Carbohydrate Polymers 77 (2009) 583–589

Contents lists available at ScienceDirect

Carbohydrate Polymers

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

Synthesis of thermosensitive P(NIPAAm-*co*-HEMA)/cellulose hydrogels via "click" chemistry

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ARTICLE INFO

Article history:
Received 28 November 2008
Received in revised form 16 January 2009
Accepted 26 January 2009
Available online 6 February 2009

Keywords:
"Click" chemistry
Hydrogel
PNIPAAm
Cellulose
Thermosensitive

ABSTRACT

Azide-modified cellulose and alkyne-modified poly(*N*-isopropylacrylamide-*co*-hydroxylethyl methacrylate) P(NIPAAm-*co*-HEMA) were synthesized. The two components were cross-linked once mixed together in the presence of Cu(I) catalyst, a type of Huisgen's 1,3-dipolar azide-alkyne cycloaddition which is also defined as "click" chemistry, leading to the *in situ* formation of a series of novel thermosensitive P(NIPAAm-*co*-HEMA)/cellulose hydrogels. The gelation process was examined via rheology. The resulted hydrogels was studied via scanning electron microscope (SEM), equilibrium swelling ratio, swelling kinetics and temperature response kinetics. The obtained data presented that the formed hydrogels exhibited favorable thermosensitive properties upon temperature changes.

1. Introduction

Hydrogels are three-dimensional cross-linked networks composed of hydrophilic synthetic or natural polymers, which are able to absorb a lot of water. Hydrogels have been widely used in biomedical fields, including drug delivery systems and tissue engineering scaffolds due to the similarity between the highly hydrated three-dimensional networks and the hydrated body tissues. The intelligent materials, which are constructed for applications in the biomedical field, need to react selectively without interference by biological functionality. Ossipov et al. investigated different crosslinking chemistries, which could afford *in-vivo* hydrogel formation and cell growth, and proved that perfect crosslinking reaction should go fast enough to give a hydrogel within a few minutes without the release of harmful side product and the ligation reaction must be chemoselective and stable toward hydrolysis and oxidation (Ossipov & Hilborn, 2006).

Chemoselective crosslinking reactions initiated by simply mixing polymer component solutions have drawn much research attention. Some kinds of reactions have been applied for hydrogel formation, such as coupling between an aldehyde and a hydrazide group (Bulpitt & Aeschlimann, 1999; Jia, Colombo, Padera, Langer, & Kohane, 2004; Luo, Kirker, & Prestwich, 2000) or a cysteine 1, 2-aminothiol group (Wathier, Jung, Carnahan, Kim, & Grinstaff, 2004). Michael additions of thiols to acrylates (Elbert, Pratt, Lutolf, Halstenberg, & Hubbel, 2001; Vernon, Tirelli, Bächi, Haldimann, & Hubbel, 2003) or vinyl sulfones (Lutolf & Hubbell, 2003; Qiu et al., 2003) were also used for preparation of the hydrogel. All these reactions can be carried out at physiological temperature and pH. A major drawback, however, is that thiol-functionalized compounds are sensitive toward oxidation. There is another biocompatible reaction, called "click" reaction, which has no need to protect from oxygen and only requires stoichiometric amounts of starting materials.

The azide/alkyne "click" reaction, which is also termed as the "Sharpless-type click reaction", is variated from the Huisgen 1,3-dipolar cycloaddition reaction between C≡C, C≡N bonds, and azides (Tornoe, Christensen, & Meldal, 2002). The Cu(I)-catalyzed 1,3-dipolar cycloaddition of azides and alkynes (CuAAC) (Kolb, Finn, & Sharpless, 2001; Wu et al., 2004) has become the typical example of "click" chemistry (Malkoch et al., 2005), with its unique characteristics of quantitative yields without side reactions, high tolerance of functional groups and extraordinary reliability. Such "click" chemistry possesses a great potential in material science, and this reaction has been proven extraordinary useful for the synthesis of dendrimers (Parrish, Breitenkamp, & Emrick, 2005; Tornoe et al., 2002), modification of proteins (Helms, Mynar, Hawker, & Frechet, 2004), and many other novel polymers and materials (Opsteen & van Hest, 2005; Tsarevsky, Sumerlin, & Matyjaszewski, 2005). Furthermore, "click" chemistry have also been applied inside living cells without affecting the cell activity already (Link & Tirrell, 2003; Wang et al., 2003).

Here, we take the advantages of the CuAAC reaction to create three-dimensional networks, without using traditional crosslinking agents. Based on this point, alkyne and azide pendent groups were introduced to thermosensitive poly(*N*-isopropylacrylamide)

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doi:10.1016/j.carbpol.2009.01.023

(PNIPAAm) based polymer and natural polysaccharide, cellulose, respectively. Attributed to the chemoselective click coupling crosslinking reaction between the alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose in the presence of Cu(I) catalyst, *in situ* formed hydrogels could be achieved. The gelation process was examined via rheology. The resulted hydrogels was studied via scanning electron microscope (SEM), equilibrium swelling ratio, swelling kinetics and temperature response kinetics.

