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Upper critical solution temperature (UCST) phase transition of halide salts of branched polyethylenimine and methylated branched polyethylenimine in aqueous solutions

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Received 00th January 20xx,
Accepted 00th January 20xx

DOI: 10.1039/x0xx00000x

www.rsc.org/

The upper critical solution temperature (UCST) phase transition of halide salts of branched polyethylenimine (PEI) and methylated branched polyethylenimine (MPEI) is first reported in aqueous solutions. In particular, iodide counter-ions can introduce the UCST property in MPEI. The importance of the counter-ion composition of MPEI for UCST transition is discussed in detail.

Polymers with temperature responsiveness have been extensively researched in past decades. The temperature-responsive phase transition arises from different interactions between polymer and solvent molecules according to temperature variation. In a lower critical solution temperature (LCST) mixture, a polymer is miscible with a solvent below a certain temperature, but it is abruptly phase-separated above that temperature. On the other hand, an upper critical solution temperature (UCST) polymer-solvent mixture exhibits phase separation below the phase transition temperature. In particular, such phenomena in aqueous mixtures have been used in various fields, including hydrogels [1], drug delivery [2], chromatography [3], water purification [4], and optical devices [5].

In many cases, UCST phase transition occurs when solute-solute or solvent-solvent interactions dominate the solute-solvent interaction to generate the positive enthalpy of mixing ($\Delta H_m > 0$), unlike LCST phase transitions, which are more dependent upon the negative entropy of mixing ($\Delta S_m < 0$) due to the strong solute-solvent interaction [6]. In aqueous solution, examples of UCST polymers have been reported less frequently than those of LCST polymers, partially due to the characteristic strong interaction between polymers and water molecules with strong polarity and hydrogen-bonding capability [7]. Some selective polymers with

hydrogen-bond-forming residues, such as urea [8] and amide [9], or zwitterion residues, such as sulfopropyl ammonium [10] and sulfopropyl imidazolium [11], exhibit UCST properties in water because the strong polymer-polymer interaction can overcome the polymer-water interaction to generate positive ΔH_m . It was also reported that complex coacervate micelles composed of oppositely charged polymeric ions showed UCST-like behaviour in aqueous solutions [12].

Because the strong electrostatic interaction between polymer residues can induce UCST phase transition, we hypothesized that simple polymer-monomeric ion salts, not zwitterionic polymers or oppositely charged polymeric ion-based coacervates, could also exhibit a similar transition in aqueous solution. Of course, a separate monomeric ion salt has a larger positive statistical ΔS_m due to a larger increase of the degree of freedom during the solvation process compared to the corresponding zwitterion or polymer-polymer ion salt. Because the phase transition occurs when the Gibbs free energy of mixing ($\Delta G_m = \Delta H_m - T\Delta S_m$) becomes zero at a certain polymer-ion salt/water composition [13], the phase transition temperature ($T = \Delta H_m/\Delta S_m$) of the separate polymer-monomeric ion salt might be lower than that of the zwitterionic or polymer-polymer ion salt if we assume that other conditions are equal in both polymers. Therefore, if we want to obtain UCST characteristics of a separate polymer-monomeric ion salt in an appropriate temperature range, ΔH_m should be increased to compensate for the increase of ΔS_m .

As a model polymer backbone for UCST transition, we chose branched polyethylenimine (*b*-PEI) with a high amine density (1 amine residue per 43 Da). Primary, secondary, and tertiary amines of *b*-PEI were gradually protonated with the addition of hydrogen halide, and the resulting ammonium residues could interact with halide counter-ions (Fig. 1A). If the interaction was strong enough to overcome the water-ion interaction, a positive ΔH_m could be generated to show UCST phase transition. As shown in Fig. 1 and Fig. S1, ESI[†], various polyethyleniminium (PEI) halide salts exhibited UCST phase transition in the temperature range between 0 °C and

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[†]Electronic Supplementary Information (ESI) available: phase transition behaviour of PEI halides and MPEI, NMR spectra of MPEI, and pH values of the PEI or MPEI halide solutions. See DOI: 10.1039/x0xx00000x

