Reducibly Degradable Hydrogels of PNIPAM and PDMAEMA: Synthesis, Stimulus-Response and Drug Release

BO-YU ZHANG, WEI-DONG HE, LI-YING LI, XIAO-LI SUN, WEN-TAO LI, KE-REN ZHANG

Department of Polymer Science and Engineering, CAS Key Laboratory of Soft Matter Chemistry, University of Science and Technology of China, Hefei, Anhui 230026, China

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ABSTRACT: Reducibly degradable hydrogels of poly(*N*-isopropylacrylamide) (PNIPAM) and poly(*N*,*N*-dimethylaminoethyl methacrylate) (PDMAEMA) were synthesized by the combination of reversible addition-fragmentation chain transfer (RAFT) polymerization and click chemistry. The alkyne-pending copolymer of PNIPAM or PDMAEMA was obtained through RAFT copolymerization of propargyl acrylate with NIPAM or DMAEMA. Bis-2-azidyl-isobutyrylamide of cystamine (AIBCy) was used as the crosslinking reagent to prepare reducibly degradable hydrogels by click chemistry. The hydrogels exhibited temperature or pH stimulus-responsive behavior in water, with rapid response, high swelling ratio, and reproducible swelling/shrinkage cycles. The loading and release of ceftriaxone sodium proved the feasibility of the hydrogels as the stimulus-responsive drug delivery system. Furthermore, the presence of disulfide linkage in AIBCy favored the degradation of hydrogels in the reductive environment. © 2010 Wiley Periodicals, Inc. J Polym Sci Part A: Polym Chem 48: 3604–3612, 2010

KEYWORDS: drug delivery systems; hydrogels; living radical polymerization (LRP); reducible degradation; stimuli-sensitive polymers; stimulus-response

INTRODUCTION Polymer-based intelligent hydrogels, which could change their volume and porous structure under the external stimulus, such as temperature, pH value, light, electric field, and chemical environment, have attracted great interest in material science, polymer science, and biomedicine.[1–7] More and more intelligent hydrogels are leading potential applications in biology and medicine fields, including gene and drug delivery system,[8] biochemical sensor,[9] bioengineering,[10] biocatalyst,[11] and size-dependent separation.[12]

Among intelligent hydrogels, poly(*N*-isopropylacrylamide) (PNIPAM) and poly(*N*,*N*-dimethylaminoethyl methacrylate) (PDMAEMA) based hydrogels are the most studied. Now the research of PNIPAM based thermoresponsive hydrogels is focused on biomedical field, such as the site-specific conjugation of vinyl sulfone-PNIPAM to N49C streptavidin,[13] the release of sodium salicylate from graft-type PNIPAM hydrogels[14] and the utility of phase-separated PNIPAM as a pseudostationary phase for capillary electrochromatography to analyze the mutation of DNA.[15] The hydrogels based on PDMAEMA exhibit pH responsive behavior due to the protonation of the tertiary amine groups,[16] and there are many reports on their synthesis and responsive behavior.[17,18] Because PDMAEMA is a polycation, researchers also have studied its application in gene delivery.[19] Thus, preparing PNIPAM or PDMAEMA hydrogels through easy ways and applying them in biology field is a hot topic in recent years.

Recently, many research works are concerned with the development of crosslinkers with degradable linkage to control the hydrogel biodegradability.[20,21] Various decomposable crosslinkers, such as ester,[22] peptide[23] and anhydride,[24] are widely used in the synthesis of hydrogels. The resultant crosslinked hydrogels are degradable to water-soluble polymers. Disulfides represent an excellent choice for a degradable crosslinker.[25,26] As the disulfide can be cleaved by reducing reagent such as 1,4-dithiothreitol (DTT),[27] the hydrogels prepared with disulfide crosslinker are reductively decomposed. Disulfide-stabilized poly(methacrylic acid) hydrogels was proposed as functional enzymatic cargo[28] and biochemical degradable gelators with disulfide group have been synthesized by Armes et al.[29] These gels show the potential used in biology field.

At present, click chemistry has gained much attention due to the high efficiency and quantitative yield. Recent reports about click chemistry indicate that it is an ideal method for the synthesis of functional polymers.[30,31] The combination of click chemistry with living polymerization makes it easy to prepare hydrogels with predetermined crosslinkage density. Thus, the design of novel disulfide crosslinker suitable for click reaction could provide a new method for preparing reductively degradable hydrogels.

In general, the response rate of polymer hydrogels is governed by the thickness of polymer matrix skin layer.[32] The

Correspondence to: W.-D. He (E-mail: wdhe@ustc.edu.cn)
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dense skin layer retards the outward diffusion of water during the hydrogel collapse and decreases the swelling/shrinkage rate of hydrogels.[33] Usually, macroporous structure, which is obtained by the addition of pore-forming reagents, could increase the response rate of polymeric hydrogel.[34] For example, Zhang et al. obtained PNIPAM macroporous gel with poly(ethylene glycol) as the pore-forming reagent and the gel exhibited the improved swelling rate and rapid shrinkage rate.[35]

This study aims to prepare intelligent hydrogels with rapid response, adjustable drug release, and reducible degradation by the combination of RAFT polymerization and click chemistry. The resultant hydrogels possess disulfide linkage, so they could degrade into water soluble polymer in the present of DTT. Also, these hydrogels have macroporous structure, which offers them with rapid response. Release of a hydrophilic drug from the hydrogels demonstrates the stimulus dependence and the potential in medical field.

