, the crystalline structure of EBCS had been remarkably modified. The peak intensity at 23.5° was greatly decreased, and diffraction peak around 10° had disappeared. This is consistent with the previous studies that the crystallinity of chitosan decreased gradually with the introducing of benzene structure units into the side chain of chitosan (Teng, Lee, Wang, Shin, & Kim, 2008). XRD patterns of Q[8]/EBCS xerogel

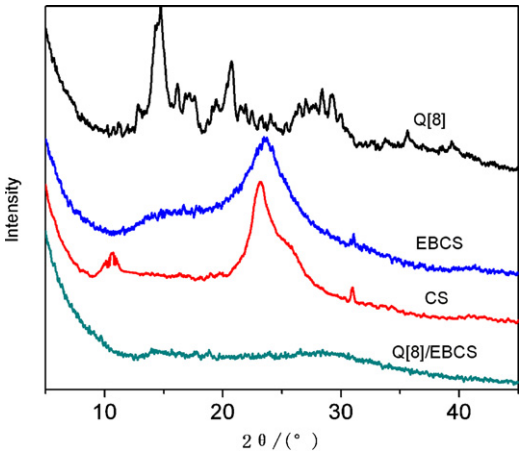

Fig. 5. XRD diagrams of CS, EBCS, Q[8] and Q[8]/EBCS gel.

did not have characteristic peaks as Q[8] and EBCS. It suggested that Q[8]/EBCS was not the physical blend of Q[8] and EBCS, instead, it is in an amorphous state after EBCS interacted with Q[8].

3.4. Analysis of thermal properties

Thermal characteristics of chitosan, EBCS, Q[8] and Q[8]/EBCS gel were examined by TG and DSC. As shown in Fig. 6, the TG curve of Q[8] showed two major mass loss steps under nitrogen. The first mass loss between 50 °C and 100 °C might be associated with the loss of adsorptive water in Q[8]. The second transition occurred near 450 °C was considered as thermal degradation of Q[8]. It suggested that Q[8] should have a higher thermostability. Chitosan began gradual mass loss (about 45% weight loss) over the temperature range of 260–380 °C, which is likely due to the glycosidic bond fracture of chitosan. Compared with the TG curve for chitosan, EBCS started to lose weight earlier and had about 43% mass loss while heating over the range of 252–373.5 °C, and decreased about 30% between 182 and 299 °C. TG curve of EBCS tended to be stable after 500 °C.

The DSC thermograms of chitosan, EBCS, Q[8] and Q[8]/EBCS gel were showed in Fig. 7. It was found that Q[8] had an exothermic peak at 471 °C, corresponding to the thermal degradation process of Q[8]. The thermo stability of EBCS was not the same as chitosan

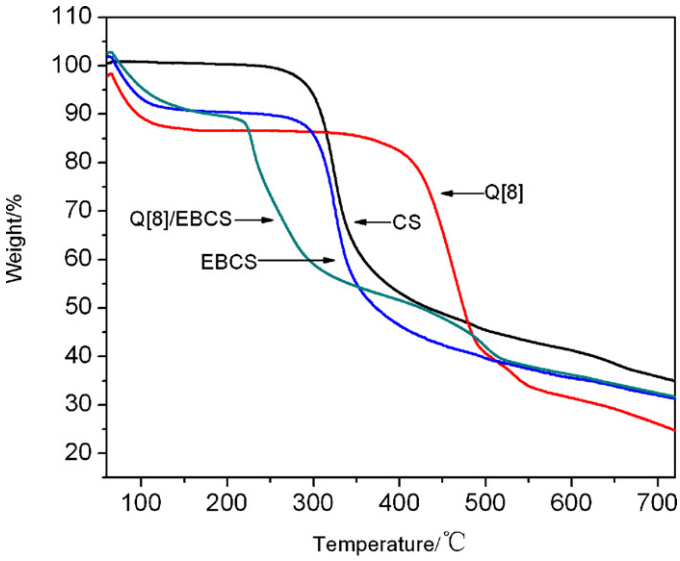

Fig. 6. TG diagrams of CS, EBCS, Q[8] and Q[8]/EBCS gel.