2. Experiment

2.1. Materials

The cellulose was purchased from AVICEL PH-101® (FLUKA, DP 260). Lithium Chloride (LiCl) was provided by Shanghai Reagent Chemical Co. (China). The cellulose and LiCl were dried in vacuum at 50 °C for 24 h before use. *N*-Isopropylacrylamide (NIPAAm), 2-bromoisobutyl bromide, copper (I) bromide (CuBr) and *N*,*N*′,*N*′,*N*′-pentamethyldiethylenetriamine (PMDETA) were purchased from Acros and used as received. Hydroxylethyl methacrylate (HEMA), propargyl alcohol, *N*,*N*′-dimethylacetamide (DMAc), triethylamine (TEA), pyridine, *N*,*N*′-dimethylformamide (DMF), tetrahydrofuran (THF), and 1,4-dioxane were obtained from Shanghai Reagent Chemical Co. (China) and distilled prior to use. *N*,*N*′-Azobisisobutyronitrile (AIBN) were provided by Shanghai Reagent Chemical Co. (China) and recrystallized before use. Succinic anhydride, 4-toluene sulfonyl chloride (TsCl) *N*,*N*′-dicyclohexylcarbodimide (DCC), 4-(dimethylamino)pyridine (DMAP), sodium azide (NaN_3) were purchased from Shanghai Reagent Chemical Co. (China) and used as received. All other reagents and solvents were of analytical grade and used without further purification.

2.2. Synthesis of 4-oxo-4-(prop-2-ynyloxy) butanoic acid

4-Oxo-4-(prop-2-ynyloxy) butanoic acid was synthesized according to our previous report (Xu et al., 2008). Briefly, 10 mL (0.085 mol) propargyl alcohol, 10.6 g (0.116 mol) succinic anhydride, 8.6 mL (0.116 mol) pyridine and 18 mL (0.116 mol) triethyl amine were dissolved in 200 mL of dry 1,4-dioxane. The mixture was stirred at room temperature for 24 h. The mixture was evaporated under vacuum and then dissolved in CH_2Cl_2 and washed with chilled 1 M HCl. After extracting three times, the organic phase was dried over Na_2SO_4, filtered, and evaporated to obtain 4-oxo-4-(prop-2-ynyloxy) butanoic acid. Yield: 48%.

^{1}H NMR (300 MHz, $CDCl_3$, δ ppm): 11.2 (s, 1H, —C═OO*H*), 4.7 (d, 2H, CH≡CC*H*$_2$O–), 2.7 (m, 4H, —C═OC*H*$_2$C*H*$_2$C═OOH), 2.5 (t, 1H, C*H*≡CCH$_2$—).

^{13}C NMR (300 MHz, $CDCl_3$, δ ppm): 176.3 (—*C*═OOH), 172.6 (—*C*═OCH$_2$CH$_2$C═OOH), 77.5 (CH≡*C*CH$_2$O—), 75.3 (*C*H≡CCH$_2$O—), 55.4 (CH≡CCH$_2$O—), 31.1 (—C═OCH$_2$*C*H$_2$C═OOH), 28.3 (—C═OCH$_2$ CH$_2$C═OOH).

2.3. Synthesis of P(NIPAAm-co-HEMA)

To a solution of 4.1 g (0.036 mol) NIPAAm and 0.4 g (0.003 mol) HEMA (NIPAAm/HEMA molar ratio = 12/1) in 20 mL DMF, 19 mg (0.116 mmol) AIBN were added, and the reaction mixture was stirred at 70 °C for 24 h under nitrogen atmosphere. After precipitated with chilled ether and filtered, the sample was dried under vacuum to obtain P(NIPAAm-*co*-HEMA). Yield: 71%.

2.4. Synthesis of alkyne-modified P(NIPAAm-co-HEMA)

0.9 g (0.006 mol) 4-oxo-4-(prop-2-ynyloxy) butanoic acid was dissolved in dry THF, then 1.5 g (0.007 mol) DCC was added. After that, the solutions of 3.2 g (0.003 mol) P(NIPAAm-*co*-HEMA) in dry THF and 0.9 g (0.007 mol) DMAP in DMSO were added. The mixture was stirred and reacted at room temperature for 24 h. The insoluble precipitation was filtered. Alkyne-modified P(NIPAAm-*co*-HEMA) was obtained by adding chilled diethyl ether to the filtrate, precipitation and drying in vacuum.