EXPERIMENTAL

Materials

2,2′-Bipyridine, 2-bromoisobutyryl bromide and 1,4-dithiothreitol (DTT, 99%) were purchased from Alfa Aesar and used as received. Acryloyl chloride was purified by vacuum distillation, and azodiisobutyronitrile (AIBN) was purified by recrystallization from ethanol. *N*-Isopropylacrylamide (NIPAM, Kohjin, Japan) was purified by recrystallization from a benzene/*n*-hexane mixture (65/35 v/v). *N*,*N*-Dimethylaminoethyl methacrylate (DMAEMA, Aldrich) was passed through basic alumina column, then vacuum-distilled over CaH_2 before use. Cystamine dihydrochloride (Shanghai Chemical Reagent, 98.5%) was purified by recrystallization from a water/ethanol mixture (25/75 v/v). Triethylamine was stirred with KOH for 12 h at room temperature, refluxed with toluene-4-sulfonyl chloride and distilled before use. Copper(I) bromide was washed with glacial acetic acid, followed by washing with methanol and ethyl ether to remove impurities, then dried under vacuum and kept under N_2 atmosphere. THF was dried over sodium/benzophenone and distilled just before use. Dibenzyltrithiocarbonate (DBTTC) as chain transfer agent (CTA) was synthesized based on literature.[36] All other reagents were of analytical grade and used as received.

Synthesis of Bis-2-azidyl-isobutyrylamide of Cystamine (AIBCy)

A solution of cystamine dihydrochloride (2.85 g, 0.013 mol) in water (100 mL) was cooled to 0–5 °C in ice-water bath. Then, the solution of 2-bromoisobutyryl bromide (6.00 g, 0.015 mol) in dichloromethane (10 mL) and NaOH aqueous solution (2.00 g, 0.050 mol, 10 mL) were added simultaneously from two separated addition funnels in dropwise way. The addition lasted for more than 1 h, and then the mixture was stirred at room temperature for 2 h. After that, the mixture was extracted with 5 × 30 mL methylene chloride. The organic phase was collected and dried over anhydrous sodium sulfate. After the rotary evaporation and further vacuum distillation, bisbromoisobutyrylamide of cystamine was obtained (5.08 g, 89%).

Bis-bromoisobutyrylamide of cystamine (2.25 g, 5 mmol) was added to the solution of sodium azide (0.65 g, 0.15 mol) in DMF (30 mL) at room temperature. The reaction mixture was stirred at 75 °C for 48 h. After reaction, the resulting solution was precipitated in 250 mL water. AIBCy was collected by filtration and dried at room temperature under vacuum for 24 h. (1.80 g, 96%)

RAFT Copolymerization of NIPAM or DMAEMA with Propargyl Acrylate

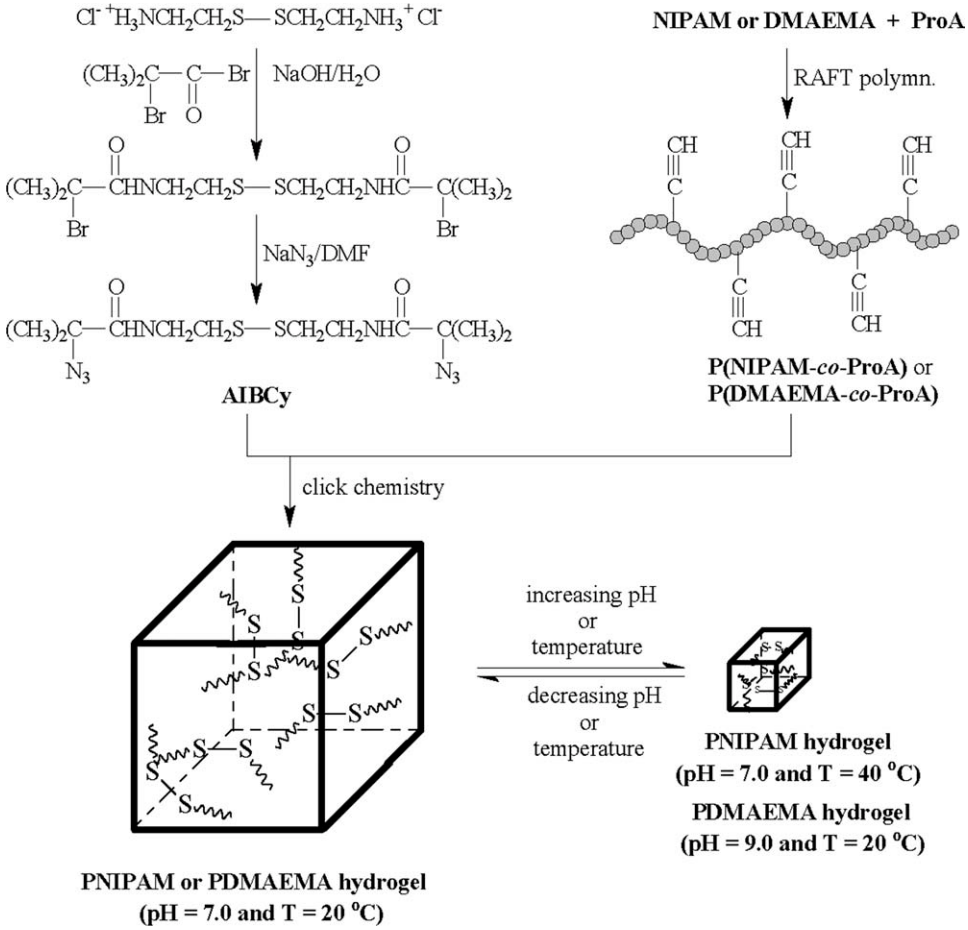
After the solution of propargyl alcohol (1.10 g, 0.019 mol), triethylamine (2.25 g, 0.020 mol), hydroquinone (0.050 g) and THF (100 mL) was cooled in ice-water bath, acryloyl chloride (1.80 g, 0.020 mol) was added dropwise in 20 min. The mixture was stirred in ice-water bath for 1 h and at room temperature for 15 h. After the filtration to remove the solid salt, the solvent was removed by rotary evaporation. Propargyl acrylate (ProA) was obtained by further vacuum distillation (1.8 g, 88%).

Proton nuclear magnetic resonance spectroscopy (^{1}H NMR, $CDCl_3$, δ, ppm): 2.45 (s, 1H, —C≡C***H***), 4.63 (s, 2H, —OC***H***$_2$—C≡CH), 5.85 (d, 1H, C***H***$_2$=CH—), 6.10 (q, 1H, C***H***$_2$=CH—), 6.34 (d, 1H, CH_2=C***H***—).

