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Thermoresponsive Homopolymer Tunable by pH and CO_2

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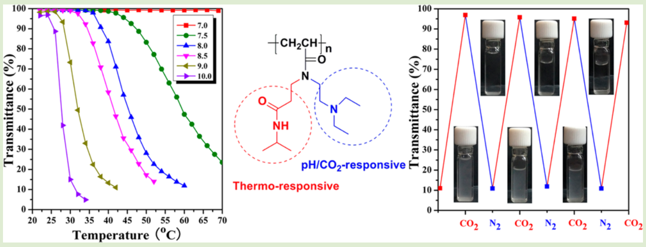
Supporting Information



ABSTRACT: A new acrylamide monomer bearing isopropylamide and *N*,*N*-diethylamino ethyl groups in the side chain, i.e., *N*-(2-(diethylamino)ethyl)-*N*-(3-(isopropylamino)-3-oxopropyl)acrylamide (DEAE-NIPAM-AM), was synthesized through Aza-Michael addition reaction followed by amidation with acryloyl chloride. The homopolymers, poly(*N*-(2-(diethylamino)ethyl)-*N*-(3-(isopropylamino)-3-oxopropyl)-acrylamide)s [poly(DEAE-NIPAM-AM)], with controlled molecular weights and relatively narrow molecular weight distributions were then prepared via RAFT polymerization. The lower critical solution temperature (LCST) of the homopolymer was examined to be influenced by molecular weight, salt concentration, and pH value of aqueous solution. The LCST of the homopolymer could be tuned in a wide temperature window by changing the pH value of aqueous solution, and it increased with the decrease of pH value. Particularly, CO_2 gas as a unique pH stimulus can also reversibly adjust the solubility of homopolymer without the addition of acids or bases.

Stimuli-responsive polymers able to respond to external stimuli have attracted increasing attention since these kinds of polymers have broad applications in areas from material science to biology.[1] For relevant applications, however, the change in behavior of a macromolecule (protein and nucleic acid) is often not the result of a single factor but a combination of environmental changes.[2] To mimic this feature, formulation of multistimulus responsive polymers by incorporating different stimulus-sensitive moieties has spurred significant interest.[3] Among them, temperature and pH dual-responsive polymers were widely studied due to the convenience in adjusting environmental pH and temperature and the ease in the preparation of "smart materials" on the basis of temperature and pH dual-responsive polymers. In general, block and random copolymers are relatively common in double-responsive systems by simply connecting moiety with pH-stimulus property with the other segment with temperature-stimulus behavior.[3−9] For the block copolymer, time-consuming steps in the preparation and purification are usually required to obtain well-defined and pure block copolymers. In addition, the self-assembly of some certain block copolymers in solution is often observed due to the mutual incompatibility of different blocks, which might retard their relative applications.[3,4] pH- and temperature-responsive random copolymers could be another alternative to avoid these drawbacks, which can be prepared by random copolymerization or postfunctionalization strategy.[5−9] However, the ill-defined monomer distribution of random copolymers might inevitably become the obstacle for investigating the influence of structure or the distribution of monomer of copolymers on their dual stimuli-responsive properties. Taking the maturity of synthesis of small molecules into account, the synthesis of well-defined thermo- and pH-responsive homopolymers with thermo- and pH-responsive moieties in each repeated unit has become a long-term goal in chemistry. Van Hest and Li et al. reported the synthesis and property of temperature and pH-responsive homopolymers.[10,11] Their results suggested that it would be much easier to prepare multiresponsive copolymers via copolymerization of a dual responsive monomer with other specific monomers.[10,11]

