Design of polymerized ionic liquids showing highly thermosensitive LCST-type phase transition in water

2022. 03

Yuki Deguchi-Nishida
Department of Biotechnology
Tokyo University of Agriculture and Technology

Table of contents

Chapter 1 Thermoresponsive polymers showing LCST-type phase transition with water

Chapter 2 Design and properties of poly(ionic liquid)s to show LCST-type phase transition with water

Chapter 3 Control of phase transition temperature of poly(ionic liquid) in water

Chapter 4 Design of thermoresponsive hydrogel derived from ionic liquids

Chapter 1

Thermoresponsive polymers showing LCST-type phase transition with water.

1.1 LCST-type phase transition

The lower critical solution temperature (LCST) is the boundary temperature of following two phases, miscible phase at low temperatures and separated phase at high temperatures. Polymers having such LCST are phase-separated when the temperature of the solution is above the LCST. The polymers become compatible with solution again when the solution is cooled below the LCST. This phenomenon is called LCST-type phase transition.

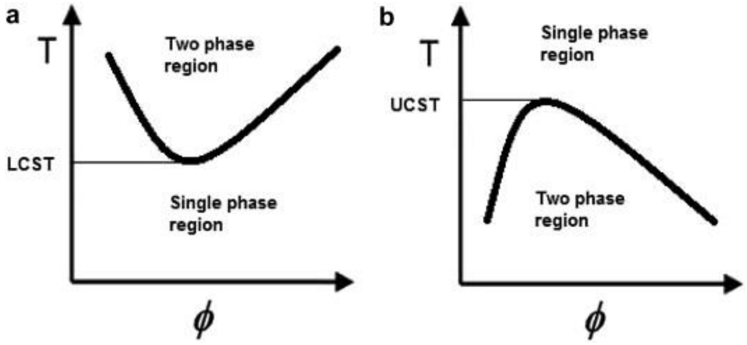


Figure1-1 Temperature vs. polymer volume fraction (ϕ). Schematic illustration of phase diagrams for polymer solution (a) lower critical solution temperature (LCST) behavior and (b) upper critical solution temperature (UCST) behavior.[1]

1.2 Polymers showing LCST-type phase transition

Since discovering LCST-type phase transition in cellulose derivatives in the 1930s, researchers have been studying LCST-type phase transition polymers, and fundamental studies on the properties of solutions have been conducted. Especially in the last several decades, applications to switching devices, and hydrogels have been proposed. Typical polymers exhibiting LCST-type phase transitions and their phase transition temperatures are shown in **Table 1-1**. The typical LCST polymers consist of *N*-isopropylacrylamide[2,3], *N,N*-diethylacrylamide[4] methyl vinyl ether[5] and *N*-vinylcaprolactam.[6,7] Poly(*N*-isopropylacrylamide) (PNIPAM), which has amide group and isopropyl group as hydrophilic and hydrophobic groups, has been the most extensively studied as a temperature-responsive polymer since the temperature-responsive behavior of aqueous solutions was first reported by Heskins and Guillet in 1968.[8] Here, the research results on PNIPAM are mainly described.

Table 1-1 Some LCST-type thermoresponsive polymers.[1]

Polymer	Phase transition temperature in water
Poly(*N*-isopropylacrylamide)	30 – 34 ºC
Poly(*N,N*-diethylacrylamide)	32 – 34 ºC
Poly(methyl vinyl ether)	37 ºC
Poly(*N*-vinylcaprolactam)	30 – 50 ºC
PEO-*block*-PPO	20 – 85 ºC
Poly(pentapeptide) of elastin	28 – 30 ºC

1.3 Phase transition behavior

LCST-type Phase Transition of Linear Polymers in water

It has been confirmed that PNIPAM aqueous solution shows phase transition around 32 ºC, and the LCST is constant molecular weight above 50000 regardless of concentration 0.01 ~ 1%.[9,10] The effect of tacticity and the stereoregularity of polymers were also studied.[11] Since the phase transition of an aqueous solution of PNIPAM was accompanied by a change in turbidity, a standard method for determining the phase transition temperature is to define the temperature when the transmittance of light decreases to a specified ratio as a cloud point (CP or T_c). However, the transition is not sharp enough against temperature change and it is difficult to observe the macroscopic light transmittance change in a low molecular weight polymers or a low concentration solution. As methods for evaluating phase transition behavior except for transmittance measurement, differential scanning calorimetry (DSC) measurement was used. The viscosity change was also examined.[12,13]

Conformation and properties of temperature-responsive polymers before and after phase transition have been studied from various viewpoints by light scattering, simulation, ultrasonic spectroscopy, NMR, and Raman spectroscopy.[14–21] Some temperature-responsive polymers, such as PNIPAM, are hydrated in dilute solutions in a fully elongated random coil state and that they change conformation from a coiled state to a collapsed globular state above the LCST.[22] The phenomenon observed macroscopically as turbidity change is due to the aggregation of molecules in the globular state by hydrophobic interaction.

Siesler *et al.* evaluated the interaction between PNIPAM and water molecules by IR spectroscopy, and in clarifying the structure, interaction, and microscopic changes in individual functional group, they confirmed the phase transition behavior in which the terminal isopropyl group was first dehydrated and then the main chain aggregated by heating.[23] Ozaki *et al.*. found that the amide group of PNIPAAM, which is hydrated at low temperatures, changed to a C = O ... NH bond above LCST with reflection/infrared (ATR/IR) spectroscopy.[24,25] Meada *et al.* also found that 13% of the carbonyl group interacts with the NH group upon heating. In addition, they clarified that dehydration occurred by heating, because CH stretching vibration of isopropyl side chain and main chain of PNIPAM shifted to the low frequency side in IR measurement.[26,27] These results suggest that dehydration of hydrophobic groups and changes in the hydrogen bonding of hydrophilic groups cause the phase transitions.

Saito *et al.* and Cho *et al.* reported that water molecules around the isopropyl group form water cages and stabilize the structure through solvent interactions.[28,29] Based on this water cage concept and the results of the structural changes obtained with the above measurements, it is considered that PNIPAM form hydrogen bonds with water at low temperatures and that water molecules are hydrophobic hydrated to surround the hydrophobic groups of PNIPAM. Heating breaks the hydrogen bonds among PNIPAAM and water molecules and forms hydrophobic hydration to wrap the entire PNIPAM chain (**Figure 1-2**).

Kitamura *et al.* examined the phase transition and phase separation speed of PNIPAM aqueous solution using a laser temperature-jump transmission light measurement.[30–32] As a result, it was reported that the coil-globule phase transition of PNIPAM occurred about 30 microseconds, while the phase separation of aqueous solution is much slower than the phase transition and occurs over several milliseconds to several 100 milliseconds. In addition, in a report in 2015[31], the following 3 points were clarified as guidelines for speed-up of phase separation: increase of aqueous solution concentration, weight average molecular weight of about 60,000, and improvement of stereoregularity.

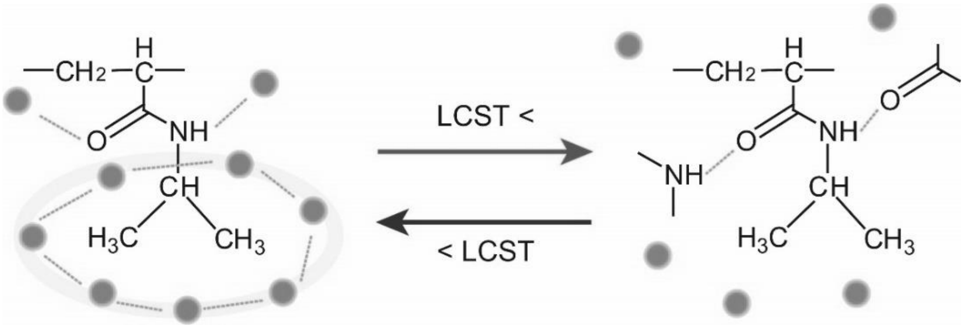


Figure 1-2 Schematic illustration of the behavior of water molecules and PNIPAM undergoing phase transition.[29]

LCST-type Phase Transition of hydrogel

There are also many studies of gel materials fabricating the termoresponsivity polymers owing to their fascinating properties and functions. Hydrogels having 3D network of polymer chains are generally filled with water. They are categorized into "physical gel" and "chemical gel" according to the difference in cross-linking method. "The physical gel is cross-linked by physical interaction such as intermolecular interaction by hydrogen bonds and ionic bonds, and physical entanglement of polymer chains. Physical gels undergo reversible sol-gel transition by external stimuli such as heating. Against these, Chemical gels formed by cross-links with covalent bonding, so that they do not dissolve into solvent unless the cross-linking is destroyed.

PNIPAM is recognized to be one of the most actively studied in hydrogel systems. The volume change process of PNIPAM gels during heating has been studied and exploited by many researchers since 1985.[33] Above the LCST, the polymer chains undergo a globule transition, which causes the hydrogel to shrink and generally become opaque. On the other hand, at temperatures lower than the LCST, the polymer chains reverse to an extended coil chain causing the hydrogel to swell and become transparent[34]. Using this volume change and solvent evacuation, various applications have been proposed, mainly in the biomedical field, such as actuators and drug delivery systems. This will be discussed in a Section 1.5. Most of the reported PNIPAMs form chemical gels; however, the size of the gels can be designed from macro to micro size. For the field of further oversized gels, the mechanical strength is often the subject of interest[35–37], while for the smaller gels, there seems to be more research on responsiveness and functionality.[38,39] These are deeply related to the applications.

1.4 Control of phase transition temperature

1.4.1 The effect of hydrophilicity of polymers

The control of the phase transition temperature of temperature-responsive polymers is essential for developing applications in various fields. The most typical way to control the LCST is to change the hydrophilicity of the polymer chain by copolymerization of monomers with different hydrophilicity.[40,41] Generally, in a polymers system that exhibit LCST-type phase transition in water, more hydrophilic polymers exhibit higher LCST, and more hydrophobic polymers exhibit lower LCST. For example, copolymer containing hydrophilic *N,N*-dimethylacrylamide (DMAAm) in the main chain shows the LCST to a higher temperature range, while copolymerization of hydrophobic buthylmethacrylate (BMA) can shift the LCST to a lower temperature range. **Figure 1-3** represents the change in the transmittance of the solution of PNIPAAM oligomer copolymerized with DMAAm or BMA upon heating.[42] LCST increases with the increase in the percentage of DMAAm fraction in the polymer chain, and conversely, when BMA is copolymerized, LCST decreases with the increase in its fraction. Thus, the LCST can be controlled step by step by changing the hydrophilicity according to the ratio of copolymerization. However, the introduction of non-temperature-responsive parts tends to slow down the temperature responsiveness of the entire polymers. **Figure 1-4** shows the change in phase behavior sensitivity of PNIPAM with difference BMA fraction.[43] The thermoresponsive sensitivity of the mixture dramatically decreased with increasing BMA fraction. This characteristic