Copolymer with low ProA content was synthesized via RAFT copolymerization of NIPAM or DMAEMA with ProA using DBTTC as CTA.[37,38] The typical procedure was followed. DMAEMA (1.57 g, 10 mmol), AIBN (1.64 mg, 0.01 mmol), ProA (0.11 g 1 mmol) and DBTTC (0.029 g, 0.1 mmol) were dissolved in THF (3.0 mL). The obtained solution was added into a polymerization tube equipped with a magnetic stirring bar. The mixture was degassed by three freeze-pump-thaw cycles. After the tube was sealed under vacuum, the polymerization reaction proceeded under stirring at 80 °C for 24 h. The tube was opened and the reaction mixture was precipitated into hexane. Poly(DMAEMA-*co*-ProA) (1.37 g, 87%) was obtained after the dryness under vacuum at 25 °C for 24 h. For the preparation of poly(NIPAM-*co*-ProA) (yield: 89%), diethyl ether was used as the precipitation medium. The chemical structure of copolymers was determined by ^{1}H NMR spectroscopy and the relative molecular weight was characterized by gel permeation chromatograph (GPC).

Synthesis of Reducible Intelligent Hydrogels with Disulfide Linkage

PNIPAN or PDMAEMA hydrogels were synthesized through click reaction between alkyne-pending copolymer and azide-containing AIBCy crosslinker. The typical procedure was followed. P($NIPAM_{100}$-*co*-$ProA_{10}$) (600 mg, the digits in subscript are the number of monomer units), AIBCy (85 mg), CuBr (77 mg) and 2,2′-bipyridine (0.25 g) were dissolved in THF (3.0 mL). The mixture was deoxygenated by three freeze-pump-thaw cycles. After the reaction at 30 °C without stirring for 24 h, the resulted gel was purified by dialysis against THF/water (1/1) for 2 d and water for 3 d. The final



SCHEME 1 Schematic illustration of the synthesis of reducible intelligent polymer hydrogel.

dry gel was obtained by the lyophilization in a freeze drier (SIMENS FD5-3) for 48 h. (0.62 g, 95%)

Characterizations

^{1}H NMR spectroscopy (300 MHz) measurements were performed on Bruker DMX300 spectrometer in $CDCl_3$ using tetramethylsilane as internal reference. Fourier transform infrared (FTIR) spectra were recorded on MAGNA-IR 750 spectrophotometer in KBr pellets. The relative molecular weight and its distribution were determined on a Waters 150C GPC equipped with Waters 1515 HPLC pump, Waters 2414 differentia refractive index detector and three Ultrastyragel columns (500, 10^3 and 10^4 Å in series) at 30 °C, using monodispersed polystyrene as calibration standard. DMF was used as eluent at a flow rate of 1.0 mL/min. The final freeze-dried gels were fractured carefully and fixed on aluminum stubs. The interior morphology of the gels was observed under scanning electron microscope (SEM, FEI-SIRION 200). Before SEM observation, the hydrogel specimens were coated with gold.

The Swelling Kinetic of Intelligent Hydrogels

For the investigation of the swelling kinetic of PDMAEMA gels at different pH values (pH = 4.0, 7.0, and 9.0), dry PDMAEMA gels (diameter: 2 cm, thickness: 0.5 cm) were immersed into buffer solutions at 25 °C. At the different determined intervals, the swollen hydrogels were taken out and weighted after the excess water on the surface was wiped off with filter paper. The results of three measurements were averaged. Swelling ratio (R_s) was calculated by eq 1.

$$R_s = \frac{W_s - W_d}{W_d} \times 100\% \quad (1)$$

where W_s and W_d is the weight of swollen gel and dry gel, respectively.

Swelling kinetics of PNIPAM hydrogel at different temperatures was studied in distilled water with similar procedure to that of PDMAEMA hydrogels.

Stimulus Response of Hydrogels with pH or Temperature

After PDMAEMA hydrogels reached the swelling equilibrium in buffer solution (pH = 7.0) at 25 °C, they were quickly transferred to the buffer solution with pH of 9.0. The samples were withdrawn at the determined intervals and weighted after wiping off the excess water on the surface. Thus, the shrinkage kinetics of PDMAEMA hydrogels with pH increase was observed. After that, the swelling-shrinkage cycles between pH = 7.0 and 9.0 were repeated at 25 °C by keeping PDMAEMA hydrogels in two buffer solutions alternatively.