Herein, we designed a new acrylamide monomer, namely, *N*-(2-(diethylamino)ethyl)-*N*-(3-(isopropylamino)-3-oxopropyl)-acrylamide (DEAE-NIPAM-AM), on the basis of well-studied pH-responsive poly((*N*,*N*-diethylamino)ethyl methacrylate) (PDEAEMA) with a pK_a around 7.3[12] and thermoresponsive poly(*N*-isopropylacrylamide) (PNIPAM) with a LCST about 32 °C.[13] One side segment of this monomer was an *N*-isopropyl amide derivative with the intent to possess thermoresponsive property, and the other side segment was the *N*,*N*-diethylamino ethyl group to keep pH sensitivity. In particular, CO_2 also could be employed as a special protonating agent for the *N*,*N*-diethylamino ethyl group, which allowed reversible switchability of the monomer between hydrophility and hydrophobility by bubbling CO_2/N_2.[12,14−18] This new monomer was synthesized from commercially available reagents of *N*,*N*-diethylethylenediamine, *N*-isopropylacrylamide, and acryloyl chloride via two-step reaction (Scheme 1). Subsequently, RAFT polymerization was utilized to get target homopolymers with controlled molecular weights and relatively narrow molecular weight distributions. The influence of molecular weight, salt concentration, and pH value on the

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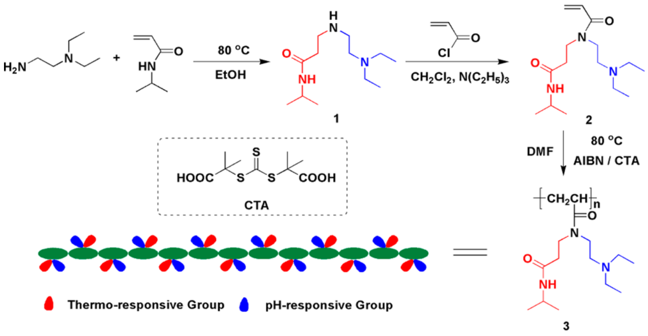
Scheme 1. Synthetic Route of Poly(DEAE-NIPAM-AM) Homopolymer



LCST of the homopolymer was examined by UV−vis transmittance measurement. This kind of homopolymer showed dual thermo- and pH-responsive properties, whose LCST could be tuned from 24.4 to 44.4 °C in a pH window from 10.0 to 7.5. In particular, the solubility of the homopolymer could be reversibly adjusted by bubbling CO_2 and N_2 without the addition of acids or bases. In addition, we need to point out that considering the good tolerance of the Aza-Michael addition reaction a wide range of acrylate- or acrylamide-based functional homopolymers can be envisioned by appropriate selection of amino-containing compounds and acrylate or acrylamide via the synthetic strategy employed in our current work, pointing to the potential broad scope of functional materials.

The key intermediate of 3-(2-(diethylamino)ethylamino)-*N*-isopropylpropanamide **1** was first prepared through Aza-Michael addition reaction using *N*,*N*-diethylethylenediamine and *N*-isopropylacrylamide as starting materials, which contained potential pH(CO_2) and thermoresponsive *N*-isopropyl amide and *N*,*N*-diethylamino ethyl moieties. Figures S1A and S1B (Supporting Information) show ^{1}H and ^{13}C NMR spectra of intermediate **1**, displaying the expected resonance signals of *N*-isopropyl amide, which confirmed the structure of target intermediate **1**.

Subsequently, the obtained intermediate **1** was treated with acryloyl chloride to afford the DEAE-NIPAM-AM monomer. The structure of the DEAE-NIPAM-AM monomer was verified by ^{1}H NMR, ^{13}C NMR, FT-IR, and ESI-MS. The proton resonance signals assured us of the structure of the monomer, showing that $I_a:I_b:I_c:I_d:I_{(e+f)}:I_g:I_i:I_j$ is 1:1:2:2:6:2:6:6 (Figure 1A). The ^{13}C NMR spectrum (Figure 1B) demonstrates the expected resonance signals of double bond (peaks of "d" and "c") and amide groups (peaks of "a" and "b"). In addition, the ESI-MS result (284.2338) was also consistent with the theoretical value (284.2333). All these results confirmed the successful synthesis of target monomer **2**.