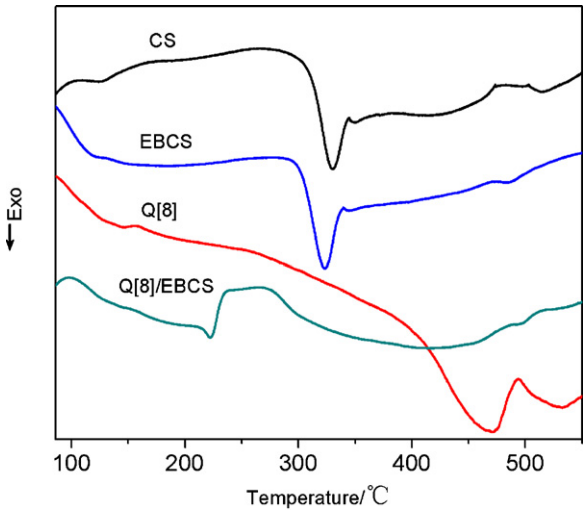

Fig. 7. DSC thermograms of CS, EBCS, Q[8] and Q[8]/EBCS gel.

due to their different crystalline properties (Wan, Creber, Peppley, & Bui, 2003; Wang et al., 2009). A thermopositive peak of chitosan at 330.5 °C was likely associated with the decomposition of chitosan main chain. And exothermic peak of EBCS appeared at 323 °C, which was lower than that for chitosan. The DSC curve of Q[8]/EBCS gel showed that the degradation temperature of xerogel at 222 °C and the position of exothermic peak was different from those of Q[8] and EBCS. EBCS had a poor thermal stability and showed a little lower degradation temperature than that of chitosan. With these distinct thermal characteristics, Q[8]/EBCS gel has been demonstrated as a novel supramolecular gel, not the simple mixture of Q[8] and EBCS.

3.5. *In vitro* drug release studies

In vitro release studies of 5-Fu from the Q[8]/EBCS supramolecular gel were carried out in mediums with three different pH values (see Fig. 8). All the releasing curves of raw 5-Fu without supramolecular gel started with an initial burst release in three pH buffer solutions. Within 180 min, the cumulative dissolution rates of raw 5-Fu were both nearly 100% at pH 6.8, 4.0, and 1.2 buffer solutions. As shown in Fig. 8, the cumulative release percentage of 5-Fu loaded in Q[8]/EBCS supramolecular gel was increased with the medium acidity decreased. Especially in HCl solution (pH 1.2), 5-Fu release rate was 41.13% from gel in 180 min and gradually increased to 72.86% at 600 min. The drug release from Q[8]/EBCS supramolecular gel has slowed down significantly in acidic solution (pH 1.2), in comparison to those in pH 4.0 and 6.8. According to these results, it was concluded that the release rates of 5-Fu could be control-released by supramolecular gel, and modulated via different pH media.

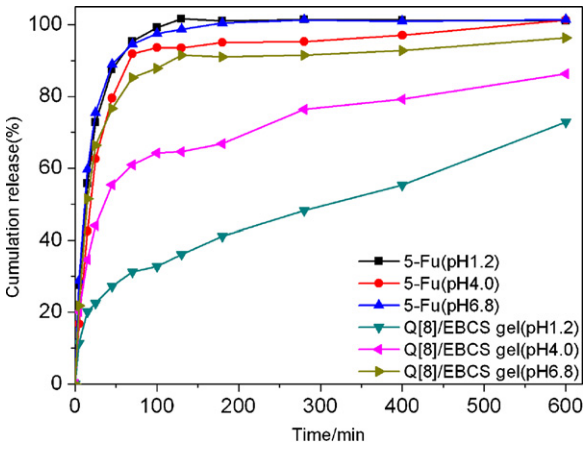

Fig. 8. The cumulative dissolution rate of 5-Fu and Q[8]/EBCS drug-loaded gel.