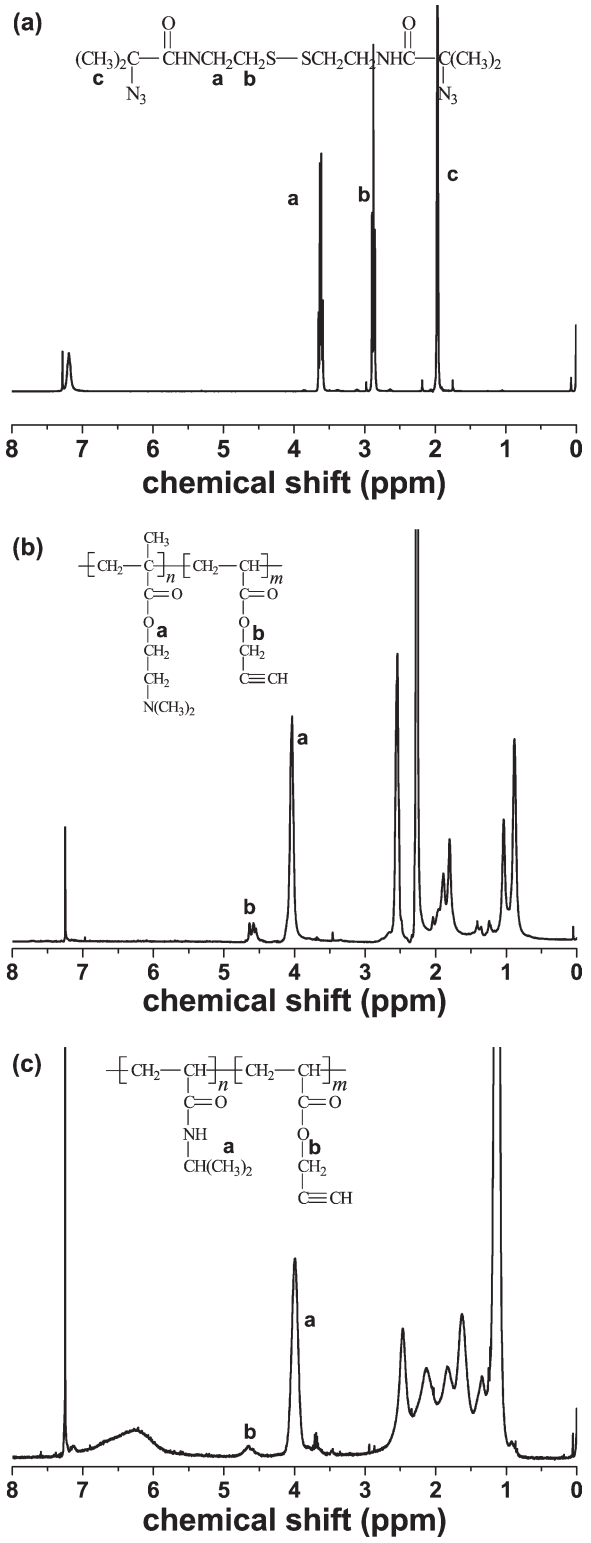


FIGURE 1 ^{1}H NMR spectrum of AIBCy (a), P(DMAEMA-*co*-ProA) (b), and P(NIPAM-*co*-ProA) (c) in $CDCl_3$.

For the study of shrinkage of PNIAPM hydrogels, they were firstly immersed in distilled water at 20 °C. After they reached the swelling equilibrium, the swollen PNIPAM hydrogels were then quickly transferred to distilled water at 40 °C. The swelling-shrinkage cycles of PNIPAM hydrogels between 20 and 40 °C were carried out.

Drug Loading and Release Behavior of Intelligent Hydrogels

Ceftriaxone sodium was chosen as the model drug to test the drug loading and release behaviors of hydrogels. The dry PNIPAM or PDMAEMA hydrogels (50 mg) was swollen in ceftriaxone sodium aqueous solution (10.0 mg/g, 10 g) at 20 °C for 12 h. After the excess water on the surface was wiped off with filter paper, the drug-swollen hydrogel was weighted. The loading amount (mg) of ceftriaxone sodium for 50 mg hydrogels was determined.

The drug release was carried out at different pHs or temperatures. For PDMAEMA hydrogels, the drug-loaded hydrogels were kept in buffer solution (100 mL, pH = 7.0 or 9.0). For PNIPAM hydrogels, the drug-loaded hydrogel was kept in distilled water at the temperature of 20 °C or 40 °C. At each of determined time intervals, 2 mL solution was taken out for UV-vis (Hitachi U-3010) measurement (λ = 272 nm) and its concentration of ceftriaxone sodium was determined using the calibration built on the absorbance of ceftriaxone sodium solutions with known concentrations. At each sampling, 2 mL fresh solution was added to keep the constant volume of medium.

Reducible Degradation of Intelligent Hydrogels

The reducible degradation of hydrogels was studied by using DTT to cleave disulfide linkage of hydrogels. For example, PDMAEMA hydrogels were immersed in DTT aqueous solution (0.1 *M*) and kept under nitrogen atmosphere for 5 d. The gel-to-sol transition was recorded by a digital camera.

RESULTS AND DISCUSSION

Herein, we reported one method to prepare reducible hydrogels. We synthesized P(NIPAM-*co*-ProA) or P(DMAEMA-*co*-ProA) by RAFT copolymerization, and obtained polymer

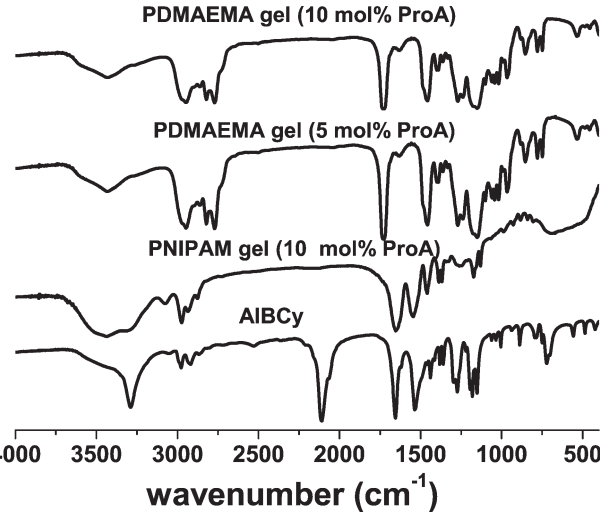


FIGURE 2 FTIR spectrum of AIBCy, PNIPAM gel with 10 mol % ProA, PDMAEMA gel with 5 and 10 mol % ProA.

TABLE 1 Synthesis of the Copolymers[a] and the Reducible Gels[b]

				GPC Results		Gel Preparation	
Copolymer Code	Main Monomer	ProA (mmol)	Molar Ratio in Copolymer[c]	M_n	M_w/M_n	AIBCy (mg)	Geltation
P(DMAEMA-*co*-ProA)-1	DMAEMA	1.0	Mm:ProA = 10	16,700	1.32	72	Yes
P(DMAEMA-*co*-ProA)-2	DMAEMA	0.5	Mm:ProA = 20	15,900	1.37	36	Yes
P(NIPAM-*co*-ProA)-1	NIPAM	1.0	Mm:ProA = 10	12,100	1.23	85	Yes
P(NIPAM-*co*-ProA)-2	NIPAM	0.5	Mm:ProA = 20	11,700	1.29	43	No

[a] RAFT copolymerizations were carried out with main monomer (10.0 mmol) in the presence of DBTTC (0.1 mmol) and AIBN (0.01 mmol) in THF (3.0 mL) at 80 °C for 24 h.
[b] The click crosslinkage reaction of copolymer (600 mg) and AIBCy was performed in the presence of CuBr (77 mg) and bipyridine (0.25 g) in THF (3.0 mL) at 30 °C without stirring for 24 h.
[c] The copolymer composition was determined by ^{1}H NMR. Mm stands for the main monomer, which is DMAEMA and NIPAM respectively.

hydrogels through click chemistry with disulfide-containing crosslinker. The overall procedure was illustrated in Scheme 1.