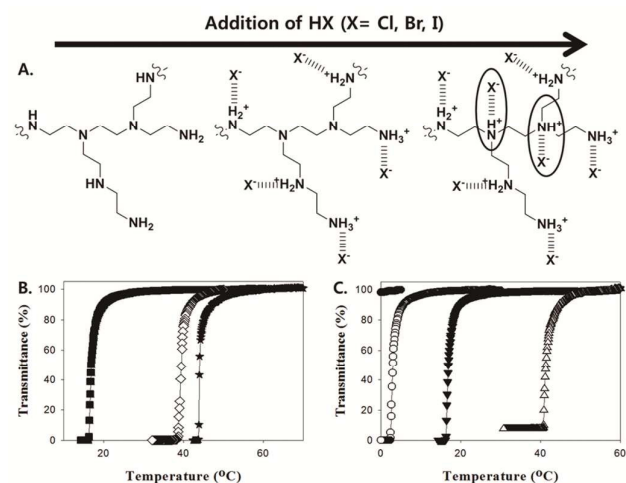


Fig. 1 (A) Interaction between ammonium residues and halide counter-ions in PEI halide salts. (B) Phase transition of PEIB at 30% (w/w) polymer composition (HBr/N ratio = 1.5 (■), 2.0 (◇), and 2.5 (★)). (C) Phase transition of PEIB at HBr/N ratio of 1.5 (polymer composition (w/w) = 20% (●), 25% (▼), and 30% (Δ)).

50 °C. UCST phase transition of PEI halide is quite sensitive to the amount of hydrogen halide. Phase transition temperature of PEI bromide (PEIB) increased as the HBr/amine (N) ratio increased (Fig. 1B). Excessive HBr might have induced the further protonation of internal tertiary amines in PEI [14], and the electrostatic interaction between ammonium and halide ions increased to elevate ΔH_m and the phase transition temperature. Excessive HBr might also elevate the phase transition temperature by salting-out type effect. Phase transition temperature of PEIB was also dependent on the composition (Fig. 1C). Both PEI chloride (PEIC) and PEI iodide (PEII) exhibited the UCST phase transition, although it was observed at a very narrow range of the composition and halide/N ratio (Fig. S1, ESI[†]). The UCST phase transition of simple polymer-halide salts in aqueous solutions is reported here for the first time, although the UCST phase transition of polymer-complex ions was reported in a few previous papers [15].

However, UCST phase transition of PEI halide can be observed only under extreme pH conditions (Table S1, ESI[†]), probably because a considerable number of cationic ammonium residues, which interact with halide anions, can be generated only under those conditions. To obtain polymeric salts, which can show the UCST phase transition under more tolerable conditions, we prepared a methylated PEI possessing permanent cationic ammonium residues irrespective of pH conditions (Fig. 2A). The methylated polyethyleniminium iodide (MPEII) was prepared by the nucleophilic substitution between primary, secondary, and tertiary amine residues of *b*-PEI and methyl iodide (CH₃I) [16]. The quaternary ammonium degree of each polymer salt was determined by the inverted gate (INVGATE) ¹³C NMR technique [17]. We could obtain an MPEII of which 35–38% of the total amine residues were quaternized (MPEII_{0.35} or MPEII_{0.38}) using the 5.0 equivalent of CH₃I (Fig. S2, ESI[†]). Most of the quaternary ammoniums originated from more-exposed primary amines on the external part of *b*-PEI through methylation reactions. Excess iodide salts were removed or exchanged into chloride or bromide to

produce methylated polyethyleniminium chloride (MPEIC) or bromide (MPEIB) by dialysis (Fig. 2A).

Interestingly, unlike the unmethylated correspondents, only MPEII exhibited UCST phase transition in the temperature range of 0–20 °C (Fig. 2B). Marginal hysteresis ($\Delta T \approx 1\text{--}2^\circ\text{C}$) was observed in the phase transition of MPEII (Fig. S3, ESI[†]), which is a typical characteristic of temperature-sensitive phase transition of polymers [18]. Both MPEIC and MPEIB are miscible with water in the temperature range of 0–100 °C. In case of MPEI, the hydrophobic interaction between the methylated ammonium residues (NR₄⁺) and the halide ions might also have been an important factor as well as the electrostatic interaction. The molar Gibbs free energies of the hydration of Cl⁻, Br⁻, and I⁻ are -340, -315, and -275 kJ/mol, respectively [19]. Due to the more hydrophobic character of I⁻ compared with other halide ions, the interaction of I⁻ with NR₄⁺ is fairly higher than Cl⁻ and Br⁻ [20]. The stronger interaction between NR₄⁺ and I⁻ can dominate the interaction between water molecules and ions to generate positive ΔH_m and give the UCST property to MPEII. However, the water-ion interaction might be stronger than the weaker interaction between NR₄⁺ and other halide ions to generate negative ΔH_m and negative ΔG_m ($=\Delta H_m - T\Delta S_m$) in the temperature range.