In lower pH media, the drug release was slower. The possible reason might be that there are more ammonium positive ions which can bind to Q[8] portals through electrostatic interaction, and promote supramolecular assembly, the network structure of Q[8]/EBCS gel remained intact in the strong acidic solutions, which leads to the slow release of model drug from the gel. Because of electrostatic repulsion between dissociative diethylamino groups and the oxygen atoms of Q[8] portals in a higher pH value (i.e. 6.8), the interaction between Q[8] and EBCS became weakened and the network of supramolecular gel was gradually degraded with increasing pH, thus the drug could be released quickly without the hindrance of gel network structure. These research results suggested that pH is the critical element for structural stabilization of Q[8]/EBCS gel and this supramolecular gel was sensitive to the changes of pH value.

In addition, it has been reported that Q[8] showed very low toxicity for using as molecular containers in advanced drug delivery system (Hettiarachchi et al., 2010; Uzunova et al., 2010) and the analogs of EBCS have been proved to be an effective gene carriers (Rojanarata et al., 2008). Therefore, based on the results in this study and above literature, the prepared Q[8]/EBCS gel could be a promising pH sensitive material for drug controlled release system.

4. Conclusions

N-(4-diethylaminobenzyl) chitosan was synthesized and characterized to prepare a novel supramolecular gel with Q[8] for the first time. The structure morphology of Q[8]/EBCS xerogel analyzed with SEM illustrated network structure. Supramolecular cross-linking via the host–guest interaction between Q[8] and aromatic ring of EBCS was confirmed by the spectrum of ^{1}H NMR. This supramolecular gel system showed thermosensitive and pH-sensitive properties. The release profiles of 5-Fu embedded in the gel demonstrated that Q[8]/EBCS gel could be a potential pH-sensitive drug carrier.

Acknowledgments

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References

Angelos, S., Khashab, N. M., Yang, Y. W., Trabolsi, A., Khatib, H. A., Stoddart, J. F., et al. (2009). pH clock-operated mechanized nanoparticles. *Journal of the American Chemical Society*, *131*, 12912–12914.

Appel, E. A., Biedermann, F., Rauwald, U., Jones, S. T., Zayed, J. M., & Scherman, O. A. (2010). Supramolecular cross-linked networks via host–guest complexation with cucurbituril. *Journal of the American Chemical Society*, *132*, 14251–14260.

Buschmann, H. J., Wego, A., Zielesny, A., & Schollmeyer, E. (2006). Structure, electronic properties and NMR-shielding of cucurbit[*n*]urils. *Journal of Inclusion Phenomena and Macrocyclic Chemistry*, *54*, 85–88.

Charlot, A., & Auzly-Velty, R. (2007). Synthesis of novel supramolecular assemblies based on hyaluronic acid derivatives bearing bivalent β-cyclodextrin and adamantane moieties. *Macromolecules*, *40*, 1147–1158.

Chenite, A., Buschmann, M., Wang, D., Chaput, C., & Kandani, N. (2001). Rheological characterization of thermogelling chitosan/glycerolphosphate solutions. *Carbohydrate Polymers*, *46*, 39–47.

Choi, H. S., Yamamoto, K., Ooya, T., & Yui, N. (2005). Synthesis of poly(ε-lysine)-grafted dextrans and their pH- and thermosensitive hydrogelation with cyclodextrins. *A European Journal of Chemical Physics and Physical Chemistry*, *6*, 1081–1086.