Synthesis of Reducible Intelligent PNIPAM or PDMAEMA Hydrogels

The disulfide-diazide crosslinker of AIBCy was synthesized through two steps. Firstly, the amino groups of cystamine react with bromoisobutyryl bromide to introduce bromide groups. Secondly, the halogen groups are transformed into azide groups. Figure 1(a) shows ^{1}H spectrum of AIBCy in $CDCl_3$. The signals of *a* at 3.71 ppm and *b* at 2.82 ppm are described to the methylene groups of cystamine, and that of *c* at 1.98 ppm to the methyl group from 2-bromoisobutyryl group. The integration ratio of a:b:c equals 1:1:3, in agreement of the structure of AIBCy. IR analysis of AIBCy shows an absorption peak at 2100 cm^{-1}, which corresponds to the asymmetric stretching vibration of the azide group (Fig. 2). This confirms the successful synthesis of the AIBCy.

P(NIPAM-*co*-ProA) and P(DMAEMA-*co*-ProA) were synthesized through RAFT copolymerization. As one of living polymerizations, RAFT polymerization could be used to easily prepare the polymers with well defined structure.[39,40] Figure 1(b) shows ^{1}H NMR spectrum of P(DMAEMA-*co*-ProA), The copolymer composition was determined by comparing the integration of DMAEMA peak (a) at 4.0 ppm and ProA peak (b) at 4.7 ppm. The ratio of a:b is 10:1 or 20:1, which is consists with the molar ratio of charged monomers. Figure 1(c) shows ^{1}H NMR spectrum of P(NIPAM-*co*-ProA) and the integration ratio of PNIPAM peak (a) at 4.0 ppm to ProA peak (b) at 4.7 ppm is 10:1 or 20:1, which is also in agreement with the monomer ratio. The details about the synthesis of the copolymers are summarized in Table 1. The molecular weight and its distribution of the copolymers were characterized by GPC and the result is also listed in Table 1.

PNIPAM and PDMAEMA gels were obtained through the click reaction between alkyne groups of copolymers and azide groups of AIBCy, which was firstly prepared and used by our group. The amount of alkyne groups is little less than azide groups (90 mol %). After thorough dialyze, the gels have no absorbance at 2100 cm^{-1} in IR spectra (Fig. 2). Herein, the crosslinkage density is defined as the molar ratio of ProA to DMAEMA or NIPAM. As shown in Table 1, the click crosslinkage between P(DMAEMA-*co*-ProA) (5 mol % and 10 mol %

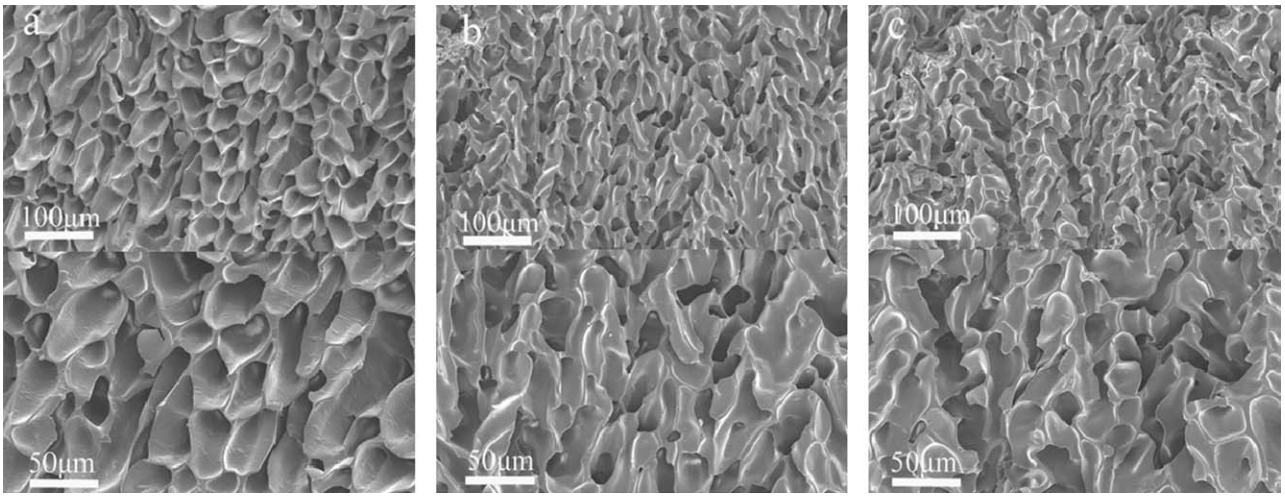


FIGURE 3 SEM micrographs of the morphology of polymer hydrogels produced from (PDMAEMA-*co*-ProA) with 5 mol % ProA (a), (PDMAEMA-*co*-ProA) with 10 mol % ProA (b), and (PDNIPAM-*co*-ProA) with 10 mol % ProA (c).