As listed in Table 1, a series of well-defined poly(DEAE-NIPAM-AM) homopolymers with relatively narrow molecular weight distributions were prepared via RAFT polymerization. The structures of the obtained homopolymers were confirmed by ^{1}H NMR (Figure S2, Supporting Information). The polymerization kinetics was investigated. Linear dependence of Ln($[M]_0/[M]$) on the time and molecular weight on the monomer conversion was observed in Figure S4A and Figure S4B (Supporting Information), respectively. These observations verified the "living"/controlled nature of RAFT polymerization of the monomer.

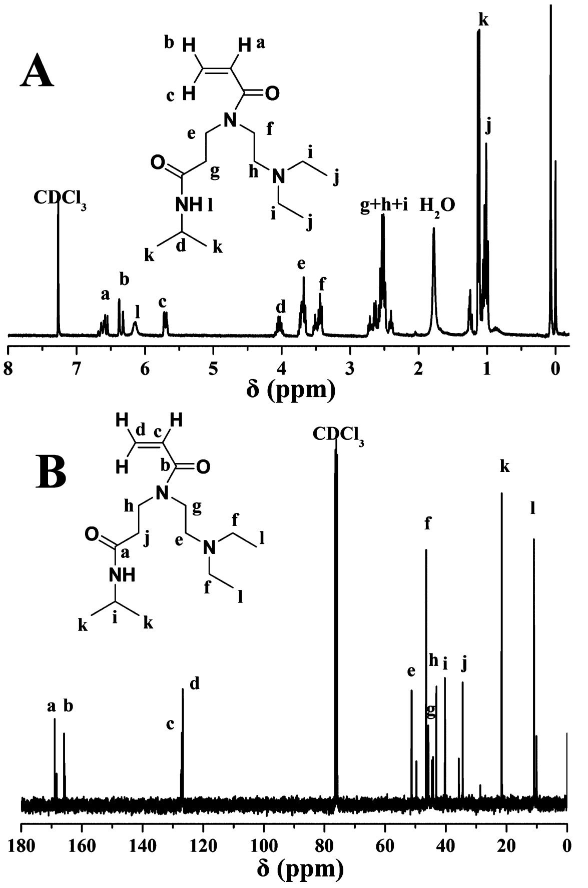


Figure 1. ^{1}H (A) and ^{13}C (B) NMR spectra of *N*-(2-(diethylamino)-ethyl)-*N*-(3-(isopropylamino)-3-oxo-propyl) acrylamide **2** (DEAE-NIPAM-AM) in $CDCl_3$.

Table 1. Synthesis of Poly(DEAE-NIPAM-AM) 3 by RAFT Polymerization[a]

sample	[2]:[CTA]	conv.[b] (%)	M_n[c] (g/mol)	PDI[c]
3a	107:1	21	5200	1.35
3b	163:1	19	7500	1.44
3c	197:1	23	9100	1.44
3d	300:1	16	10900	1.38

[a] [AIBN]:[CTA] = 1:3, solvent: DMF, temperature: 80 °C, time: 16 h. [b] Measured by ^{1}H NMR. [c] Measured by GPC at 35 °C using LiBr-added DMF as eluent.

Thermoresponsive phase transition behavior of the poly-(DEAE-NIPAM-AM) homopolymer was tested by UV−vis approach as shown in Figure 2. The transmittance of homopolymer solution (λ = 600 nm) vs temperature was measured to determine the LCST, which was defined as the temperature at which the transmittance decreased by 5%.[19] Figure 2A shows the change of transmittance of poly(DEAE-NIPAM-AM) **3b** solution during heating and cooling processes. This homopolymer obviously displayed a reversible and abrupt change in transmittance from 26 to 40 °C during the course of heating and cooling.