2.5. Synthesis of cellulose p-toluenesulfonates (tosylates)

Tosylated cellulose was synthesized according to the previous report (Ranh, Diamantoglou, Klemm, Berghmans, & Heinze, 1996). After the substitution between cellulose and *p*-toluenesulfonyl chloride, the crude product was collected via the precipitation in ice water. Thereafter, the precipitation was suspended in acetone and re-precipitated into distilled water. After the filtration and washing with ethanol, the purified tosylated cellulose was obtained after dried in vacuum at 50 °C for 24 h with a yield of 75%.

Degree of substitution (DS): 1.46 (calculated from *S*-content determined by elemental analysis).

Elemental analysis (EA): C 48.32, H 4.93, S 11.35%.

2.6. Synthesis of azide-modified cellulose

By using the prepared tosylated cellulose above, the azide-modified cellulose was synthesized according to the literature (Liu & Hsieh, 2006; Matthias, Jens, Frank, & Thomas, 2008; Matthias & Thomas, 2008; Thomas, Michael, Matthias, & Frank, 2008; Tim, Claudia, & Thomas, 2005). After the reaction, the product was precipitated in distilled water. By washing the precipitation with water and ethanol for five times, the sample was dried at 60 °C under vacuum to obtain azide-modified cellulose.

Degree of substitution (DS): 0.95 (calculated from *N*-content determined by elemental analysis).

Elemental analysis (EA): C 37.52, H 4.67, N 21.71%.

2.7. Hydrogel preparation

Alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose with various ratios were dissolved in DMSO. Then, 5 mg CuBr as a catalyst and 50 μL PMDETA as an accelerator were added. The mixture was put still at room temperature for the desire time. According to Table 1, by changing the ratio of azide-modified cellulose and alkyne-modified P(NIPAAm-*co*-HEMA), and the amount of DMSO, a series of thermosensitive P(NIPAAm-*co*-HEMA)/cellulose hydrogels were prepared. The resulting hydrogel was immersed in distilled water for 7 days in order to remove the residues such as catalyst in the formed hydrogel. The distilled water was refreshed every several hours.

2.8. FT-IR measurement

The frozen hydrogel samples were analyzed by FT-IR (Perkin–Elmer Spectrum One) spectrophotometer. Before the measurement, the hydrogel samples were pressed into potassium bromide (KBr) pellet, respectively.

Table 1
The feed compositions of P(NIPAAm-*co*-HEMA)/cellulose hydrogels.

	Gel1	Gel2	Gel3	Gel4
Alkyne-modified P(NIPAAm-*co*-HEMA) (mg)	50	100	150	200
Azide-modified cellulose (mg)	50	50	50	50
CuBr (mg)	5	5	5	5
PMDETA (μL)	50	50	50	50
DMSO (mL)	1	1.5	2	2.5

2.9. ^{1}H NMR and ^{13}C NMR measurements

The ^{1}H NMR spectra of the polymers were recorded on a Mercury VX-300 spectrometer at 300 MHz (Varian, USA) by using $CDCl_3$ as a solvent and TMS as an internal standard.

The ^{13}C NMR spectra of the polymers were recorded on a Mercury VX-300 spectrometer at 300 MHz (Varian) by using DMSO as a solvent and TMS as an internal standard.

2.10. Oscillatory rheology

Rheology experiments were performed at room temperature on ARES-RFS III rheometer (TA Instruments, USA). Alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose solutions (4 mL, 0.1 g/mL) were prepared. After mixing these two types of solutions, PMDETA, CuBr were added. Subsequently, the mixed solution was quickly transferred to the rheometer within several minutes. The storage modulus (G′) and loss modulus (G″) were recorded.

2.11. Interior morphology

Scanning electron micrograph (SEM) was taken with a Hitachi X-650 microscope. The hydrogel, equilibrated in distilled water at 22 °C, was frozen in liquid nitrogen immediately, and then freeze-dried under vacuum at −45 °C for 3 days. The facture surface of the hydrogel was observed after sputtering with gold for 7 min.

2.12. Temperature dependence of swelling ratios

The temperature dependence of swelling ratio of P(NIPAAm-*co*-HEMA)/cellulose hydrogels was examined to evaluate their temperature-sensitive properties. Swelling ratio of the hydrogels was measured gravimetrically in the temperature range from 15 to 45 °C, after the sample surfaces had been wiped with moistened filter paper. The hydrogel samples were incubated in distilled water for at least 24 h at each temperature. The swelling ratio (SR) of hydrogel is defined as follows:

$SR = W_s/W_d$where W_s is the weight of water in a swollen hydrogel at each temperature and W_d is the dried weight of the hydrogel after vacuum drying.