Corma, A., Garcia, H., Montes-Navajas, P., Primo, A., Calvino, J. J., & Trasobares, S. (2007). Gold nanoparticles in organic capsules: A supramolecular assembly

of gold nanoparticles and cucurbituril. *Chemistry – A European Journal*, *13*, 6359–6364.
Ge, Z., Hu, J., Huang, F., & Liu, S. (2009). Responsive supramolecular gels constructed by crown ether based molecular recognition. *Angewandte Chemie International Edition*, *48*, 1798–1802.
Guo, M., Jiang, M., Pispas, S., Yu, W., & Zhou, C. (2008). Supramolecular hydrogels made of end-functionalized low-molecular-weight PEG and α-cyclodextrin and their hybridization with SiO_2 nanoparticles through host–guest interaction. *Macromolecules*, *41*, 9744–9749.
Hennig, A., Bakirci, H., & Nau, W. M. (2007). Label-free continuous enzyme assays with macrocycle-fluorescent dye complexes free. *Nature Methods*, *4*, 629–632.
Hettiarachchi, G., Nguyen, D., Wu, J., Lucas, D., Ma, D., Isaacs, L., et al. (2010). Toxicology and drug delivery by cucurbit[*n*]uril type molecular containers. *PloS ONE*, *5*, e10514.
Hwang, I., Jeon, W. S., Kim, H. J., Kim, D., Kim, H., Selvapalam, N., et al. (2007). Cucurbituril: A simple macrocyclic, pH-triggered hydrogelator exhibiting guest-induced stimuli-responsive behavior. *Angewandte Chemie International Edition*, *46*, 210–213.
Kim, K. (2002). Mechanically interlocked molecules incorporating cucurbituril and their supramolecular assemblies. *Chemical Society Reviews*, *31*, 96–107.
Kim, C., Agasti, S. S., Zhu, Z., Isaacs, L., & Rotello, V. M. (2010). Recognition-mediated activation of therapeutic gold nanoparticles inside living cells. *Nature Chemistry*, *11*, 962–966.
Kim, D., Kim, E., Kim, J., Park, K. M., Baek, K., Jung, M., et al. (2007). Direct synthesis of polymer nanocapsules with a noncovalently tailorable surface. *Angewandte Chemie International Edition*, *46*, 3471–3474.
Lagona, J., Mukhopadhyay, P., Chakrabarti, S., & Isaacs, L. (2005). The cucurbit[*n*]uril family. *Angewandte Chemie International Edition*, *44*, 4844–4870.
Lehn, J. M. (1988). Supramolecular chemistry-scope and perspectives molecules, supermolecules and molecular devices (Nobel Lecture). *Angewandte Chemie International Edition in English*, *27*, 89–112.
Lin, Y., Chen, Q., & Luo, H. (2007). Preparation and characterization of *N*-(2-carboxybenzyl) chitosan as a potential pH-sensitive hydrogel for drug delivery. *Carbohydrate Research*, *73*, 265–273.
Liu, S. M., Xu, L., Wu, C. T., & Feng, Y. Q. (2004). Preparation and characterization of perhydroxyl-cucurbituril bonded silica stationary phase for hydrophilic-interaction chromatography. *Talanta*, *64*, 929–934.
Rauwald, U., Del Barrio, J., Loh, X. J., & Scherman, O. A. (2011). "On-demand" control of thermoresponsive properties of poly(*N*-isopropylacrylamide) with cucurbituril host–guest complexes. *Chemical Communications*, *47*, 6000–6002.
Ravi Kumar, M. N. V., Muzzarelli, R. A. A., Muzzarelli, C., Sashiwa, H., & Domb, A. J. (2004). Chitosan chemistry and pharmaceutical perspectives. *Chemical Reviews*, *104*, 6017–6084.
Rojanarata, T., Petchsangsai, M., Opanasopit, P., Ngawhirunpat, T., Ruktanonchai, U., Sajomsang, W., et al. (2008). Methylated *N*-(4-*N*,*N*-dimethylaminobenzyl) chitosan for a novel effective gene carriers. *European Journal of Pharmaceutics and Biopharmaceutics*, *70*, 207–214.