Figure 2B shows transmittance vs temperature for aqueous solutions of poly(DEAE-NIPAM-AM) **3** with different molecular weights. For homopolymers **3a** (5200 g/mol) and **3b** (7500 g/mol), their LCSTs were 24.2 and 24.5 °C, respectively. As the molecular weight increased to 9100 g/mol (**3c**) and 10 900 g/mol (**3d**), LCSTs of **3c** and **3d** increased to

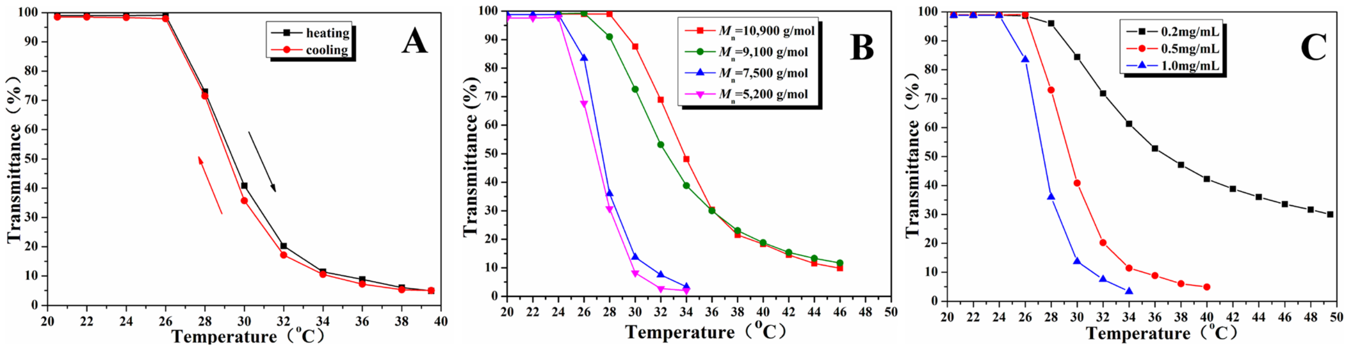


Figure 2. Transmittance vs temperature for aqueous solution of poly(DEAE-NIPAM-AM) when pH = 9.0, (A) during one heating and cooling cycle, [**3b**] = 0.5 mg/mL, (B) with different M_n's, [**3**] = 1.0 mg/mL, and (C) **3b** with different concentrations.

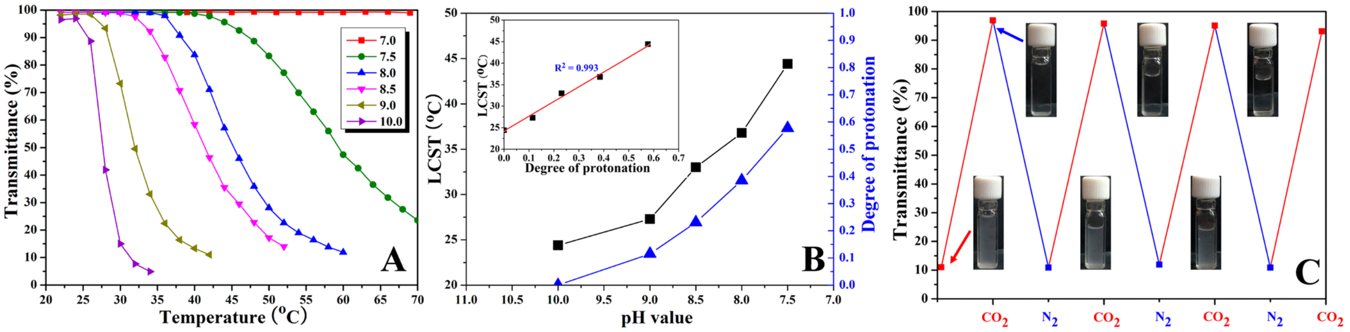


Figure 3. (A) Transmittance vs temperature for aqueous solutions of poly(DEAE-NIPAM-AM) **3b** at different pHs, [**3b**] = 0.5 mg/mL, [NaCl] = 10 mM. (B) Influence of pH on the LCST of the homopolymer and degree of protonantion of the *N*,*N*-diethylamino ethyl group. Inset of the figure shows relationship between the LCST of the homopolymer with the degree of protonation of the *N*,*N*-diethylamino ethyl group. (C) Transmittance change of poly(DEAE-NIPAM-AM) **3b** in aqueous solution at 50 °C by alternating bubbling with CO_2/N_2 stimulation, [**3b**] = 0.5 mg/mL.