2.13. Deswelling kinetics at 50 °C

The deswelling kinetics of the equilibrated swollen hydrogels was measured gravimetrically in distilled water at 50 °C. At predetermined time intervals, the samples were taken out from the hot water (50 °C) and weighed after the sample surfaces were wiped with moistened filter paper to remove the excess water. The average value of three measurements was used. Water retention was defined as follows:

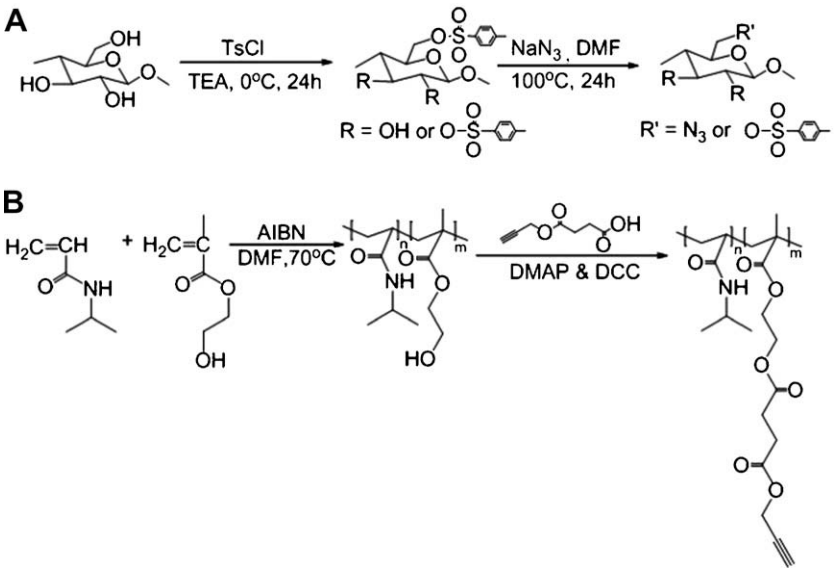
$[\text{Water retention}]_t = [(W_t - W_d)/W_s] \times 100$where W_t is the weight of the equilibrated sample at time t at 50 °C, W_s is the equilibrium water weight at room temperature, and the other symbols are the same as defined above.

2.14. Reswelling kinetics at 15 °C

The vacuum dried samples were immersed in distilled water at 15 °C and taken out from water to be weighed after removing the water on the surface with moistened filter paper at regular time intervals. Each sample was measured three times and the average value was used. The swelling ratio during the reswelling course of hydrogel is defined as defined above.

3. Results and discussion

"Click" chemistry is an efficient strategy for effective controlling the crosslinking during the preparation of hydrogels. Alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose were synthesized to investigate the possibility of *in situ* hydrogel formation by "click" chemistry. Moreover, we established a new method to prepare hydrogel from unsubstituted cellulose, which have been scarcely reported because of the insolubility of cellulose in aqueous solutions due to its strong intermolecular and intramolecular hydrogen bondings (Zhou, Chang, Zhang, & Zhang, 2007). The synthesis routes of the two polymers were presented in Scheme 1. To prepare the alkyne-modified P(NIPAAm-*co*-HEMA) and



Scheme 1. The synthesis of the azide-modified cellulose (A) and alkyne-modified P(NIPAAm-*co*-HEMA) (B).

azide-modified cellulose, the precursors, P(NIPAAm-*co*-HEMA) and cellulose tosylate were synthesized. Then alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose were fabricated via coupling and nucleophilic displacement reactions, respectively. From the FT-IR spectra (Fig. 1A), the typical amide I and II bands of NIPAAm units were obvious at 1641 and 1540 cm^{-1}. Besides, the stretching variation absorbance of C=O in HEMA units existed at 1725 cm^{-1}. The real molar ratio of NIPAAm units to HEMA units in P(NIPAAm-*co*-HEMA) was 12.2: 1, calculated from its ^1H NMR spectrum (Fig. 2A). To demonstrate the existence of alkyne pendant group in the corresponding polymer, FT-IR (Fig. 1B) and ^1H NMR (Fig. 2B) characterizations were carried out. From the FT-IR spectrum of the alkyne-modified P(NIPAAm-*co*-HEMA), the typical absorbent peak of the alkyne pendant group existed at 2125 cm^{-1}. Based on the ^1H NMR spectra, after the coupling reaction between P(NIPAAm-*co*-HEMA) and 4-oxo-4-(prop-2-ynyloxy) butanoic acid, the original chemical shift at 3.8 ppm belonging to the hydroxyl groups of HEMA units disappeared in the ^1H NMR spectra of the alkyne-modified P(NIPAAm-*co*-HEMA) as shown in Fig. 2B, indicating that nearly all the hydroxyl groups have reacted and transformed to the alkyne pendant groups. Moreover, the chemical shift at 4.7 ppm was attributed to the methylene protons of $CH{\equiv}CCH_2O{-}$. And the chemical shifts at 2.5 and 2.7 ppm were mainly associated with the protons of alkyne pendant groups ($CH{\equiv}CCH_2{-}$) and methylene groups ($-C{=}OCH_2CH_2O{=}C-$), respectively.