Samuels, R. J. (1981). Solid state characterization of the structure of chitosan films. *Journal of Polymer Science: Polymer Physics Edition*, *19*, 1081–1105.
Sangeetha, N. M., & Maitra, U. (2005). Supramolecular gels: Functions and uses. *Chemical Society Reviews*, *34*, 821–836.
Singh, J., & Dutta, P. K. (2009). Preparation, circular dichroism induced helical conformation and optical property of chitosan acid salt complexes for biomedical applications. *International Journal of Biological Macromolecules*, *45*, 384–392.
Steed, J. W. (2011). Supramolecular gel chemistry: Developments over the last decade. *Chemical Communications*, *47*, 1379–1383.
Ta, H. T., Dass, C. R., & Dunstan, D. E. (2008). Injectable chitosan hydrogels for localised cancer therapy. *Journal of Controlled Release*, *126*, 205–216.
Teng, S. H., Lee, E. J., Wang, P., Shin, D. S., & Kim, H. E. (2008). Three-layered membranes of collagen/hydroxyapatite and chitosan for guided bone regeneration. *Journal of Biomedical Materials Research Part B: Applied Biomaterials*, *87*, 132–138.
Tien, C. L., Lacroix, M., Ispas-Szabo, P., & Mateescu, M. A. (2003). *N*-acylated chitosan: Hydrophobic matrices for controlled drug release. *Journal of Controlled Release*, *93*, 1–13.
Uzunova, V. D., Cullinane, C., Brix, K., Nau, W. M., & Day, A. I. (2010). Toxicity of cucurbituril and cucurbituril: An exploratory in vitro and in vivo study. *Organic and Biomolecular Chemistry*, *8*, 2037–2042.
Van De Manakker, F., Vermonden, T., El Morabit, N., Van Nostrum, C. F., & Hennink, W. E. (2008). Rheological behavior of assembling PEG-β-cyclodextrin/PEG-cholesterol hydrogels. *Langmuir*, *24*, 12559–12567.
Wan, Y., Creber, K. A. M., Peppley, B., & Bui, V. T. (2003). Synthesis, characterization and ionic conductive properties of phosphorylated chitosan membranes. *Macromolecular Chemistry and Physics*, *204*, 850–858.
Wang, X. L., Huang, Y., Zhu, J., Pan, Y. B., He, R., & Wang, Y. Z. (2009). Chitosan-graft poly(*p*-dioxanone) copolymers: Preparation, characterization and properties. *Carbohydrate Research*, *344*, 801–807.
Yang, X., & Kim, J. C. (2010). β-Cyclodextrin hydrogels containing naphthaleneacetic acid for pH-sensitive release. *Biotechnology and Bioengineering*, *106*, 295–302.
Yang, H., Tang, Y., & Wang, Y. (2009). Fabrication and properties of cucurbituril induced thermo-responsive supramolecular hydrogels. *Soft Matter*, *5*, 3511–3516.
Yang, Z., Xu, K., Wang, L., Gu, H., Wei, H., Zhang, M., et al. (2005). Self-assembly of small molecules affords multifunctional supramolecular hydrogels for topically treating simulated uranium wounds. *Chemical Communications*, *35*, 4414–4416.
Zhang, G. L., Wu, Z. Y., Li, Y. T., Wang, D. D., & Dou, J. M. (2006). Synthesis and crystal structure of a binuclear gadolinium (III) complex bridged by cucurbituril. *Chinese Chemical Letters*, *17*, 93–96.
Zhou, H. Y., Chen, X. G., Kong, M., Liu, C. S., Cha, D. S., & Kennedy, J. F. (2008). Effect of molecular weight and degree of chitosan deacetylation on the preparation and characteristics of chitosan thermosensitive hydrogel as a delivery system. *Carbohydrate Polymers*, *73*, 265–273.