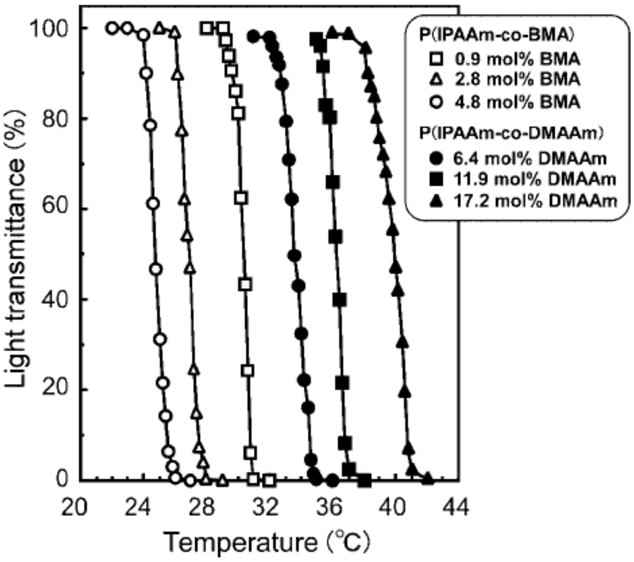


Figure 1-3 Temperature-dependent transmittance change of copolymer/water mixture.[42]

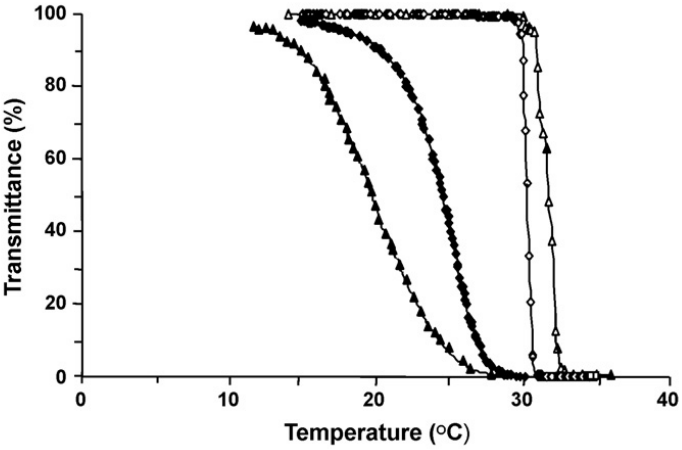


Figure 1-4 Temperature-dependent transmittance change of PNIPAM with difference BMA fraction. (△); PNIPAAm, (◇); BMA-1%, (◆); BMA-3%,(▲); BMA-5%.[43]

prevents the wide application of thermoresponsive polymer.

Since the LCST is influenced by the hydrophobicity of the polymers, the LCST can also be controlled by designing the monomer structure such as the alkyl chain length and introduced functional groups. Furthermore, the LCST decrease as the alkyl chain length, but the structural isomers also effected the LCSTs even at the same carbon number of the alkyl chain. This result may be comprehended by the steric hindrance because polymers with branched structures generally show higher LCST than the linear ones.

1.4.2 The effect of molecular weight of polymers

A method to control the phase transition temperature has been reported by using the polymer with precisely controlling the degree of polymerization by living polymerization.[9] According to Stöver *et al.*, the phase transition temperature of PNIPAM decreases with increasing molecular weight. **Figure 1-5** shows that the phase transition temperature decreased from 43.0 to 33.3 °C as the molecular weight increased from 2.8 to 26.5 kD. The dependence of the phase transition temperature on the molecular weight of polymers is attributed to the difference in the free volumes caused by the polymer chains and water. It is known that the phase transition temperature is proportional to the critical value of the Flory–Huggins interaction parameter and that the phase transition temperature decreases with the ratio of the molar volume of the polymer to that of water.[44] Therefore, when the molar volume of the polymer increases with the length of the chain, the phase transition temperature of the polymer decrease with the molecular weight of the polymer.

Regarding polymers other than PNIPAM, Aoshima's group examined the effect of molecular weight using various vinyl ether polymers having oxyethylene chains. Since these polymers were prepared by living radical polymerization, the molecular weight was controlled precisely.[45] As **Figure 1-6** shows, for both 0.1 and 1 wt% of polymer aqueous solutions, the phase transition temperature decreased with increasing the average molecular weight in the range from 1×10^4 to 2×10^4. However, when the molecular weight is above 2×10^4, the phase transition temperature is almost constant or slightly decreased with increasing molecular weight. In contrast, for a 10 wt% aqueous solution, no drastic change is observed. These results suggest that the effect of molecular weight on phase transition temperature is significant at the low concentration of low molecular weight polymer.

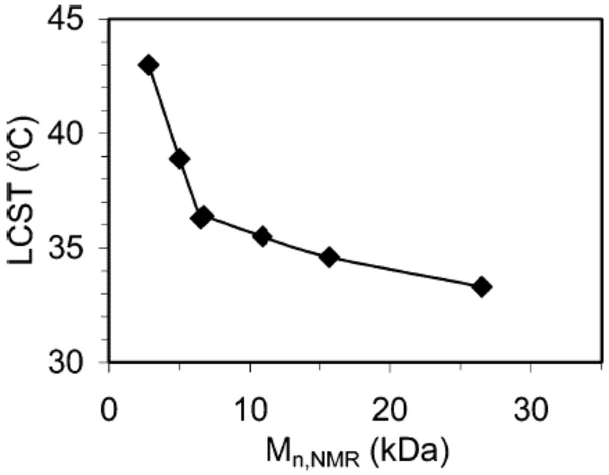


Figure 1-5 Cloud point (50%T) *vs* polymer molecular weight ($M_{\mathrm{n,NMR}}$) for narrow-disperse PNIPAM samples made by ATRP. Cloud points determined by turbidimetry on 1 wt % solutions; heating rate = 0.2 °C/min.[45]

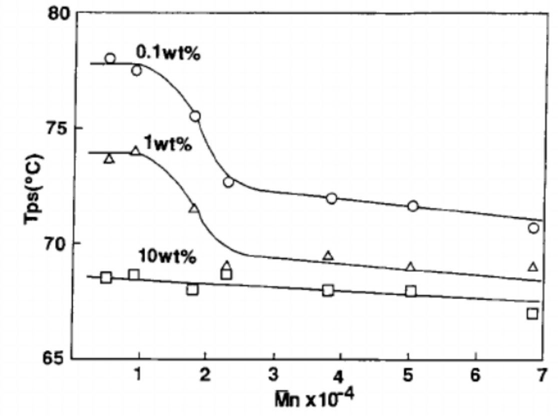


Figure 1-6 Dependance of number-average molecular weight of polymerized (2-methoxyethoxy)ethene with a narrow molecular weight distribution on the phase transition temperature in aqueous solution in concentration of 0.1, 1, and 10wt%.[45]

1.4.3 The effect of initiator

Structure of polymerization initiator also affect the phase transition behavior. Stover *et al.* selected 2-chloropropionamide (CP), *N*-isopropyl-2-chloropropionamide (i-PrCP), ethyl 2-chloropropionate (ECP), and *N*-phenyl-2-chloropropionamide (PhCP) as initiators to prepare polymers with end groups of varying polarity, ranging from hydrophilic for CP to hydrophobic for PhCP (**Figure 1-7**).[46] **Figure 1-8** shows that the phase transition temperature of the polymers having each end group was different at the same molecular weight and inversely depended on molecular weight of the polymer. The hydrophilic initiator, CP, increased the phase transition temperature to 45.3 °C at 3 kDa, while the hydrophobic ECP and PhCP showed the phase transition temperature at 40.6 and 37.4 °C at 3 kDa, respectively. The effect of initiator was significant for the low molecular weight polymer.

The amount of enthalpy generated by the phase transition was also affected by the hydrophilicity of initiator, and the polymer prepared using a hydrophobic polymerization initiator showed a larger enthalpy. This may be because the hydrophobic end group promotes desolvation of the polymer above the phase transition temperature.

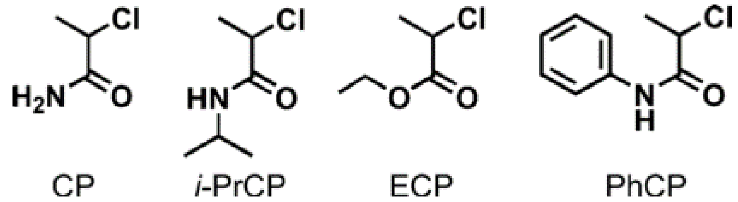


Figure 1-7 Chemical structures of 2-chloropropionamide (CP), isopropyl 2-chloropropionamide (*i*-PrCP), ethyl 2-chloropropionate (ECP), and phenyl 2-chloropropionamide (PhCP).[46]

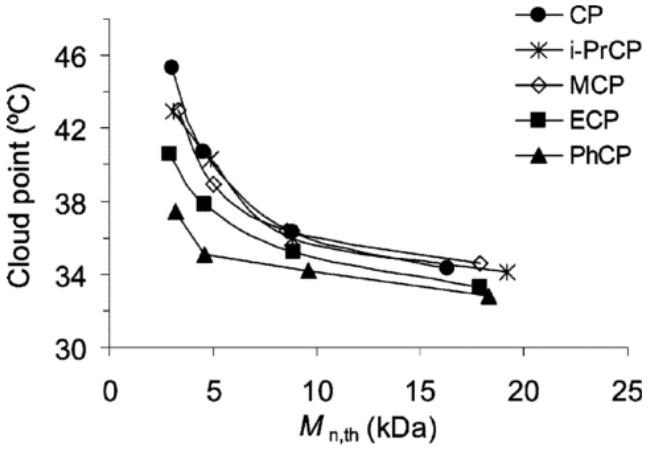


Figure 1-8 phase transition temperature (50%T) *vs* polymer molecular weight ($M_{\mathrm{n,th}}$) for narrow-disperse PNIPAM samples with different initiator made by ATRP.[46]

1.5 Applications

Polymers exhibiting LCST type phase transition with water have been proposed for application as functional materials in various fields, especially in the biomedical field. This section introduces examples of polymer and gel applications.

1.5.1 Separation and extraction

Kanazawa's group reported the use of a temperature-responsive polymer as a chromatographic carrier.[47–50] They modified the amino silica surface with a copolymer of NIPAM and BMA. The column packed with PNIPAM-modified silica showed drastic changes in retention of solutes with slight changes in column temperatures. The elution time of highly hydrophobic substances could be controlled by stepwise temperature gradient (**Figure 1-9**). Since the mobile phase does not require an organic solvent and only water was sufficient, it is possible to purify and analyze proteins, cells, and the like, which have not been possible using chromatography.

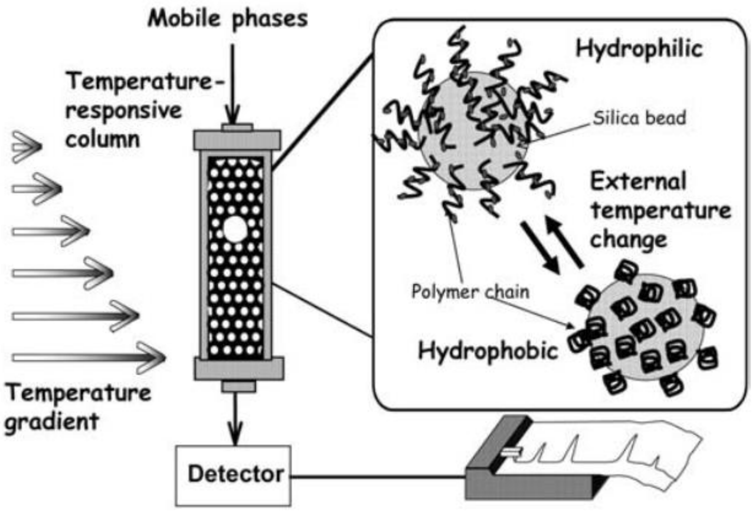


Figure 1-9 Schematic illustration of novel chromatography using a column packed with PNIPAM hydrogel-modified silica. [49]

Attempts have been made to use temperature-responsive polymers as part of a seawater desalination system. Reverse osmosis, currently the most commonly used method for plant-level desalination; RO) method, is a membrane separation technique in which solvents are moved by applying pressure above the osmotic pressure. On the other hand, the forward osmosis; FO) method is a method that utilizes the transfer of solvent according to the osmotic pressure between solutions and does not require the application of pressure. The high osmotic solvent used to separate water from seawater, sludge, and sewage by the FO method is called the driving solution. When the process for isolating pure water from the driving solution was carried out, pure water should be obtained at lower energy than the RO method because pressurization is not required.