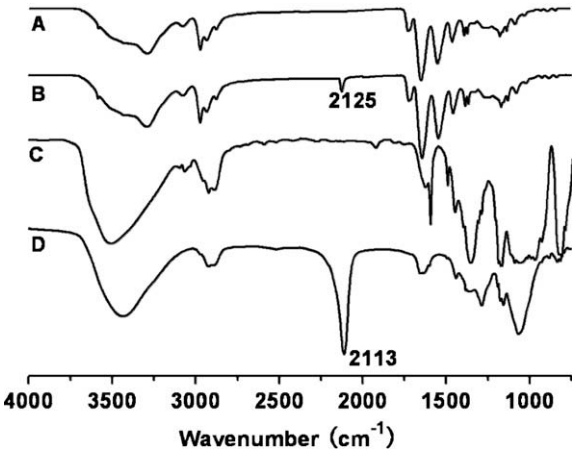


Fig. 1. FT-IR spectra of the P(NIPAAm-*co*-HEMA) (A), alkyne-modified P(NIPAAm-*co*-HEMA) (B), tosylated cellulose (C) and azide-modified cellulose (D).

Cellulose derivatives were synthesized and purified according to the previous reports (Liu & Hsieh, 2006; Matthias & Thomas, 2008; Matthias et al., 2008; Ranh et al., 1996; Thomas et al., 2008). FT-IR spectrum of cellulose tosylate (Fig. 1C) showed the typical absorptions of the cellulose backbone as well as signals of the aromatics at 3067, 1598, 1495 and 815 cm^{-1}, respectively. Furthermore, two bands with high intensity at 1359 and 1175 cm^{-1} (υSO_2) confirmed the presence of the tosyl groups (DS = 1.46). Fig. 2C showed a standard ^{13}C NMR spectrum of cellulose tosylate. The signals of the tosyl methyl and aromatic ring carbons were detectable at 21.7 and 128.3–148.2 ppm, respectively. And resonances assigned to the carbons of cellulose backbone were visible in the region of 63.9 to 104.7 ppm. In this spectrum, a small signal for tosylated C-6 atoms (δ = 67.5 ppm) as well as the one for non-tosylated C-6 atoms (δ = 63.9 ppm) occurred. C-1 atoms gave two signals at δ = 97.5 (C-1′) and δ = 102.7 (C-1) ppm in its spectral region. The splitting pattern of the peak of C-1 (C-1′) indicated an incomplete tosylation at C-2, from which a faster esterification of O-6 than O-2 and O-3 can be estimated.

As for the cellulose tosylate, it was simply transformed to azide-modified cellulose by a homogeneous nucleophilic displacement reaction in DMF with sodium azide. The FT-IR spectrum of azide-modified cellulose (Fig. 1D) showed a significant band at 2113 cm^{-1} for the azide moiety. But one can also find the weak aromatic bands of the tosyl groups at 1598, 1500, and 1453 cm^{-1}, because only the primary tosylate moieties which positioned on C-6 could be easily substituted by NaN_3, which is demonstrated in previous report (Ranh et al., 1996). And the degree of substitution is 0.95, calculated based on the element analysis. The ^{13}C