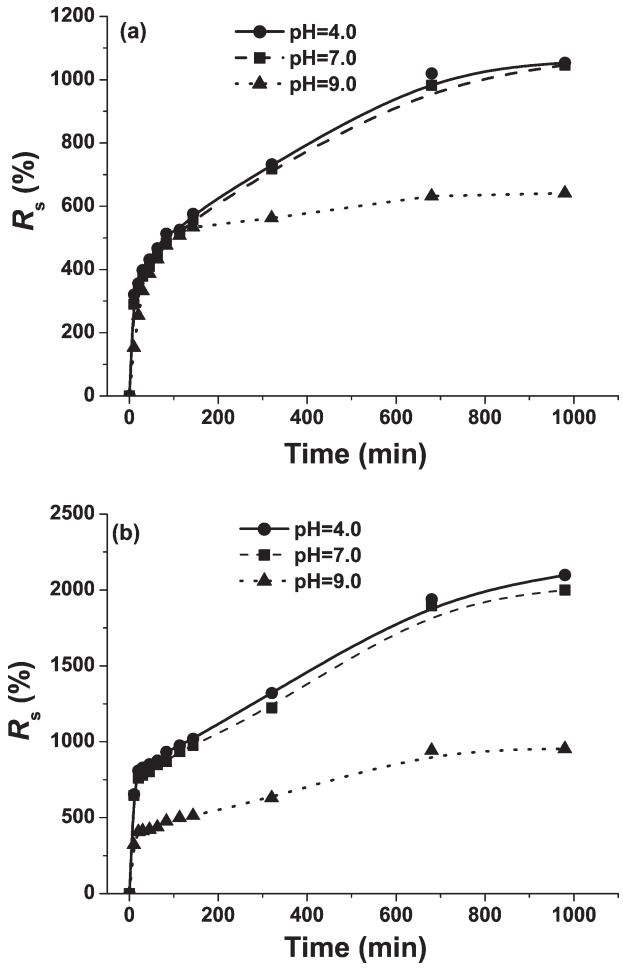


FIGURE 4 Swelling kinetics of PDMAEMA hydrogels at different pHs [(a) 10 mol % ProA, (b) 5 mol % ProA].

of ProA to DMAEMA) and AIBCy leads to the formation of bulk gels. On the contrary, the click crosslinkage between P(NIPAM-*co*-ProA) produce the bulk gel only at 10 mol % of ProA to NIPAM. No formation of bulk gel was observed P(NIPAM-*co*-ProA) with 5 mol % of ProA to NIPAM.

Interior Morphology of Reducible Intelligent Hydrogels

The interior morphology of reducible intelligent hydrogels was observed and their SEM images reveal the macroporous morphology with averaged pore diameter of ~50 μm as shown in Figure 3. The pores are regular and arrange along the same direction, suggesting the controlled porosity structure of the gels. Generally, some pore-forming agents, such as PEG and cellulose, are needed in preparing macroporous hydrogels. Herein, we prepare macroporous networks without them. As expected, PDMAEMA gel from P(DMAEMA-*co*-ProA) with 10 mol % ProA has smaller pore size than that from 5 mol % ProA. It indicates that the porosity structure could be modulated simply by the copolymer composition. This macroporous structure might improve the property of intelligent hydrogels and impart the gels with a rapid response rate in swelling/shrinkage experiment.

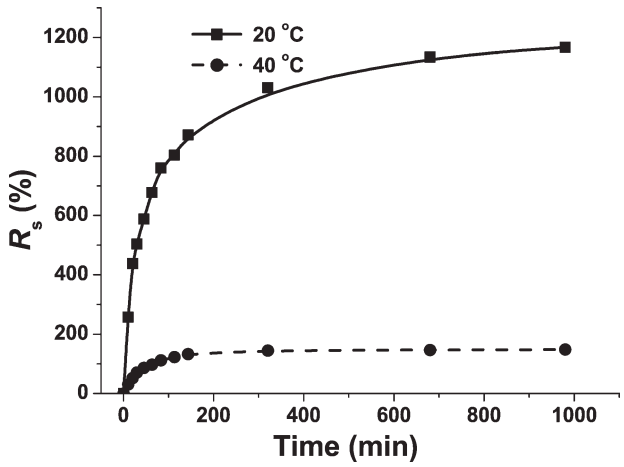


FIGURE 5 Swelling kinetics of PNIPAM hydrogels with 10 mol % ProA at 20 °C and 40 °C.

The Swelling Kinetics of Reducible Intelligent Hydrogels

After the thorough dialysis and freeze-dryness, the swelling kinetics of reducible intelligent hydrogels was investigated to evaluate their water-absorption behavior. Figure 4 shows the swelling kinetics of PDMAEMA hydrogels with two crosslinkage densities at 20 °C in buffer solution with different pHs. Figure 5 shows the swelling kinetics of PNIPAM gel from P(NIPAM-*co*-ProA) with 10 mol % ProA at 20 and 40 °C in distilled water. From the figures, we can find that all the hydrogels have excellent ability to uptake water with swelling ratio of 1000–2000% at 20 °C and pH 7.0, which might be attributed to the macroporous structure of polymer networks. Usually, there are two ways for the hydrogels to uptake water. One is the water binding by polymer matrix via hydrogen bond and another is the capillary action.[41] As for our gels, the pore volume is much larger than the polymer volume, so the capillary action is dominated. Their macroporous structure offers them enough space to hold large amount of water.

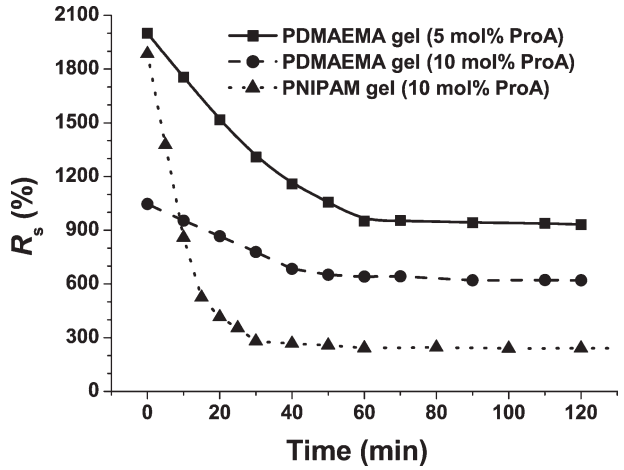


FIGURE 6 Shrinkage kinetics of PDMAEMA hydrogels at pH variation from 7.0 to 9.0 and PNIPAM gel at temperature variation from 20 to 40 °C.