Conventionally, high-concentration sugar solution, ammonium carbonate solution, superparamagnetic nanoparticle solution, salt solution, etc., have been used as the driving solution when the FO method is used for seawater desalination. Studies using polyelectrolyte solutions and ILs have been carried out, but all still have many improvements. Wang *et al.* attempted to extract pure water using a semi-IPN gel consisting of a polyelectrolyte and a temperature-responsive polymer as a driving force (**Figure 1-10**).[51–53] In this system, dry particulate gels are fixed on the surface of the semipermeable membrane, and the gel absorbs water toward swelling. However, at present, since the osmotic pressure of seawater is enormous compared with that of gel, it is almost impossible to recover water, and accordingly further improvement is required.

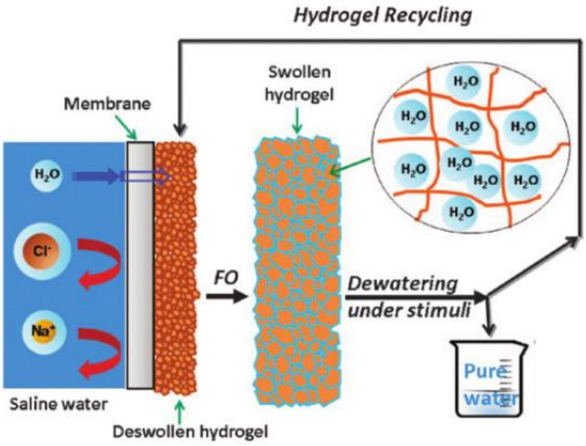


Figure 1-10 Schematic representation of the polymer hydrogel-forward osmosis desalination process.[52]

1.5.2 Tissue engineering

Okano *et al.* grafted PNIPAM onto surfaces of commercial polystyrene dishes in which cells were cultured and adhered to and recovered cultured cells by changing temperature.[54–56] Above the LCST, the surface of the culture dish becomes hydrophobic, and the cells adhere to each other through cell adhesion proteins, as in the case of a conventional culture dish, so that seeded cells can be cultured. When the temperature was below the LCST, the surface of the culture dish became hydrophilic so that the cells were quickly recovered by peeling off the adhesive proteins. Desorption and recovery of cultured cells are generally carried out by treatment with trypsin, dispase, calcium chelating agent, etc. In these cases, adhesive proteins and membrane proteins were decomposed, and accordingly cell function deteriorates. However, the LCST-system can be recovered without impairing proteins and function of the cell.

When the cells are grown on a monolayer and then subjected to low-temperature treatment, all the cells can be recovered as a cell sheet connected by adhesion between the cells.[55,56] It is impossible to obtain such a monolayer cell sheet because conventional methods such as tryptosine treatment disrupt cell-cell adhesion. The resulting cell sheet, in which almost all the cells remain alive, functions similarly to the medium they were transferred. So far, this has already been applied to vascular endothelial cell sheets[55,57], epidermal cell sheets[58], renal epithelial cell sheets[59,60], myocardial cell sheets[61], etc., and some of them have already been applied. The use of cell sheets is considered adequate in that the cells, extracellular matrix, humoral factors, etc. contained in the sheets are manipulated as a very non-invasive single structural unit to enable the formation of layered tissue close to the living body.

1.5.2 Actuator

Since PNIPAM gels were reported by Tanaka *et al.* in 1984[62], gels consisting of temperature-responsive polymers have also been extensively studied. It is known that the rate of volume change of a polymer gel is proportional to the square of its size.[63] An actuator is designed to utilize the mechanical energy generated in the swelling/shrinking process of the gel. In addition, the polymer gel is an intelligent material with control functions, so it is a very effective material for use as a micro-actuator that does not require wiring to control. Accordingly, various applications are being promoted, such as microfluidic control using polymers that respond to temperature[64] and micro temperature/chemical sensors that convert changes in gel volume caused by temperature and pH into electrical signals.[65]

Figure 1-11 is an example of micro-lens-driven research using temperature-responsive hydrogels as a part of the perimeter of a liquid-liquid lens.[66] The temperature change affected the volume of the hydrogel around the liquid-liquid lens, which further changes the lens's curvature and the focal length. When the gel is swollen at low temperature, the focal length of the lens takes a negative value, and when heated to the phase transition temperature, the focal length takes a positive value (**Figure 1-11 d**).

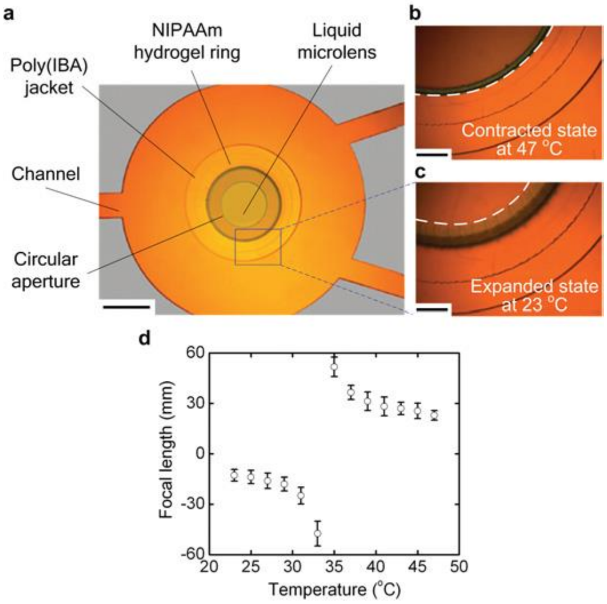


Figure 1-11 A smart temperature-sensitive liquid microlens using NIPAAm hydrogel. (a) An optical image of the device. Scale bar; 2.0 mm. The contracted (b) and expanded (c) states of the hydrogel ring. The dashed lines show the boundaries of the inside periphery of the hydrogel ring at two temperature states. Scale bars, 500 mm. (d) The focal length of the microlens as a function of temperature.[66]

1.5.3 Drug delivery

The coil-to-globule phase transition in the PNIPAM hydrogel rapidly reduces the volume of the gel, resulting in a fast release of an entrapped drug, subsequently causing more sustained, linear, and controlled release. Using a copolymer gel of PNIPAM, DMAAm, and BMA, Sakurai *et al.* achieved complete on-off control of the encapsulated indomethacin with temperature changes in between 36 °C and 38 °C.[67] The gel releases the drug at temperature below the LCST and ceases the drug release above the LCST to form a skin layer on the gel surface.

Okano *et al.* proposed device that release drugs at only high temperatures, such as fever, but no release was driven at average temperatures.[68] The device consists of a gel produced by copolymerizing PNIPAM with acrylamide, a hydrophilic monomer, and the skin layer is difficult to form on the gel surface. The gel is used in an impermeable capsule having a hole in a part. No drug release occurs when the gel is swollen at a low temperature. When the gel shrinks at high temperatures, space is created in the capsule, and the drug released from the gel diffuses through the pores of the capsule (**Figure 1-12**).

As described above, conventional thermoresponsive polymers, such as PNIPAM, have been developed for various applications. However, it has been difficult to control the phase transition temperature of PNIPAM while maintaining the sensitivity of its temperature response. Therefore, the available temperature of PNIPAM was limited to around 32 °C, and the use of the polymer in lower or higher temperature range was difficult. In order to utilize thermoresponsive polymers in various situations, a new polymer that can control the phase transition temperature at the desired temperature with a sharp temperature response has been required.

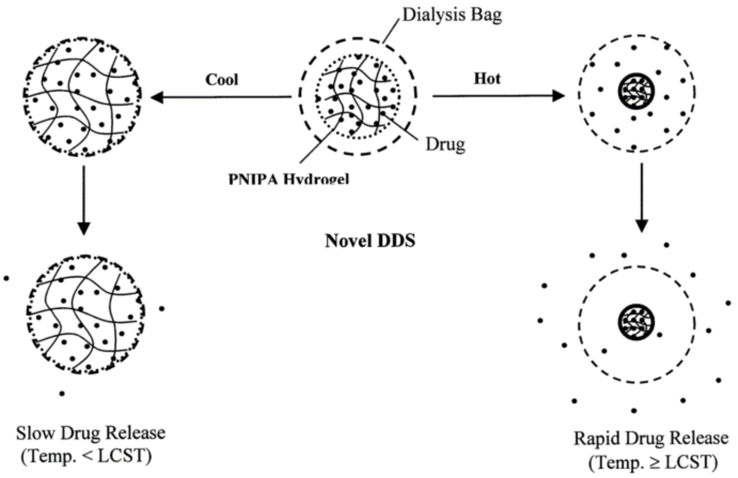


Figure 1-12 Schematic illustrations of the novel, thermo-responsive drug delivery system to give a positive drug release by modulating the external temperature.[68]

1.6 Poly(ionic liquids) showing LCST-type-phase transition with water

1.6.1 Ionic liquids and poly(ionic liquid)s

Ionic liquids (**ILs**) [69] are organic salts with very low melting points, and have attracted a keen attention as a new type of liquid-state materials due to their unique physicochemical properties such as negligibly low vapor pressure, liquid state in the wide range of temperature, and high charge density. Another important feature of ILs is that the physicochemical properties can be freely designed by selecting anions and cations. For this reason, ILs with various constituent ions have been produced in order to obtain desired physical properties. Early ILs were unstable in air, but in 1992, Wilkes *et al.* first reported an IL with 1-ethyl-3-methylimidazolium cation that was stable in air and water and had a melting point below room temperature.[70] After this discovery, the number of studies on ILs has increased continuously, and research area in various fields has been spread. The variety of ILs reported today is so great that the only feature that can comprehensively describe all of them is that they are salts that melt at room temperature and are composed entirely of ions.

In 1998, Ohno's group reported for the first time an IL-based polyelectrolyte referred to as polymerized ionic liquids poly(ionic liquids), **PILs**, containing a repeating unit of IL in the polymer chain.[71] Following this report, the functionalization of polymers composed of ILs has been studied in establishing a new classification of polyelectrolytes and in a manner termed PIL. PILs have been designed for various applications such as solid polymer electrolytes[72,73], lithium batteries[74,75], solar cells,[76,77] electrochromic devices[78], however most of the research is conducted especially for solid-state applications. The most PIL is simple polymers obtained from monomers having polymerizable group on a cation or an anion.

1.6.2 IL/water mixture showing LCST-type phase transition

The first report of an IL exhibiting the LCST type phase transition with water was presented by Ohno *et al.*. The temperature responsiveness is achieved by chemically modified amino acid as a component ion so as to bring the hydrophilicity to an appropriate range (**Figure 1-13**).[79] **Figure 1-14** shows the phase behavior of the tetrabutylphosphonium *N*-trifluoromethanesulfonyl leucine/water mixture, in which ILs are colored with red pigment. The solution has a liquid-liquid phase separation at 25.0 °C.