27.0 and 28.7 °C. Previous studies on PNIPAM and other thermoresponsive polymers showed that the LCST could be independent,[20] directly dependent,[21] or inversely dependent[22] upon molecular weight. Generally, the end group of thermoresponsive polymers and dilution effect may relate to polymer–solvent interaction and hydrophobic/hydrophilic balance of polymer and, thus, the dependence of LCST with molecular weight, especially in the relatively low molecular weight regime. First, *S*,*S*′-bis(*α*,*α*′-dimethyl-*α*″-acetic acid) trithiocarbonate (BDMAT) was used as a chain transfer agent (CTA) to prepare poly(DEAE-NIPAM-AM) **3** homopolymer (Scheme 1), and carboxyl should be located at both chain ends. Therefore, the hydrophilic carboxyl would perturb the hydrophobic/hydrophilic balance and increase the LCST of the homopolymer with a lower molecular weight.[23] Second, a previous result indicated that for the same mass concentration a higher molecular weight would result in a lower molar concentration, that is, a more "dilute" polymer solution with less polymer chains.[24] In order to test the dilution effect on the LCST of the homopolymer, the concentration dependence on LCST of homopolymer **3b** (M_n = 7500 g/mol) (Figure 2C) and LCSTs of poly(DEAE-NIPAM-AM) **3** with a lower concentration of 0.5 mg/mL (Figure S5, Supporting Information) were examined. The LCST of **3b** was 24.5 °C as the concentration was 1.0 mg/mL. It increased to 26.3 and 28.2 °C with the decrease of the concentration to 0.5 and 0.2 mg/mL, respectively (Figure 2C). One also can see that a lower concentration led to an increase in LCSTs for all of homopolymer **3** with different molecular weights (Figure S5, Supporting Information). These observations showed that the LCST increased upon lowering the concentration of the homopolymer, which was consistent with previous reports.[25,26] Thus, for the same mass concentration, the homopolymer with a higher molecular weight will show a higher LCST. In the current case, the dilution effect might overwhelm the chain end effect, which led to direct dependence of the LCST on the molecular weight of the homopolymer.

Poly(DEAE-NIPAM-AM) **3** homopolymer can be considered to have pH responsiveness for the existence of a tertiary amino group. A previous report showed that the hydrogen bond between the amide group and water molecule would be weakened when salts (e.g., NaCl) were present due to their polarization of adjacent water molecules.[27] One might argue that the change of solution ionic strength (Na^+ and Cl^-) might bring out the difference in LCST at different pHs since Na^+ or Cl^- was introduced during the adjustment of pH by NaOH or HCl. So we first examined the influence of concentration of NaCl on the LCST of homopolymer **3b** (Figure S6, Supporting Information). We found that a higher concentration of NaCl led to a slightly higher LCST. To exclude the influence of salt on LCST, NaCl was added into each solution with a concentration of 10 mM as the adjustment of pH. The solution turbidity for aqueous solution of homopolymer **3b** (0.5 mg/mL) vs temperature at different pHs is shown in Figure 3A. One can see that LCSTs of homopolymer **3b** were 24.4, 27.3, 33.0, 36.8, and 44.4 °C at pH of 10.0, 9.0, 8.5, 8.0, and 7.5, respectively (Figure 3B). The solution was transparent at pH of 7.0 over the experimental temperature range even at 70 °C. Previous reports showed that LCST was primarily determined by the balance between the thermo-dependent hydrophobic unit and ionizable group.[12,14−18] Therefore, we conducted a titration experiment to examine the degree of protonation of the *N*,*N*-diethylamino ethyl group at different pHs (Figure S7, Supporting Information). As the pH decreased from 10.0 to 7.5, the degree of protonation of *N*,*N*-diethylamino ethyl groups increased from 0 to 58% with the increase of LCST from 24.4 to 44.4 °C (Figure 3B). This observation indicated that a higher phase transition temperature was required so as to compensate the improvement of hydrophilicity from the protonated *N*,*N*-diethylamino ethyl group. As shown in the inset of Figure 3B, the LCST of the homopolymer is plotted against the degree of protonation at different pHs, and one can see an obviously linear relationship. It is interesting that no LCST could be observed at pH of 7.0, at which the degree of protonation was 80%, while an obvious LSCT of 44.4 °C was detected at pH of 7.5 with only slightly lower degree of protonation of 73%. This high sensitivity of thermoresponsive behavior to pH, especially at the range around 7, invoked our investigation on a homopolymer's CO_2-trigger thermoresponsive behavior.