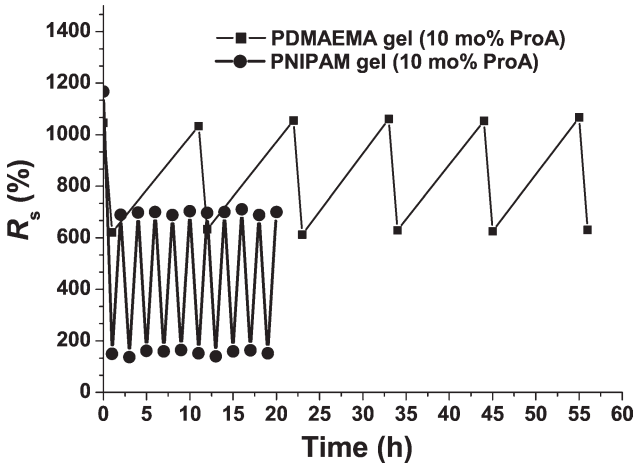


FIGURE 7 Shrinkage-swelling circles of PDMAEMA hydrogels between pH 7.0 and 9.0, and PNIPAM hydrogel between 20 and 40 °C.

Based on the feature of PDMAEMA, PDMAEMA hydrogels exhibit pH responsive behavior. Equilibrium swelling ratio and swelling rate decrease while pH changes from 7.0 to 9.0. This might be cause by two reasons. Firstly, the hydrophilicity of PDMAEMA chains decreases gradually with the increase of pH value,[42] leading to the decrease of binding interaction between water and polymer matrix. Secondly, electrostatic repulsion between PDMAEMA segment decreases while the amount of ammonium groups from PDMAEMA decreased drastically.[43] Thus, the pore size becomes smaller and the capacity of hydrogels for water uptake decreases. It also shows that the saturated swelling ratio also decreases with increasing crosslinking density. This is because the hydrogel with low crosslinking density has larger pore, thus it could uptake more water at quicker rate than the hydrogel with high crosslinking density.

Stimulus Response of Intelligent Hydrogels with pH or Temperature

Figure 6 shows the shrinkage kinetics of PDMAEMA hydrogels when environment pH changes from 7.0 to 9.0. Since the hydrophilicity of PDMAEMA hydrogels become worse and the decrease of electrostatic repulsion between PDMAEMA segments induce the shrinkage of hydrogel network, the swelling ratio of hydrogels decreases. After 40 and 60 min, the shrinkage equilibrium reaches with swelling ratio of 900 and 600% for PDMAEMA hydrogel with 5 and 10 mol % ProA, respectively. The swelling ratio at shrinkage equilibrium keeps relative high, due to the fact that the PDMAEMA is a weak base with pKa about 7.0 and is water-bound within wide pH range. When pH is 9.0, it is partially hydrophilic and still could absorb water somewhat.

As shown in Figure 6, PNIPAM hydrogel undergoes the shrinkage after the environment temperature changes from 20 to 40 °C. PNIPAM homopolymer has LCST about 32 °C in aqueous solution, thus PNIPAM hydrogel also shrinks their volume when the temperature rises above LCST. From the figure, we could find that PNIPAM gel shrinks very quickly at 40 °C. The swelling ratio dramatically decreases from 1200% to 150% in about 30 min. It should be assigned to responsive properties of PNIPAM segment in gel network. Because a majority of gel space is pore capacity, PNIPAM chains only take up a little volume. After it is immersed in water, the size of pore enlarges quickly through capillary action. At the temperature above PNIPAM LCST, the shrinkage of PNIAM induces the exclusion of the most absorbed water from the hydrogel, thus skin of the fully shrunk gel seems dry. Due to a little physical absorption of water by PNIPAM gel, the gel could have 150% swelling ratio at shrinkage equilibrium. This rapid and full shrinkage of PNIPAM hydrogels could help to devise immediate responsive drug delivery system.

To testify these polymer hydrogels can be used circularly and have the dynamistic responses of shrinkage and swelling behavior via pH or temperature variation, their shrinkage-swelling circles are investigated and the results are showed in Figure 7.

Figure 7 shows the shrinkage-swelling circles of PDMAEMA hydrogel with 10 mol % ProA between pH 7.0 and 9.0.

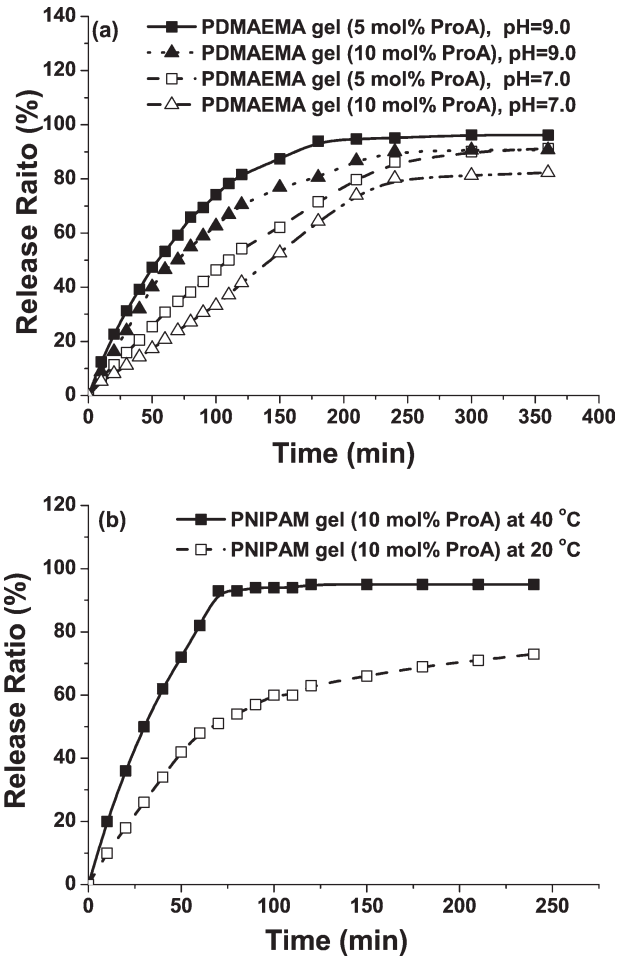


FIGURE 8 (a) Drug release of PDMAEMA hydrogels with time at different pHs, (b) Drug release of PNIPAM hydrogels with time for at two temperatures.