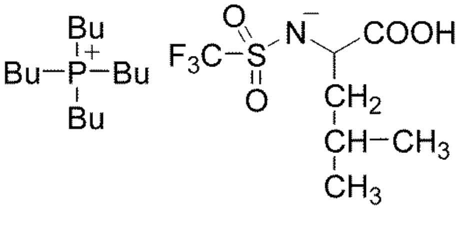


Figure 1-13 Structure of tetrabutylphosphonium *N*-trifluoromethanesulfonyl leucine.[79]

The IL phase gradually increased upon cooling the solution and became a uniform liquid phase at 22.0 °C. Then, when the solution was heated again, it became turbid at once and gradually separated into an aqueous and an IL phase. This quite sharp temperature dependence has never been found in other organic molecules /water mixtures.

Since the IL has an amino acid structure as an anion, the phase separation temperature can be easily controlled by changing the side chain structure. For example, a hydrophilic amino acid causes a phase transition at a higher temperature, whereas a hydrophobic amino acid gives a lower phase transition temperature (**Figure 1-15**). A similar tendency was observed when the hydrophobicity of the cations was changed (see **Figure 1-15** □ and ■). As the increase in the carbon number of one alkyl chain on the cation from 4 to 8, the transition temperature decreased.

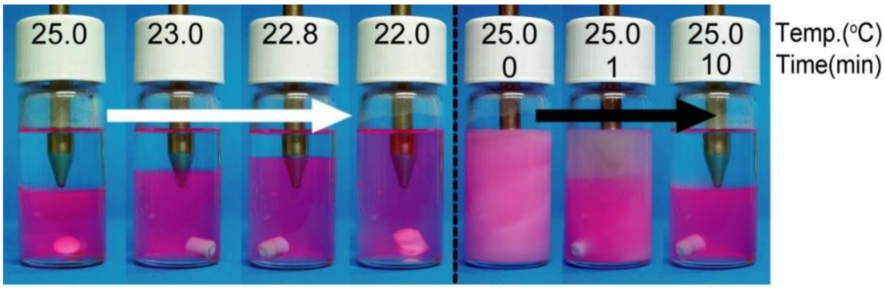


Figure 1-14 Temperature-dependent phase transition of IL/water mixture.[79]

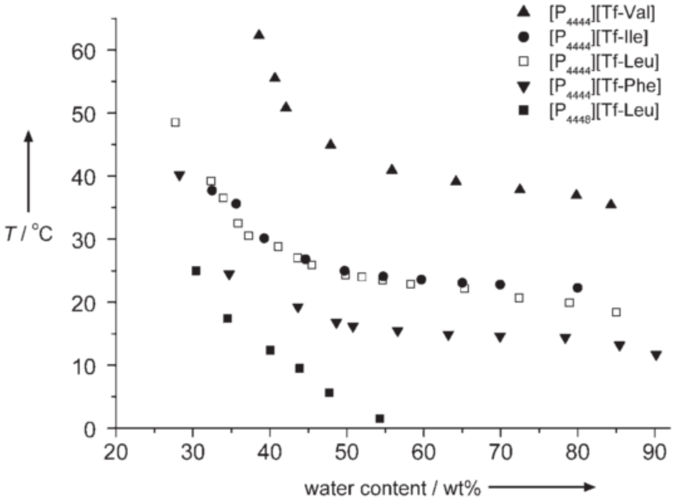


Figure 1-15 Phase-separation temperature versus water content.[79]

Ohno *et al.* also reported the phase behavior of ILs composed of various constituent ions. ILs undergoing LCST-type phase transitions with water were found in the region between hydrophobic and hydrophilic ion pairs.[80] $[P_{4444}]$ cation was coupled with several different anions to examine and the phase behavior of the IL/water mixture containing 50 wt% water. ILs having hydrophilic anions such as $[CH_3SO]^-$, Cl^-, and Br^- were always miscible with water regardless of temperatures. On the other hand, ILs containing hydrophobic anions such as $[Tf_2N]^-$ and $[BF_4]^-$ were phase-separated from water. ILs with anions having moderate hydrophobicity such as $[CF_3COO]^-$ exhibited the LCST-type phase transition. When the phase transition behavior of ILs having different cations was similarly evaluated, ILs having moderate hydrophobicity underwent the LCST type phase transition with water. These results indicate that the LCST-type phase transition should be found only when ILs with moderate hydrophobicity were used. In other words, ILs designed with adequate hydrophilicity have the potential to show the LCST-type phase transition with water. In order to quantify the required range of hydrophilicity showing the LCST-type phase transition, the amount of dissolved water in the separated IL-rich phase was measured at 60 °C and calculated the amount of water per ion pair, which is referred to as hydrophobicity index (HI).[81] They proposed that relatively hydrophobic ILs with the HI above 7 had the potential to show LCST-type phase transition with water.

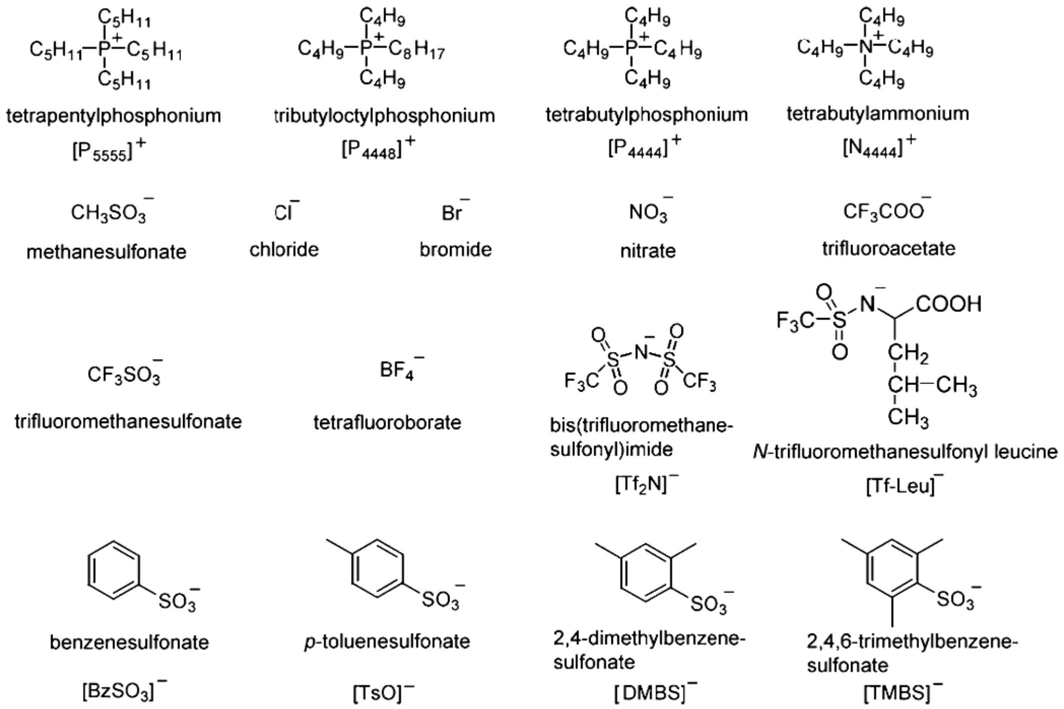
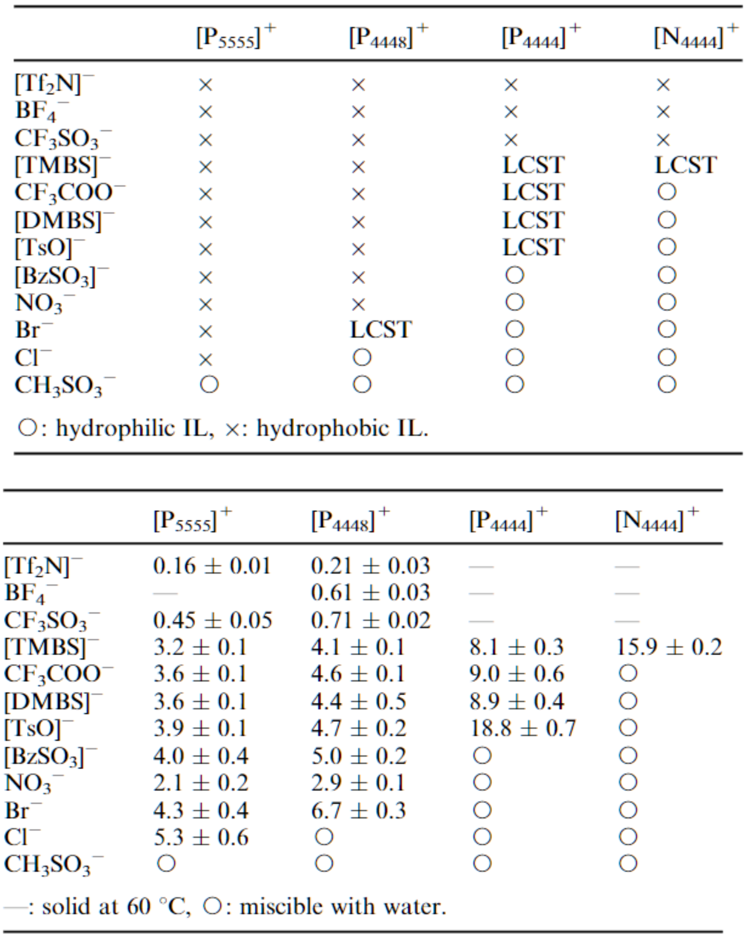


Figure 1-16 Component ions used to prepare ILs.[79]

Table1-2 Phase behavior of ILs mixed with an equal amount of water (above) and Hydrophilicity index (HI) of each ILs (below).[81]



	$[P_{5555}]^+$	$[P_{4448}]^+$	$[P_{4444}]^+$	$[N_{4444}]^+$
$[Tf_2N]^-$	×	×	×	×
BF_4^-	×	×	×	×
$CF_3SO_3^-$	×	×	×	×
$[TMBS]^-$	×	×	LCST	LCST
CF_3COO^-	×	×	LCST	○
$[DMBS]^-$	×	×	LCST	○
$[TsO]^-$	×	×	LCST	○
$[BzSO_3]^-$	×	×	○	○
NO_3^-	×	×	○	○
Br^-	×	LCST	○	○
Cl^-	×	○	○	○
$CH_3SO_3^-$	○	○	○	○

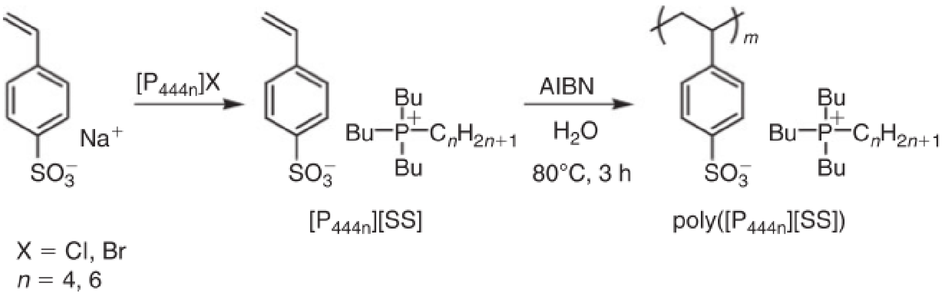
○: hydrophilic IL, ×: hydrophobic IL.