The composition-temperature phase diagram of MPEII/water mixtures showed a typical convex shape that has been observed in many other UCST mixtures (Fig. 2C) [21]. Fig. 2C also shows the effect of molecular weight on the phase transition temperature. High-molecular-weight MPEII_{0.35} (25 kDa) has a higher phase transition temperature than low-molecular-weight MPEII_{0.38} (0.80 kDa). High-molecular-weight MPEII has a less-positive ΔS_m than low-molecular-weight MPEII due to a smaller combinatorial entropy gain during mixing. Therefore, MPEII_{0.35} (25 kDa) showed a higher phase transition temperature ($T = \Delta H_m / \Delta S_m$) than MPEII_{0.38} (0.80 kDa), assuming that both derivatives have a similar ΔH_m at similar degrees of quaternization (35% vs 38%).

To investigate the effect of counter-ions, the phase transition of MPEI halide was observed by varying the counter-ion composition

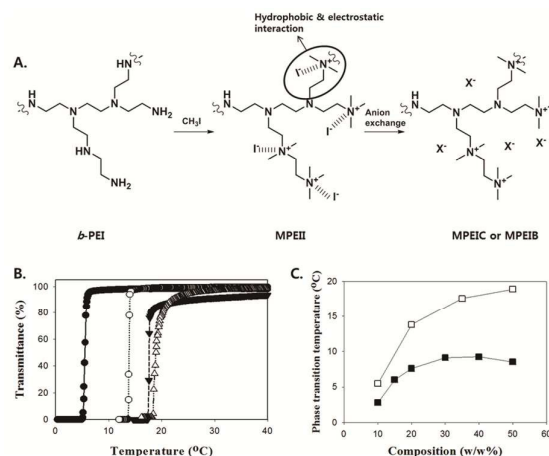


Fig. 2 (A) Preparation of various methylated PEI (MPEI) salts by methylation and anion exchange. (B) Phase transition of MPEII_{0.35} (25 kDa) (polymer composition (w/w) = 10 % (●), 20 % (○), 35 % (▼), and 50% (Δ)). (C) Composition-temperature phase diagram of MPEII_{0.38} (0.80 kDa) (■) and MPEII_{0.35} (25 kDa) (□).

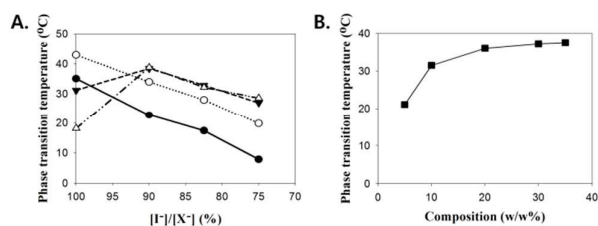


Fig. 3 (A) Change of phase transition temperature of MPEI_{0.35} (25 kDa) at various counter-ion composition and the total anion concentration ($[A^-] = [Cl^-] + [I^-] = 1.75$ M (●), 2.00 M (○), 2.25 M (▼), and 2.50 M (△)). (B) Composition-temperature phase diagram of MPEI(BF₄)_{0.35} (25kDa) (■).

and total anion concentrations ($[A^-] = [I^-] + [Cl^-]$) (Fig. 3A). When only I^- existed as the counterion ($[I^-]/[A^-] = 100\%$), the phase transition temperature increased until the total anion concentration ($[A^-]$) was 2.00 M and decreased above that concentration. The elevation of phase transition temperature at lower anion concentrations (< 2.00 M) indicated the increase of the polymer-ion interaction. Similar to the salting-out effect, the increased ionic strength might strengthen the hydrophobic interaction between ammonium and I^- in this range of concentrations. However, at higher anion concentrations (> 2.00 M), the reduction of electrostatic interaction by the decreased Debye length might have dominated the increased hydrophobic interaction to exhibit the salting-in type phenomena of MPEI halide. The former salting-out effect seemed to be dominant when Cl^- co-existed with I^- . Generally, smaller and more hydrophilic ions, such as Cl^- , exhibit a stronger salting-out effect than larger and more hydrophobic ions, such as I^- [22]. Therefore, the phase transition temperature increased as the total anion concentration increased when the I^- content ($[I^-]/[A^-]$) was lower than 90%, although no salting-out was observed with an $[A^-]$ greater than 2.25 M. Meanwhile, the phase transition temperature decreased as the I^- content decreased below 90%, and the phase separation could not be observed at the I^- content below 70%. This result clearly supports the importance of the interaction between NR_4^+ and hydrophobic I^- in MPEI for obtaining the UCST property. With an I^- content of between 100% and 90%, the stronger salting-out effect of Cl^- and the stronger $NR_4^+ \cdots I^-$ interaction competed with each other to show a rather complex intersection of graphs.