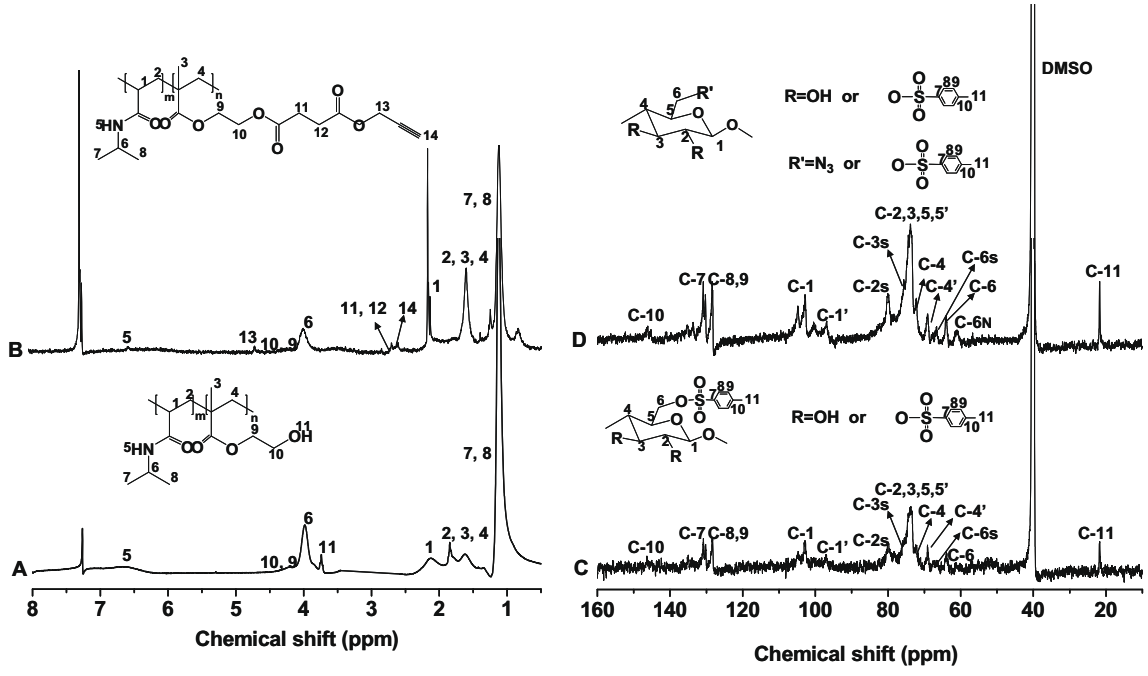


Fig. 2. ^1H NMR spectra the P(NIPAAm-*co*-HEMA) (A), alkyne-modified P(NIPAAm-*co*-HEMA) (B) and ^{13}C NMR spectra of the tosylated cellulose (C), azide-modified cellulose (D).

NMR spectrum of azide-modified cellulose was similar to the cellulose tosylate one, except that the signal for C-6 atoms influenced by azide groups appeared at 61.0 ppm while that of the corresponding carbon of tosylated cellulose appeared at 66.6 ppm.

Through "click" chemistry, the alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose reacted in the presence of CuBr/PMDETA in DMSO at room temperature. Table 1 displayed the feed compositions of four kinds of P(NIPAAm-*co*-HEMA)/cellulose hydrogels, which were designated as Gel1, Gel2, Gel3 and Gel4 correspondingly. The molar ratio of alkyne-modified P(NIPAAm-*co*-HEMA) and azide-modified cellulose increased from Gel1 to Gel4. All of the gelations were initiated quickly and the yield of the gel formed was almost 100%.

Fig. 3 exhibited the oscillatory rheology of the formed hydrogels and FT-IR spectra of four hydrogels. The rheology measurement was performed to examine the detailed gelation process. From the rheology behavior of the hydrogel, the storage modulus (G′) was higher than the loss modulus (G″) within 10 min, which was the characteristic rheological behavior of solid like gel materials (Fig. 3A). This could confirm the expected gelation of two polymers in the presence of catalyst Cu(I). Also from the IR spectrum, it can be found that the typical peak of alkyne groups at around 2125 cm^{-1} disappeared and the typical peak of azide groups at around 2113 $cm^{-1}$was weakened compared to the IR spectrum of the alkyne-modified P(NIPAAm-*co*-HEMA) (Fig. 1B) and azide-modified cellulose (Fig. 1D) before gelation. As the IR spectrum of

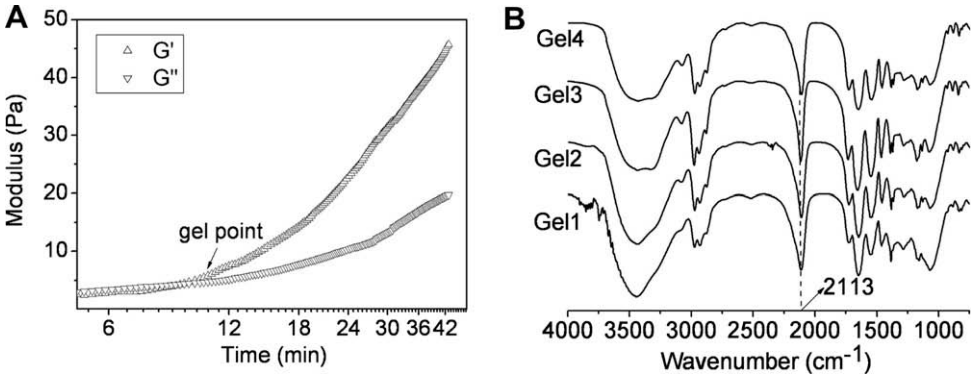


Fig. 3. Oscillatory rheology of the P(NIPAAm-*co*-HEMA)/cellulose based hydrogels (A) and the FT-IR of P(NIPAAm-*co*-HEMA)/cellulose hydrogels (B).