	$[P_{5555}]^+$	$[P_{4448}]^+$	$[P_{4444}]^+$	$[N_{4444}]^+$
$[Tf_2N]^-$	0.16 ± 0.01	0.21 ± 0.03	—	—
BF_4^-	—	0.61 ± 0.03	—	—
$CF_3SO_3^-$	0.45 ± 0.05	0.71 ± 0.02	—	—
$[TMBS]^-$	3.2 ± 0.1	4.1 ± 0.1	8.1 ± 0.3	15.9 ± 0.2
CF_3COO^-	3.6 ± 0.1	4.6 ± 0.1	9.0 ± 0.6	○
$[DMBS]^-$	3.6 ± 0.1	4.4 ± 0.5	8.9 ± 0.4	○
$[TsO]^-$	3.9 ± 0.1	4.7 ± 0.2	18.8 ± 0.7	○
$[BzSO_3]^-$	4.0 ± 0.4	5.0 ± 0.2	○	○
NO_3^-	2.1 ± 0.2	2.9 ± 0.1	○	○
Br^-	4.3 ± 0.4	6.7 ± 0.3	○	○
Cl^-	5.3 ± 0.6	○	○	○
$CH_3SO_3^-$	○	○	○	○

—: solid at 60 °C, ○: miscible with water.

1.6.3 PIL/water mixture showing LCST-type phase transition

After finding adequate HI of IL showing LCST-type phase transition, Kohno *et al.* successfully prepared a few IL monomers having vinyl groups and polymerized them to obtain PILs (see **scheme 1-1**).[82] The reported poly(tetrabutylphosphonium styrenesulfonate) (poly($[P_{4444}]$[SS])) was a polyanionic type PIL. The transmittance of the aqueous solution containing 10 wt% of this PIL decreased to 90% upon heating to 57 °C (**Figure 1-17**; black line). In addition, copolymers of $[P_{4446}]$[SS]) with different hydrophobic IL monomers, also exhibit the LCST-type phase transitions (**Figure 1-17**; blue line). This shows that the transition temperature of PILs can also be controlled by the total hydrophobicity.



Scheme 1-1 Preparation of $[P_{444n}][SS]$ and its polymers.[82]

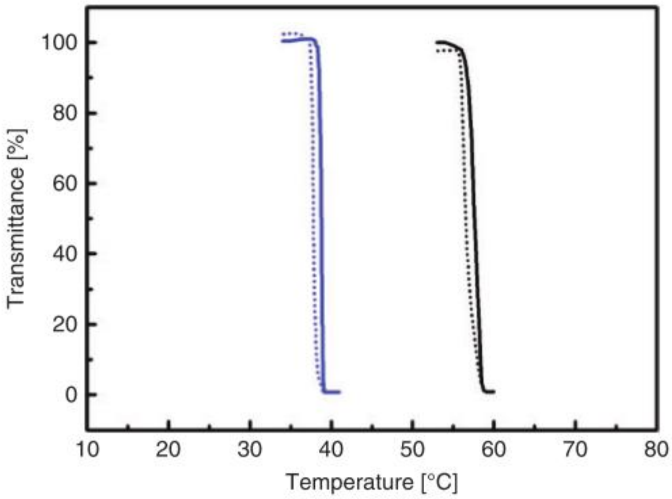


Figure 1-17 Turbidity change of an aqueous solutions containing 10 wt% poly($[P_{4444}][SS]$) (black line) and poly($[P_{4444}][SS]_{0.7}$-*co*-$[P_{4446}][SS]_{0.3}$; molar ratio of $[P_{4444}][SS]$ to $[P_{4446}][SS]$ was 7.0 to 3.0) (blue line); solid line: heating process, dotted line: cooling process.[82]

1.6.4 The phase transition behavior of thermoresponsive IL and PIL in water

The phase transition mechanism of the thermoresponsive IL and PIL has been investigated in various methods, a comprehensive explanation has not been obtained yet. Wang *et al.* analyzed the behavior of thermoresponsive IL, $[P_{4444}][CF_3COO]$, in water by DLS, ^{1}H NMR, and freeze-fracture transmission electron microscopy in order to explore the phase transition mechanism.[83] The size and morphology of the IL aggregates were found to change upon heating even under the phase transition temperature. **Figure 1-18** shows that all systems demonstrated a similar variation tendency for any concentration; the aggregate size increased gradually with increasing temperature and then increased rapidly as the temperature approached the phase transition temperature. The concentration of IL also affected the aggregate size. For example, when the temperature was fixed at 298 K, upon increasing the concentration from 20 to 25 and then 40 wt%, the aggregate size was increased from 5.3 to 10.7 and further to 30.5 nm. They considered that the size increases with increasing the concentration of IL because IL molecules form self-assembled aggregates in water as the water content of IL increases. On the other hand, if the size decreases as the IL concentration is further increased, water dissolved into the IL and forms liquid-like associated aggregates.

The aggregates size change of thermoresponsive IL, tetrabutylphosphonium 2,4-dimethylbenzene sulfonate ([P_{4444}][DMBS]), in water over a wide temperature range was reported by Kang *et al.*[84] They found that the size of aggregates changes rapidly around the phase transition temperature and that aggregates exist in both the aqueous and ionic liquid-rich phases. The ionic liquid rich phase shows aggregates on the order of 500 nm at 37 °C, just above the phase transition temperature (**Figure 1-19**). The size of aggregates in the IL-rich phase subsequently decrease with

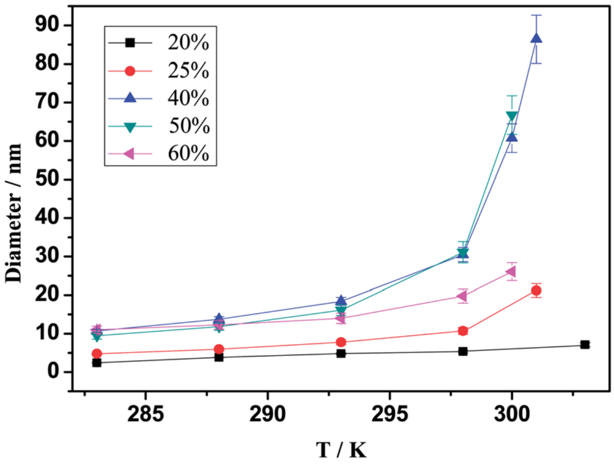


Figure 1-18 Variation of the size of [P_{4444}][CF_3COO] aggregates with temperature in solutions containing different amounts of [P_{4444}][CF_3COO]. [83]

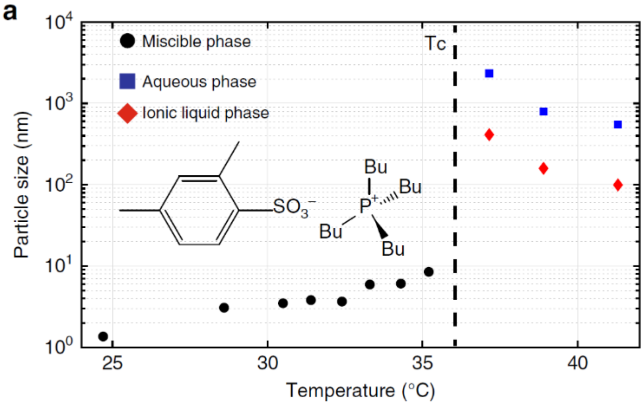


Figure 1-19 Particle size vs temperature of [P_{4444}][DMBS]/H_2O 50 wt.% over the fully miscible region and phase separated regions of the phase diagram. Phase separation temperature, T_c ca. 36 °C. [84]

increasing temperature to 150 nm at 39 °C and then to 100 nm at 41 °C. The initial 50-fold increase in IL aggregate size going from the miscible phase to IL rich phase would likely arise from the sudden decrease in water concentration and additionally the reduced electrostatic screening from water enabling larger aggregates to form. The aqueous-rich phase shows nearly an order of magnitude increase scattering diameter of 3000 nm compared to the IL-rich phase just above phase separation.

Wu *et al.* have been analyzed phase transition behavior of some ILs and PILs with DSC, turbidity method, temperature-variable ^{1}H-NMR, and FT-IR spectroscopy measurements in combination with perturbation correlation moving window and two-dimensional correlation analysis.[85,86] They revealed that the dehydration of the alkyl chain of the [P_{4444}] cation occurred initially in monomer systems upon heating then water molecules would be excluded out to the temperature response behavior between IL/water mixture and PIL/water mixture arises from the periphery. Following dehydration of cations, the hydration interaction and hydrogen bond between SO_3^- group and water molecular begin to break. Regarding poly([P_{4444}][SS])/water mixture, on the other hand, SO_3^- groups of the polyanion firstly dehydrated upon heating. The difference of hydrophobic backbone contained in the polyanion. As for to tributylhexylphosphonium 3-sulfopropyl methacrylate ([P_{4446}][MC3S]), on the other hand, the dehydration of ester group of (poly)anion first occurred upon heating both in IL/water mixture and in PIL/water mixture.

Thus, various methods have been used to analyze the phase transition behavior. However, these methods have not yet given the point where the phase transition behavior can be controlled by the hydrophilic/hydrophobic control of the whole system by mixing PIL, and further development is requested in the future.

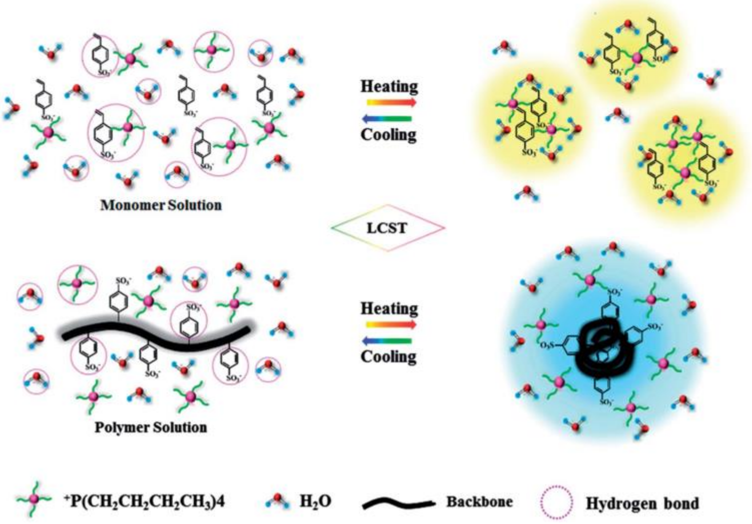


Figure 1-20 Schematic illustration of the dynamic phase transition mechanism of [P_{4444}][SS] and poly([P_{4444}][SS]) solutions during the heating and cooling processes.[86]

1.6.5 Applications of thermoresponsive PILs

Following my work of PIL showing LCST-type phase transition in water described in Chapter 2 to 4, many study using thermoresponsive PIL/water mixture or PIL hydrogel have been carried out until today. Most of them are applications of the PILs the author prepared, and there are few reports of novel thermoresponsive PIL undergoing LCST-type phase transition with water.