To investigate the effect of the hydrophobicity of counter-ions in MPEI on UCST phase transition behaviour, the I^- of MPEI was replaced with other hydrophobic anions, such as thiocyanate (SCN^-) or tetrafluoroborate (BF_4^-), of which the molar Gibbs free energies of hydration are -280 and -190 kJ/mol, respectively [19]. As shown in Fig. 3B, the tetrafluoroborate salt of MPEI (MPEI(BF₄)_{0.35}) exhibited a UCST phase transition with higher phase transition temperatures than MPEI_{0.35}. The stronger hydrophobicity of BF_4^- over I^- would contribute to the elevation of the phase transition temperature in aqueous solutions. Interestingly, even though the molar Gibbs free energies of hydration of SCN^- is similar to that of I^- , the thiocyanate salt of MPEI (MPEI(SCN)_{0.35}) did not exhibit the UCST phase transition and was miscible with water in the temperature range of 0 – 100 °C. It is probable that other characteristics of anions would affect the interaction with MPEI polymers. However, the hydrophobicity of counter-anions seemed

to be an important factor for the introduction of the UCST property into alkylated quaternary ammonium polymers.

In conclusion, we demonstrated that the simple polymer-ion salt such as the halide salts of PEI and MPEI could show the UCST phase transition in aqueous solutions if the polymer-ion interaction is strong enough to overcome the solute-water interaction. Without the complex synthesis of polymers containing zwitterionic or hydrogen bonding moieties, the simple exchange of counter-ions or methylation could introduce the UCST property into pre-existing polymers. After future research varying the cationic and anionic parts of polymers and ions, the characteristics of temperature-responsive polymeric salts will be understood in more detail, and they will be more helpful in the development of practical temperature-responsive smart materials for various applications.

This work was supported by the GAIA project (G113-00055-3004-0) funded by Ministry of Environment, Korea, and the grant of the Korea Health Technology R&D Project (HI14C2310) funded by the Ministry of Health & Welfare, Korea.

Experimental section

Measurement of the UCST phase transition

The phase transition was monitored by a V-650 UV/vis spectrophotometer (Jasco, Japan) equipped with an STR-707 water thermostatted cell holder. Each sample was completely dissolved in aqueous solution above phase transition temperature and then positioned in the temperature-controlled holder. The phase transition temperature was determined as the temperature showing 50% of the transmittance when the cooling rate was set at a 1 °C/min. For the adjustment of total anionic concentration in Fig. 3A, NaCl and NaI were added to the mixture of MPEI and MPEIC at a specific $[I^-]/[A^-]$ ratio.

Preparation of MPEI halide

A solution of *b*-PEI ($M_w = 25,000$ ($PD(M_w/M_n)=2.5$) or $M_w = 800$ ($PD(M_w/M_n)=1.3$)) in *N,N*-dimethylformamide (DMF) was added to a stirred solution of *N,N*-diisopropylethylamine in DMF at 30 °C. CH_3I (5.0 eq. of total amines in *b*-PEI) was slowly added to the solution. After 48 h of stirring, ammonium hydroxide solution was added to the solution for quenching. For the purification of MPEI (25 kDa), the reaction mixture was dialyzed against ethanol ($\times 3$) and deionized water ($\times 5$) using a membrane (Spectrum Laboratories, Inc. (USA); MWCO = 6-8 kDa). MPEI (0.80 kDa) was purified by ethanol precipitation. The purified MPEI was lyophilized to prepare the product as a pale yellow powder. The structure was characterized using the inverted gate (INVGATE) ¹³C NMR technique (Agilent 400-MR DD2 (400 MHz)). Other MPEI salts were obtained by excessive dialysis of MPEI against corresponding sodium salt solutions (NaCl, NaBr, NaSCN, or NaBF₄).

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