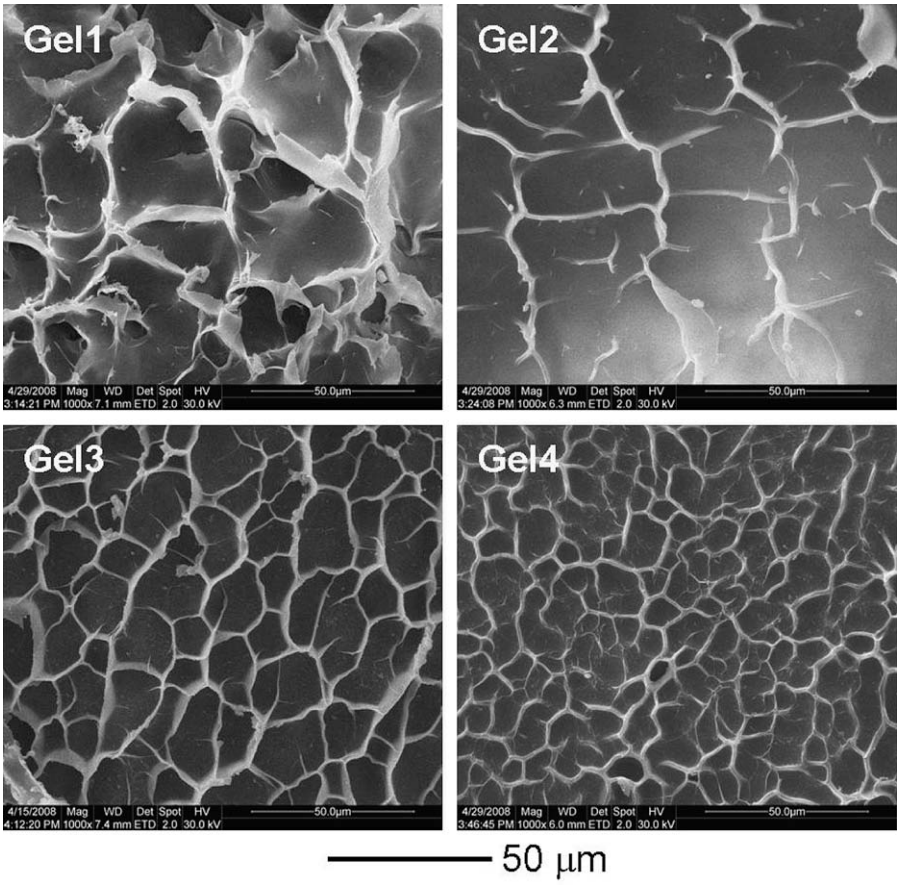


Fig. 4. The SEM images of P(NIPAAm-*co*-HEMA)/cellulose hydrogels.

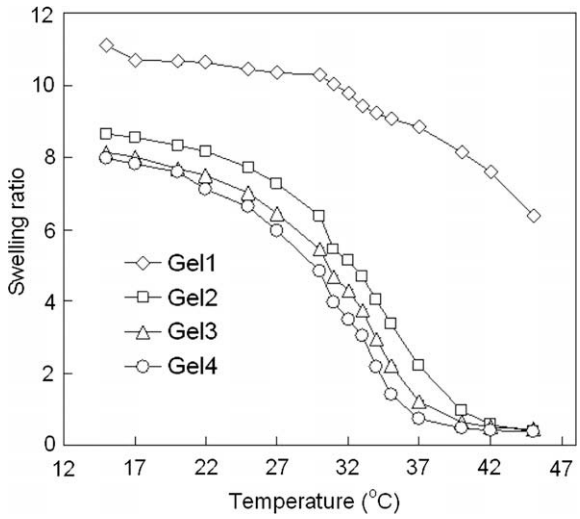


Fig. 5. The temperature dependence of swelling ratio of P(NIPAAm-*co*-HEMA)/cellulose hydrogels.

azide-modified cellulose displayed, the absorption peak at 2113 cm^{-1} for the azide moiety was much stronger than that of the cellulose backbone at 3067 cm^{-1} (Fig. 1D) while the intensity of absorption at 2113 cm^{-1} became weaker than that at 3067 cm^{-1} after gelation (Fig. 3B), which indirectly indicated that the two polymers have reacted already and azide groups were excessive. This is consistent with the structure of the two components as discussed above. Since the real molar ratio of NIPAAm units to HEMA units in P(NIPAAm-*co*-HEMA) was 12.2: 1, and all the hydroxyl groups have completely reacted and transformed to the alkyne pendant groups, it can be inferred that the amount of triple bonds in the synthetic copolymer is 8.2%. Compared with the degree of substitution of the cellulose derivatives, we can conclude that azide groups were of high excessiveness in the "click" reaction, which can ensure a thorough reaction.

Fig. 4 presented the SEM images of the P(NIPAAm-*co*-HEMA)/cellulose hydrogels. All of the hydrogels displayed discontinuous porous inner structure. The pore sizes were between 10 and 40 μm. It is known that the reaction junctions increased with the increasing alkynyl groups content from Gel1 to Gel4. Thus, the crosslinking levels of the hydrogels improved correspondingly, leading to the decreasing average pore sizes of hydrogels from Gel1 to Gel4 as shown in Fig. 4.