The forward osmosis (FO) applications are one of the most active field of use of thermoresponsive PIL. Lee *et al.* compared the performance of [P_{4444}][SS] and oligomeric [P_{4444}][SS] as draw solutes.[87] Although the water permeation flux values in the FO system using the oligomer as a draw solute were slightly smaller than those using the monomer, the reverse solute flux values using the oligomer were found to be much smaller than those using the monomer, indicating that the oligomers are more efficient draw solute materials in the FO system than the monomer. The thermoresponsive PIL hydrogel was also used as draw agent of desalination system. Liu *et al.* used the PIL hydrogels prepared with tributyl-4-vinylbenzylphosphonium-based IL monomers with an alkyl sulfonate counterion. [88] This PIL hydrogel had the highest water content and the highest recovery rate, and thus had the highest desalination efficiency compared to conventional gels used as draw agents. They insisted that the excellent desalination properties of PIL hydrogel make it suitable for the permanent production of demineralized water. Urban *et al.* combined IL, 2,4-dimethylbenzensulfonate, and [P_{4444}][SS] hydrogel in order to obtain more sustained driving force in FO systems.[89] When the IL/water mixture was used as a draw solute, the water flux gradually decreased due to the decrease in osmotic pressure caused by the water absorbed from the NaCl solution. Once PIL gel was added to

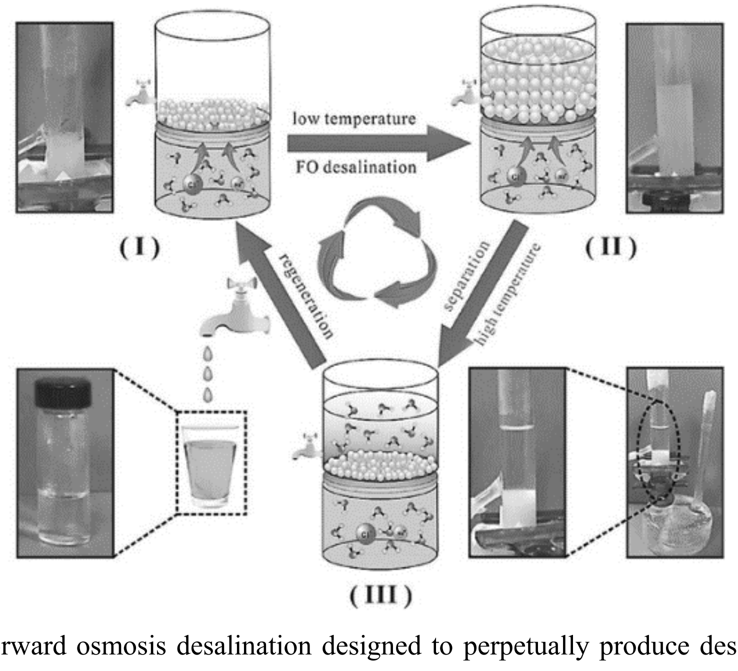


Figure 1-21 A forward osmosis desalination designed to perpetually produce desalinated water by cyclic drawing–dewatering of the PIL hydrogel.[89]

the solution, the decrease in osmotic pressure of the solution was suppressed by the PIL gel swelling with water absorbed into the solution, and the solution showed continuous water absorption ability. They concluded that the synergistic effect of using the IL/hydrogel binary draw solute system has been demonstrated in terms of creating a more durable draw solute with less active materials, the water recovered from the hydrogel contains less solutes compared to direct phase separation of IL solutions, and the hydrogels are in a mobile phase which makes a continuous FO process possible.

Besides FO applications, Chen *et al* used poly([P_{4444}][SS]) as the shell of a hybrid drug nanocarrier developed with $CuCo_2S_4$ nanoparticles as the core (**Figure 1-22**).[90] pH- and thermal-responsive characteristics of poly([P_{4444}][SS]) endow the multifunctional hybrid nanocarrier system Doxorubicin-$CuCo_2S_4$/PIL with sensitive dual-stimuli-triggered drug release behaviors. In this system, instead of heating the solution, the $CuCo_2S_4$ core converts near-infrared radiation into thermal energy, which induces shrinkage of the PIL shell. The nanocarrier achieved favorable controlled release of 90.5% under pH/thermo dual stimuli.

Yuan *et al.* utilized the PIL as the stabilizer of graphene.[91] The conventional PILs are known to disperse graphene and metal nanoparticles. [92,93] It was considered that the stabilization was designed by the interaction between the imidazolium ring of the cations and nanostructure surface. However, poly([P_{4444}][SS]) containing no imidazolium rings also dispersed graphene in water. **Figure 1-23A** shows the transmittance change of the solution. An IL/water mixture was further mixed with graphene, sonicated, and left standing for 24 hours. The resulting solution is uniformly black, indicating that graphene was stably dispersed. Upon heating the solution, poly([P_{4444}][SS]) was aggregate with graphene. Upon heating the solution above 61 °C, PIL underwent a phase transition to aggregate graphene. They also found that the aggregation state of graphene in the solution could be controlled

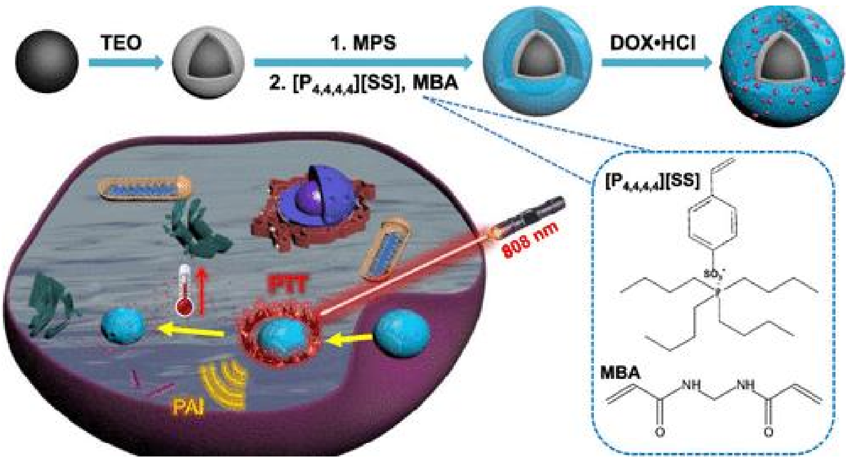


Figure 1-22 A schematic diagram of hybrid drug nanocarrier with $CuCo_2S_4$ nanoparticles as the core and Poly[P_{4444}][SS] as the shell. [90]

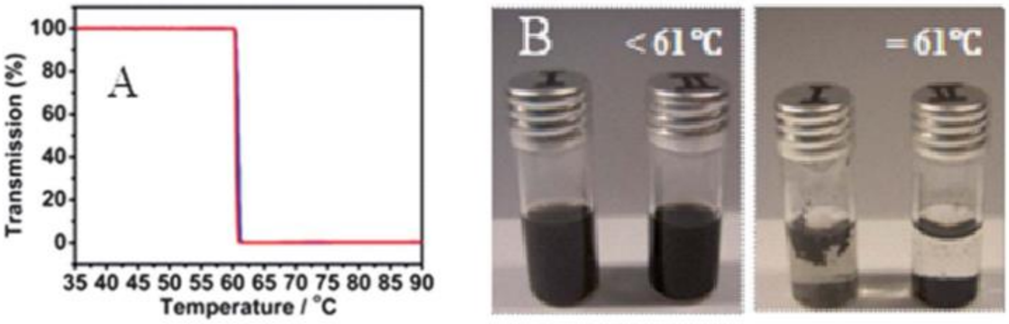


Figure 1-23 (A) Turbidity curves of aqueous solution of poly([P_{4444}][SS]) (red line) without added salt and poly([P_{4444}][SS]) (black line) with [P_{4444}]Br salt. (B) Precipitation of poly([P_{4444}][SS])-stabilized reduced graphene in the aforementioned two aqueous solutions below and at 61 °C.[91]

by the added salt. When adding [P_{4444}]Br to the solution, the graphene formed floated aggregates above the phase transition temperature (**Figure 1-23** B left). On the other hand, when [P_{4444}]Br was not added to the solution, the graphene formed precipitated aggregates above the phase transition temperature (**Figure 1-23** B right).

Diamond *et al.* have developed PIL hydrogels prepared by [P_{4446}][MC3S] as polymer actuators in microfluidic systems (**Figure 1-24**).[94] They found that the results exhibited by the IL polymer flow controlactuator followed a sigmoidal fit. The characterization of the hydrogel indicates that the hydrogel shrinks by~58% of its swollen area when the temperature is raised from 20 to 70 °C and ~39% when the temperature is raised from 20 to 50 °C. Furthermore, the hydrogel exhibited repeatable shrink/reswell behavior when subjected to temperature cycles between 20 ◦C and 50 °C. In addition, by assembling the hydrogel in a microfluidic device as a temperature-controlled actuator, the microfluidic channel was tuned between ~110 nL/min and ~27 nL/min by varying the temperature between 50 °C and 20 °C. Diamond *et al.* also utilized direct laser writing method in order to prepare PIL hydrogel for actuator created from crosslinked [P_{4446}][MC3S] at sub-micron resolution.[95] The resulting hydrogel actuator showed a very fast response due to the small scale of the structure and could not be distinguished from the rate of temperature cycling.

There are also studies in which the PIL was used for separation and extraction materials. Noble *et al.* designed thermoresponsive membrane containing [P_{4444}][SS] for concentrating proteins from aqueous media (**Figure 1-25**).[96] The observed extraction efficiency value varies widely depending on both the protein species and the temperature. The extraction efficiency of cytochrome *c* exhibited by PIL membrane was highest than that of Myoglobin and Peroxidase, and the value increased as the increasing temperature to reach 99.7% at 30 °C. They explained that the mechanism of this selectivity result from the salting-out and salting-in profiles of the proteins.

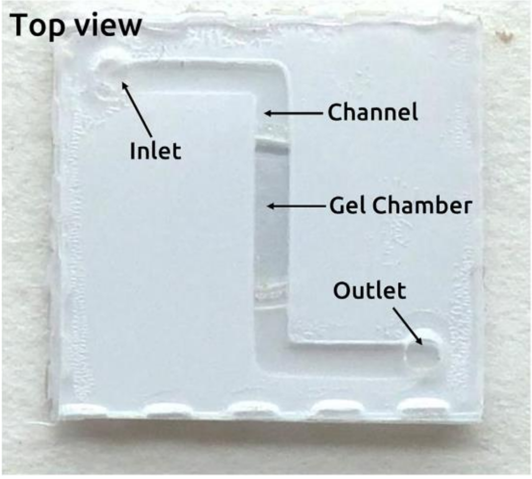


Figure 1-24 The assembled microfluidic chip containing the [P_{4446}][MC3S] hydrogel actuator. [95]

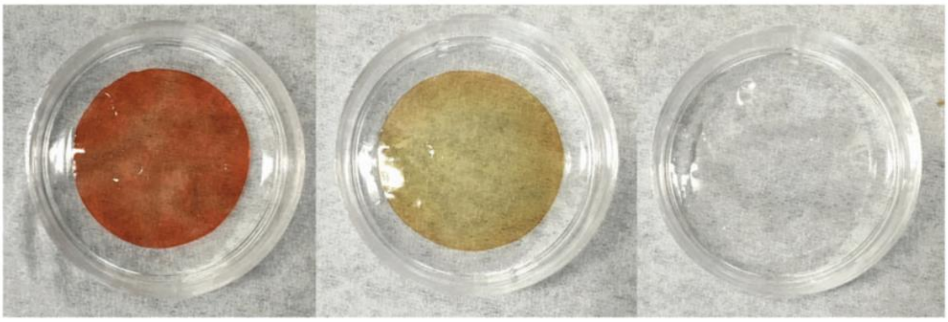


Figure 1-25 Photographs of PIL membrane after immersion in 0.1 mol L^{-1} aq. potassium phosphate buffer solution with the proteins cytochrome *c* from equine heart (left), myoglobin from equine heart (middle), and horseradish peroxidase (right) at 22 °C for 24 h. [96]

1.7 Aim of this study

The aim and objectives in this study is to design PIL/water mixture showing sharp LCST-type phase transition at desired temperature through physico-chemical analysis of required factors. It is also to design a fully charged thermo-responsive hydrogel composed of the PILs thus obtained. The design of component ions of IL monomer is first step to prepare thermoresponsive PIL, however, there is a limit to the adjustment of physical properties of PIL through the design of ionic structures only. Then, the author investigated some factor to control the phase transition temperature such as PIL concentration, additive salt, copolymerization.