As we know, CO_2 gas is a unique pH stimulus which forms a chemical equilibrium with weak carbonic acid in aqueous solution. Unlike most acids, if the solution was purged with an inert gas such as N_2 or Ar, dissolved CO_2 can be easily removed. It was affirmed that tertiary amine groups can be protonated by CO_2 to form a charged ammonium bicarbonate which can be recovered upon CO_2 removal.[12,14−18] Figure 3C presents the transmittance change of poly(DEAE-NIPAM-AM) **3b** in aqueous solution at 50 °C by alternating bubbling with CO_2/N_2. Before treating with CO_2, the turbid solution can be well observed at 50 °C. After the treatment with CO_2, the transmittance increased rapidly from 10% to 97%, accompanied by the decrease of pH from 8.5 to 5.2. The decrease in pH indicated the protonation of tertiary amine groups. Subsequently, a sharp drop of transmittance can be observed by passing N_2 through the solution to remove CO_2 due to the deprotonation of the tertiary amine of the *N*,*N*-diethylamino group. Repeatable cycles under an alternating CO_2/N_2 stimulation as shown in Figure 3C indicated gas-responsive reversibility of the homopolymer.

To conclude, a new kind of poly(DEAE-NIPAM-AM) homopolymer with controlled molecular weights and relatively narrow molecular weight distributions was prepared from *N*,*N*-diethylethylenediamine, *N*-isopropylacrylamide, and acryloyl chloride via a two-step reaction and RAFT polymerization. UV−vis measurement showed that the LCST of poly(DEAE-NIPAM-AM) homopolymer in aqueous solution was influenced by pH value, molecular weight, and polymer and salt (NaCl) concentration. In particular, the LCST of the homopolymer could be adjusted by changing the pH of aqueous solution in the range from 7.0 to 10.0 for the existence of tertiary amino groups, and a lower pH would result in a higher LCST. Given the high sensitivity of the LCST of poly(DEAE-NIPAM-AM) to pH, the tertiary amine groups of the homopolymer can also be protonated by bubbling CO_2 to make the hydrophobic homopolymer hydrophilic at the temperature of 50 °C, and the protonation process can be reversed through the simple bubbling of N_2. CO_2 as a nontoxic, benign, and cheap trigger can make this kind of dual thermo- and pH-responsive homopolymer show more promising applications in the preparation of multiresponsive polymers and hydrogels for controlled drug delivery[28] and biomedical sensors.

ASSOCIATED CONTENT

Supporting Information

Experimental details for the polymer synthesis and characterization of DEAE-NIPAM-AM monomer, ^{1}H NMR spectrum of poly(DEAE-NIPAM-AM) homopolymer, kinetic study on RAFT polymerization, dependence of NaCl concentration on LCST of **3b**, and titration experiment. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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(19) LCSTs of homopolymers under different conditions were estimated and defined as the temperature at which the transmittance decreased by 5%. There might be certain errors in the measurement of

LCSTs via UV−vis approach since the data obtained might not be abundant enough to ensure the accuracy of the LCST. Thus, we assumed an uncertainty of ±5% in the LCSTs of homopolymers.

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