The temperature dependence of swelling ratio is one of the most important parameters to evaluate the temperature-sensitive properties of hydrogels. As exhibited in Fig. 5, all the P(NIPAAm-*co*-HEMA)/cellulose hydrogels demonstrated similar thermoresponsive profiles. Below the LCST, all the hydrogels exhibited swollen states with high swelling ratios. As the external temperature increased, the swelling ratios of hydrogels decreased and reached the lowest value at 45 °C (above LCST). Moreover, the swelling ratios of Gel1–4 at 22 °C were 10.6, 8.1, 7.5 and 7.1, respectively. This tendency was attributed to the difference in interior morphology of the hydrogels. As mentioned earlier, from Gel1 to 4, the average pore sizes decreased. Therefore, the corresponding water holding capacities of the hydrogels decreased, resulting in the decreasing swelling ratios from Gel1 to Gel4.

The temperature dependence of the swelling ratio only demonstrates the equilibrium hydration states of hydrogels at different temperatures. In practical applications, the temperature response kinetics or deswelling kinetics upon the suddenly altered temperature are critical important. The deswelling kinetics of four hydrogels were carried out by transferring the equilibrated swollen samples at 22 °C (below LCST) to hot water at 50 °C (above LCST) and the results were illustrated in Fig. 6A. It was found that all the hydrogels tended to shrink and lost water once immersed into hot water at 50 °C. The shrinking rate of the hydrogels was as follows: Gel2 > Gel3 > Gel4 > Gel1. Being consistent with Fig. 5, Gel1 shrunk much slower than the other three hydrogels for its unfavorable thermosensitivity. From Gel2 to Gel4, the shrinking rates got decreased. The reason was mainly attributed to the decreasing average pore sizes of the hydrogels. Because the macroporous structure facilitated the diffusion of water molecules out of the hydrogel matrix. For instance, the water retention of Gel1 decreased from 100% to 75% within 90 min, while that of Gel2, Gel3 and Gel4 decreased from 100% to 15%, 17% and 31% within the same time frame, respectively.

Fig. 6B demonstrated the reswelling behaviors of the P(NIPAAm-*co*-HEMA)/cellulose hydrogels. From the figure, it can be seen that the reswelling rates of four hydrogels improved from Gel1 to Gel4. Generally, the reswelling procedure is related to the hydrophilicity of the hydrogel. Since P(NIPAAm-*co*-HEMA) chain was water-soluble at 15 °C, with the increasing content of P(NIPAAm-*co*-HEMA) from Gel1 to Gel4, the hydrophilicity of related hydrogel was enhanced, resulting in the improving reswelling rates of hydrogels from Gel1 to Gel4.

4. Conclusions

In this study, azide-modified cellulose and the alkyne-modified P(NIPAAm-*co*-HEMA) were synthesized. Then a series of novel hydrogels were prepared by mixing the solution of two components functionlized with alkyne and aizde groups through

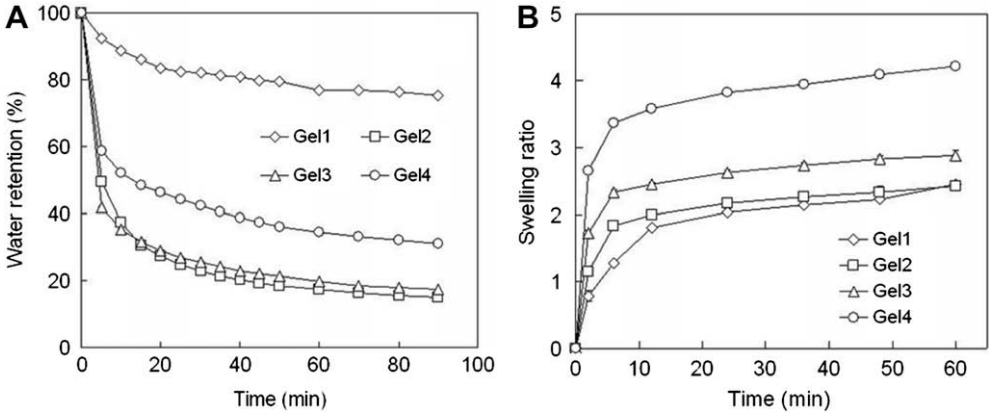


Fig. 6. The deswelling kinetic (A) and reswelling kinetic (B) of P(NIPAAm-*co*-HEMA)/cellulose hydrogels.

Huisgen's 1,3-dipolar cycloaddition with Cu(I)-catalyzed, which is also defined as "click" chemistry. The resulted hydrogels exhibited a porous network structure and favorable thermosensitivity in terms of temperature dependence of swelling ratio, deswelling kinetic and reswelling kinetic. The strategy described here provides a great potential for the *in situ* formation of hydrogels from natural polysaccharides and synthesized intelligent materials.

Acknowledgements

This work was financially supported by National Natural Science Foundation of China (20774069, 50633020) and Ministry of Education of China (Cultivation Fund of Key Scientific and Technical Innovation Project 707043).

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