Although there are many conventional non-ionic thermoresponsive polymers, it has been difficult to control the phase transition temperature with maintaining temperature sensitivity. Hence, there was a limit to usable temperature range of the polymers. On the other hand, it has already been reported that the phase transition temperature of a temperature-responsive IL/water mixture can be controlled by the hydrophobicity of component ions of IL, and the temperature responsiveness remains sharp. Based on this knowledge, the authors consider that by polymerizing an IL monomer and producing a new class of temperature-responsive polymer, a temperature-responsive polymer that overcomes problems of conventional nonionic temperature-responsive polymers can be provided.

Both structure of the IL monomers and the method to adjust the phase transition temperature of the PIL in water are discussed here. In addition, the hydrogel using IL monomer was prepared, and its thermoresponsive behavior was investigated.

1.8 References

1) A. Gandhi, A. Paul, S. O. Sen, and K. K. Sen,. "Studies on thermoresponsive polymers: Phase behaviour, drug delivery and biomedical applications",. *Asian Journal of Pharmaceutical Sciences*, **2015**, 10, 99–107.

2) H. G. Schild,. "Poly(*N*-isopropylacrylamide): experiment, theory and application",. *Progress in Polymer Science*, **1992**, 17, 163–249.

3) M. Shibayama, T. Norisuye, and S. Nomura,. "Cross-link Density Dependence of Spatial Inhomogeneities and Dynamic Fluctuations of Poly(*N*-isopropylacrylamide) Gels",. *Macromolecules*, **1996**, 29, 8746–8750.

4) I. Idziak, D. Avoce, D. Lessard, D. Gravel, and X. X. Zhu,. "Thermosensitivity of Aqueous Solutions of Poly(*N*,*N*-diethylacrylamide)",. *Macromolecules*, **1999**, 32, 1260–1263.

5) R. A. Horne, J. P. Almeida, A. F. Day, and N. T. Yu,. "Macromolecule hydration and the effect of solutes on the cloud point of aqueous solutions of polyvinyl methyl ether: A possible model for protein denaturation and temperature control in homeothermic animals",. *Journal of Colloid and Interface Science*, **1971**, 35, 77–84.

6) K. V. Durme, S. Verbrugghe, F. E. D. Prez, and B. V. Mele,. "Influence of Poly(ethylene oxide) Grafts on Kinetics of LCST Behavior in Aqueous Poly(*N*-vinylcaprolactam) Solutions and Networks Studied by Modulated Temperature DSC",. *Macromolecules*, **2004**, 37, 1054–1061.

7) E. E. Makhaeva, H. Tenhu, and A. R. Khokhlov,. "Conformational Changes of Poly(vinylcaprolactam) Macromolecules and Their Complexes with Ionic Surfactants in Aqueous Solution",. *Macromolecules*, **1998**, 31, 6112–6118.

8) M. Heskins, and J. E. Guillet,. "Solution Properties of Poly(*N*-isopropylacrylamide)",. *Journal of Macromolecular Science: Part A - Chemistry*, **1968**, 2, 1441–1455.

9) Y. Xia, X. Yin, N. A. D. Burke, and H. D. H. Sto,. "Thermal Response of Narrow-Disperse Poly (*N* -isopropylacrylamide) Prepared by Atom Transfer Radical Polymerization",. *Macromolecules*, **2005**, 38, 5937–5943.

10) C. M. Schilli, A. H. E. Muller, E. Rizzardo, S. H. Thang and Y. K. Chong, "Advances in Controlled/Living Radical Polymerization", *ACS symp. Ser.*, **2003**, 854, 603-618.

11) B. Ray, Y. Okamoto, M. Kamigaito, M. Sawamoto, K. Seno, S. Kanaoka, and S. Aoshima,. "Effect of Tacticity of Poly(*N*-isopropylacrylamide) on the Phase Separation Temperature of Its Aqueous Solutions",. *Polymer Journal*, **2005**, 37, 234–237.

12) Y. Ding, X. Ye, and G. Zhang,. "Microcalorimetric Investigation on Aggregation and Dissolution of Poly(*N* -isopropylacrylamide) Chains in Water",. *Macromolecules*, **2005**, 38, 904–908.

13) P. M. Reddy, and P. Venkatesu,. "Ionic liquid modifies the lower critical solution temperature (LCST) of poly(*N*-isopropylacrylamide) in aqueous solution.",. *The journal of physical chemistry. B*, **2011**, 115, 4752–4757.

14) J. Heyda, S. Soll, J. Yuan, and J. Dzubiella,. "Thermodynamic Description of the LCST of Charged Thermoresponsive Copolymers",. *Macromolecules*, **2014**, 47(6), 2096-2102.

15) E. I. Tiktopulo, V. E. Bychkova, J. Ricka, and O. B. Ptitsyn,. "Cooperativity of the Coil-Globule Transition in a Homopolymer: Microcalorimetric Study of Poly(*N*-isopropylacrylamide)",. *Macromolecules*, **1994**, 27, 2879–2882.

16) J. Dybal, M. Trchová, and P. Schmidt,. "The role of water in structural changes of poly(*N*-isopropylacrylamide) and poly(*N*-isopropylmethacrylamide) studied by FTIR, Raman spectroscopy and quantum chemical calculations",. *Vibrational Spectroscopy*, **2009**, 51, 44–51.

17) P. V. Yushmanov, I. Furó, and I. Iliopoulos,. "Kinetics of Demixing and Remixing Transitions in Aqueous Solutions of Poly(*N*-isopropylacrylamide): A Temperature-Jump ^{1}H NMR Study",. *Macromolecular Chemistry and Physics*, **2006**, 207, 1972–1979.

18) K. Van Durme, L. Delellio, E. Kudryashov, V. Buckin, and B. Van Mele,. "Exploration of high-resolution ultrasonic spectroscopy as an analytical tool to study demixing and remixing in poly(*N*-isopropyl acrylamide)/water solutions",. *Journal of Polymer Science Part B: Polymer Physics*, **2005**, 43, 1283–1295.

19) Y. Okada, and F. Tanaka,. "Cooperative Hydration, Chain Collapse, and Flat LCST Behavior in Aqueous Poly(*N* -isopropylacrylamide) Solutions",. *Macromolecules*, **2005**, 38, 4465–4471.

20) H. Cheng, L. Shen, and C. Wu,. "LLS and FTIR Studies on the Hysteresis in Association and Dissociation of Poly(*N*- isopropylacrylamide) Chains in Water",. *Macromolecules*, **2006**, 39, 2325–2329.

21) H. Inoue, K. Katayama, K. Iwai, A. Miura, and H. Masuhara,. "Conformational relaxation dynamics of a poly(*N*-isopropylacrylamide) aqueous solution measured using the laser temperature jump transient grating method.",. *Physical chemistry chemical physics* , **2012**, 14, 5620–5627.

22) I. Nishio, S.-T. Sun, G. Swislow, and T. Tanaka,. "First observation of the coil–globule transition in a single polymer chain",. *Nature*, **1979**, 281, 208–209.

23) B. Sun, Y. Lin, P. Wu, and H. W. Siesler,. "A FTIR and 2D-IR Spectroscopic Study on the Microdynamics Phase Separation Mechanism of the Poly(*N* -isopropylacrylamide) Aqueous Solution",. *Macromolecules*, **2008**, 41, 1512–1520.

24) Y. Katsumoto, T. Tanaka, and Y. Ozaki,. "Relationship between the coil-globule transition of an aqueous poly(*N*-isopropylacrylamide) solution and structural changes in local conformations of the polymer",. *Macromolecular Symposia*, **2004**, 205, 209–224.

25) Y. Katsumoto, T. Tanaka, H. Sato, and Y. Ozaki,. "Conformational Change of Poly(*N* -isopropylacrylamide) during the Coil−Globule Transition Investigated by Attenuated Total Reflection/Infrared Spectroscopy and Density Functional Theory Calculation",. *The Journal of Physical Chemistry A*, **2002**, 106, 3429–3435.

26) Y. Maeda,. "IR Spectroscopic Study on the Hydration and the Phase Transition of Poly(vinyl methyl ether) in Water",. *Langmuir*, **2001**, 17, 1737–1742.

27) Y. Maeda, T. Nakamura, and I. Ikeda,. "Changes in the Hydration States of Poly(*N*-alkylacrylamide)s during Their Phase Transitions in Water Observed by FTIR Spectroscopy",. *Macromolecules*, **2001**, 34, 1391–1399.

28) K. Otake, H. Inomata, M. Konno, and S. Saito,. "Thermal analysis of the volume phase transition with N-isopropylacrylamide gels",. *Macromolecules*, **1990**, 23, 283–289.

29) E. C. Cho, J. Lee, and K. Cho,. "Role of Bound Water and Hydrophobic Interaction in Phase Transition of Poly(*N* -isopropylacrylamide) Aqueous Solution",. *Macromolecules*, **2003**, 36, 9929–9934.

30) Y. Tsuboi, T. Tada, T. Shoji, and N. Kitamura,. "Phase-Separation Dynamics of Aqueous Poly (*N*-isopropylacrylamide) Solutions: Characteristic Behavior of the Molecular Weight and Concentration Dependences",. *Macromolecular Chemistry and Physics*, **2012**, 213, 1879–1884.

31) T. Tada, T. Hirano, K. Ute, Y. Katsumoto, T.-A. Asoh, T. Shoji, N. Kitamura, and Y. Tsuboi,. "Effects of Syndiotacticity on the Dynamic and Static Phase Separation Properties of Poly(*N*-isopropylacrylamide) in Aqueous Solution",. *Journal of Physical Chemistry B*, **2016**, 120, 7724–

7730.

32) M. Matsumoto, T. Tada, T.-A. Asoh, T. Shoji, T. Nishiyama, H. Horibe, Y. Katsumoto, and Y. Tsuboi,. "Dynamics of the Phase Separation in a Thermoresponsive Polymer: Accelerated Phase Separation of Stereocontrolled Poly(*N*, *N*-diethylacrylamide) in Water",. *Langmuir*, **2018**, 34, 13690–13696.

33) T. Tanaka, E. Sato, Y. Hirokawa, S. Hirotsu, and J. Peetermans,. "Critical Kinetics of Volume Phase Transition of Gels",. *Physical Review Letters*, **1985**, 55, 2455–2458.

34) Y. Pei, J. Chen, L. Yang, L. Shi, Q. Tao, B. Hui, and J. Li,. "The effect of pH on the LCST of poly(*N*-isopropylacrylamide) and poly(*N*-isopropylacrylamide-co-acrylic acid)",. *Journal of Biomaterials Science, Polymer Edition,* **2004**, 15(5), 585-594.

35) Y. Zhang, K. Chen, Y. Li, J. Lan, B. Yan, L. Shi, and R. Ran,. "High-Strength, Self-Healable, Temperature-Sensitive, MXene-Containing Composite Hydrogel as a Smart Compression Sensor",. *ACS Applied Materials & Interfaces*, **2019**, 11, 47350–47357.

36) C. Teng, J. Qiao, J. Wang, L. Jiang, and Y. Zhu,. "Hierarchical Layered Heterogeneous Graphene-poly(*N*-isopropylacrylamide)-clay Hydrogels with Superior Modulus, Strength, and Toughness",. *ACS Nano*, **2015**, 10, 413–420.