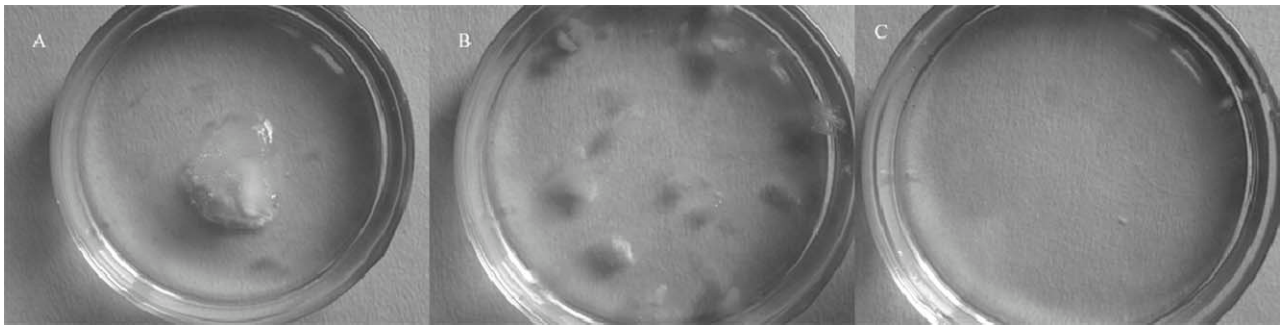


FIGURE 9 Reducible degradation of PDMAEMA hydrogel with 10 mol % ProA in DTT aqueous solution (0.1 *M*) [(A) 1 d, (B) 3 d, and (C) 5 d].

PDMAEMA hydrogel presents an asymmetrical change of swelling ratio in each circle. The shrinkage lasts for about 1 h to reach its equilibrium while the swelling process needs about 11 h to achieve its equilibrium status. However, the shrinkage-swelling circle can be repeated with high reproducibility for thirty times. As for PNIPAM hydrogel with 10 mol % ProA, its shrinkage-swelling circles are quite different form PDMAEMA hydrogel. PNIPAM hydrogel shows a symmetrical shrinking-swelling behavior when changing temperature. This rapid response consists with the result in Figure 6. Only in 1 h, PNIPAM hydrogel can get to swelling/shrinkage balance status. From the figure, we find that PNIPAM gel still shows a quickly responsive behavior in 10 circles. This stable and repeatable responsive behavior endows the hydrogels with the potential in gene or drug delivery system.

The Loading and Release of Ceftriaxone Sodium

Ceftriaxone sodium, one of the third generation of cephalosporin drugs, is well dissolved in water and has been chosen as a model drug in loading and release experiment.[44] It has strong UV-vis absorption at 273 nm, thus its concentration in water can be easily determined. After the hydrogels (50 mg) were saturately swollen in the aqueous solution of ceftriaxone sodium, the amount of loaded drug was 9.5 mg, 4.5 mg, and 5.0 mg for PDMAEMA hydrogel (50 mg) with 5 mol % ProA, 10 mol % ProA, and PNIPAM hydrogel (50 mg) with 10 mol % ProA, respectively. Then, the drug release experiments of drug-loaded hydrogels were carried out at various environments.

Figure 8(a) shows the drug release behavior of PDMAEMA hydrogels as function of time. The hydrogels with different crosslinking degrees demonstrate the varied drug release profiles. PDMAEM hydrogel with low crosslinking density releases the drug at faster rate. It might be caused by larger pore size, which leads to better drug holding/releasing of the hydrogel. The drug release in alkalic environment is quicker than that in neutral system, caused by the shrinkage of PDMAEMA hydrogel. Figure 8(b) shows the drug release behavior of PNIPAM gel. It is obvious that the drug release rate at 20 °C is slower than 40 °C and the maximum amount of release drug is 60 and 95% for 20 and 40 °C, respectively. Faster release and larger release amount are also attributed to the shrinkage of PNIPAM hydrogel at the temperature above LCST.

Reducible Degradation of Intelligent Hydrogels

Because these intelligent hydrogels are prepared by using disulfide crosslinker, they can be degraded by DTT. The decrosslinkage of intelligent gels was carried out in water with DTT as the cleavage reagent. The hydrogels firstly broke down into tiny fragments in DTT aqueous solution. After 5 days, the intelligent hydrogels were totally dissolved in water. The reducible degradation of PDMAEMA hydrogel with 10 mol % ProA is illustrated in Figure 9. Because the rate of gel degradation is much lower than drug release rate, this reducible degradation of intelligent hydrogels exercises little influence to the drug release.

CONCLUSIONS

Reducibly degradable intelligent hydrogels of PNIPAM and PDMAEMA were successfully synthesized by the combination of RAFT polymerization and click chemistry. SEM observation displayed the regular macroporous morphology of the resultant hydrogels. The final hydrogels exhibit temperature or pH stimulus-response with swelling-shrinkage reproducibility in water. The macroporous structure imparts the hydrogels with rapid response and high swelling ratio. The loading and release of ceftriaxone sodium prove the feasibility of the hydrogels as the stimulus-responsive drug delivery system. Furthermore, the experiment of reaction with DTT testifies that the hydrogels could be degraded in the reductive environment, which can facilitate the removal of empty hydrogels, as well as might improve the release of encapsulated drug molecules.

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