37) M. A. Haq, Y. Su, and D. Wang,. "Mechanical properties of PNIPAM based hydrogels: A review",. *Materials Science and Engineering: C*, **2017**, 70, 842–855.

38) Tiantian Gan, Ying Guan, and Yongjun Zhang,. "Thermogelable PNIPAM microgel dispersion as 3D cell scaffold: effect of syneresis",. *Journal of Materials Chemistry*, **2010**, 20, 5937–5944.

39) Y. Guan, and Y. Zhang,. "PNIPAM microgels for biomedical applications: from dispersed particles to 3D assemblies",. *Soft Matter*, **2011**, 7, 6375–6384.

40) Y. G. Takei, T. Aoki, K. Sanui, N. Ogata, T. Okano, and Y. Sakurai,. "Temperature-responsive bioconjugates. 2. Molecular design for temperature-modulated bioseparations",. *Bioconjugate Chemistry*, **1993**, 4, 341–346.

41) H. Feil, Y. H. Bae, J. Feijen, and S. W. Kim,. "Effect of comonomer hydrophilicity and ionization on the lower critical solution temperature of *N*-isopropylacrylamide copolymers",. *Macromolecules*, **1993**, 26, 2496–2500.

42) M. Nakayama,. "Thermoresponsive polymeric materials for drug delivery systems",. *drug delivery system*, **2008**, 627–636.

43) T. Nishio, R. Suzuki, Y. Tsukada, H. Kanazawa, T. Okano, and T. Miyabe-Nishiwaki,. "Aqueous chromatographic system for the quantification of propofol in biological fluids using a temperature-responsive polymer modified stationary phase.",. *Journal of chromatography. A*, **2009**, 1216, 7427–7432.

44) K. Dušek, and D. Patterson,. "Transition in swollen polymer networks induced by intramolecular condensation",. *Journal of Polymer Science Part A-2: Polymer Physics*, **1968**, 6, 1209–1216.

45) S. Aoshima, H. Oda, and E. Kobayashi,. "Synthesis of thermally-induced phase separating polymer with well-defined polymer structure by living cationic polymerization. I. Synthesis of poly(vinyl ether)s with oxyethylene units in the pendant and its phase separation behavior in aqueous solution",. *Journal of Polymer Science Part A: Polymer Chemistry*, **1992**, 30, 2407–2413.

46) Y. Xia, N. A. D. Burke, and H. D. H. Stöver,. "End Group Effect on the Thermal Response of Narrow-Disperse Poly(*N* -isopropylacrylamide) Prepared by Atom Transfer Radical Polymerization",. *Macromolecules*, **2006**, 39, 2275–2283.

47) H. Kanazawa, K. Yamamoto, and Y. Matsushima,. "Temperature-Responsive Chromatography Using Poly(*N*-isopropylacrylamide)-Modified Silica",. *analytical chemistry*, **1996**, 68, 100–105.

48) T. Yakushiji, K. Sakai, A. Kikuchi, T. Aoyagi, Y. Sakurai, and T. Okano,. "Graft Architectural Effects on Thermoresponsive Wettability Changes of Poly(*N* -isopropylacrylamide)-Modified Surfaces",. *Langmuir*, **1998**, 14, 4657–4662.

49) H. Kanazawa,. "Temperature-responsive polymers for liquid-phase separations.",. *Analytical and bioanalytical chemistry*, **2004**, 378, 46–48.

50) C. Sakamoto, Y. Okada, H. Kanazawa, E. Ayano, T. Nishimura, M. Ando, A. Kikuchi, and T. Okano,. "columns grafted with copolymers of *N* -isopropylacrylamide",. **2004**, 1030, 247–253.

51) Y. Cai, W. Shen, S. L. Loo, W. B. Krantz, R. Wang, A. G. Fane, and X. Hu,. "Towards temperature driven forward osmosis desalination using Semi-IPN hydrogels as reversible draw agents.",. *Water research*, **2013**, 47, 3773–81.

52) D. Li, X. Zhang, J. Yao, P. Simon, H. Wang, G. P. Simon, H. Wang, P. Simon, and H. Wang,. "Stimuli-responsive polymer hydrogels as a new class of draw agent for forward osmosis desalination.",. *Chemical communications*, **2011**, 47, 1710–1712.

53) D. Li, X. Zhang, G. P. Simon, and H. Wang,. "Forward osmosis desalination using polymer hydrogels as a draw agent: influence of draw agent, feed solution and membrane on process performance.",. *Water research*, **2013**, 47, 209–215.

54) T. Okano, N. Yamada, H. Sakai, and Y. Sakurai,. "A novel recoverv svstern for cultured cells using plasrna-trGatid polystyrene dishes grafted with poly (N4sopropylacrylarnide)",. **1993**, 27, 1243–1251.

55) M. Hirose, O. H. Kwon, M. Yamato, A. Kikuchi, and T. Okano,. "Creation of Designed Shape Cell Sheets That Are Noninvasively Harvested and Moved onto Another Surface",. *Biomacromolecules*, **2000**, 1, 377–381.

56) M. Harimoto, and T. Okano,. *kagakutoseibutsu*, **2001**, 39, 671–676.

57) A. Kushida, M. Yamato, C. Konno, A. Kikuchi, Y. Sakurai, and T. Okano,. "Decrease in culture temperature releases monolayer endothelial cell sheets together with deposited fibronectin matrix from temperature-responsive culture surfaces.",. *Journal of biomedical materials research*, **1999**, 45, 355–362.

58) M. Yamato, M. Utsumi, A. Kushida, C. Konno, A. Kikuchi, and T. Okano,. "Thermo-responsive culture dishes allow the intact harvest of multilayered keratinocyte sheets without dispase by reducing temperature.",. *Tissue engineering*, **2001**, 7, 473–480.

59) A. Kushida, M. Yamato, A. Kikuchi, and T. Okano,. "Two-dimensional manipulation of differentiated Madin – Darby canine kidney (MDCK) cell sheets : The noninvasive harvest from temperature-responsive culture dishes and transfer to other surfaces",. *Journal of biomedical materials research*, **2001**, 54, 37–46.

60) A. Kushida, M. Yamato, Y. Isoi, A. Kikuchi, and T. Okano,. "A noninvasive transfer system for polarized renal tubule epithelial cell sheets using temperature-responsive culture dishes",. *european cells and materials*, **2005**, 10, 23–30.

61) T. Shimizu, M. Yamato, A. Kikuchi, and T. Okano,. "Cell sheet engineering for myocardial tissue reconstruction",. *Biomaterials*, **2003**, 24, 2309–2316.

62) Y. Hirokawa, and T. Tanaka,. "Volume phase transition in a nonionic gel",. *The Journal of Chemical Physics*, **1984**, 81, 6379-6380.

63) T. Tanaka, and D. J. Fillmore,. "Kinetics of swelling of gels",. *The Journal of Chemical Physics*, **1979**, 70, 1214.

64) D. J. Beebe, J. Moore, J. Bauer, Q. Yu, R. Liu, C. Devadoss, and B. Jo,. "Functional hydrogel structures for autonomous flow control inside microfluidic channels",. *Nature*, **2000**, 404, 588–90.

65) G. Gerlach, M. Guenther, J. Sorber, G. Suchaneck, K.-F. Arndt, and A. Richter,. "Chemical and pH sensors based on the swelling behavior of hydrogels",. *Sensors and Actuators B: Chemical*, **2005**, 111–112, 555–561.

66) S. K. De, N. R. Aluru, B. Johnson, W. C. Crone, D. J. Beebe, and J. Moore,. "Equilibrium swelling and kinetics of pH-responsive hydrogels: models, experiments, and simulations",. *Journal of Microelectromechanical Systems*, **2002**, 11, 544–555.

67) R. Yoshida, K. Sakai, T. Okano, and Y. Sakurai,. "Modulating the phase transition temperature and thermosensitivity in *N*-isopropylacrylamide copolymer gels",. *Journal of Biomaterials Science, Polymer Edition*, **1995**, 6, 585–598.

68) R. Yoshida, Y. Kaneko, K. Sakai, T. Okano, Y. Sakurai, Y. H. Bae, and S. W. Kim,. "Positive thermosensitive pulsatile drug release using negative thermosensitive hydrogels",. *Journal of Controlled Release*, **1994**, 32, 97–102.

69) J. S. Wilkes, J. A. Levisky, R. A. Wilson, and C. L. Hussey,. "Dialkylimidazolium chloroaluminate melts: a new class of room-temperature ionic liquids for electrochemistry, spectroscopy and synthesis."., *Inorganic Chemistry,* **1982**, 21(3), 1263–1264.

70) J. S. Wilkes, and M. J. Zaworotko,. "Air and water stable 1-ethyl-3-methylimidazolium based ionic liquids",. *Chemical Communications*, **1992**, 13, 965–967.

71) H. Ohno, and K. Ito,. "Room-Temperature Molten Salt Polymers as a Matrix for Fast Ion

Conduction",. *Chemistry Letters*, **1998**, 27, 751–752.

72) M. Yoshizawa, W. Ogihara, and H. Ohno,. "Novel Polymer Electrolytes Prepared by Copolymerization of Ionic Liquid Monomers",. **2002**, 594, 589–594.

73) A. Tsurumaki, M. A. Navarra, S. Panero, B. Scrosati, and H. Ohno,. "N-n-Butyl-N-methylpyrrolidinium hexafluorophosphate-added electrolyte solutions and membranes for lithium-secondary batteries",. *Journal of Power Sources*, **2013**, 233, 104–109.

74) G. B. Appetecchi, G. T. Kim, M. Montanino, M. Carewska, R. Marcilla, D. Mecerreyes, and I. De Meatza,. "Ternary polymer electrolytes containing pyrrolidinium-based polymeric ionic liquids for lithium batteries",. *Journal of Power Sources*, **2010**, 195, 3668–3675.

75) A.-L. Pont, R. Marcilla, I. De Meatza, H. Grande, and D. Mecerreyes,. "Pyrrolidinium-based polymeric ionic liquids as mechanically and electrochemically stable polymer electrolytes",. *Journal of Power Sources*, **2009**, 188, 558–563.

76) E. Stathatos, V. Jovanovski, B. Orel, I. Jerman, and P. Lianos,. "Dye-Sensitized Solar Cells Made by Using a Polysilsesquioxane Polymeric Ionic Fluid as Redox Electrolyte",. *Journal of Physical Chemistry C*, **2007**, 111, 6528–6532.

77) T. Mizumo, T. Watanabe, N. Matsumi, and H. Ohno,. "Preparation of ion conductive inorganic–organic composite systems by in situ sol–gel reaction of polymerizable ionic liquids",. *Polymers for Advanced Technologies*, **2008**, 19, 1445–1450.

78) R. Marcilla, F. Alcaide, H. Sardon, J. A. Pomposo, C. Pozo-Gonzalo, and D. Mecerreyes,. "Tailor-made polymer electrolytes based upon ionic liquids and their application in all-plastic electrochromic devices",. *Electrochemistry Communications*, **2006**, 8, 482–488.

79) K. Fukumoto, and H. Ohno,. "LCST-Type Phase Changes of a Mixture of Water and Ionic Liquids Derived from Amino Acids",. *Angewandte Chemie*, **2007**, 46, 1852–1855.

80) Y. Kohno, H. Arai, S. Saita, and H. Ohno,. "Material design of ionic liquids to show temperature-sensitive LCST-type phase transition after mixing with water",. *Australian Journal of Chemistry*, **2011**, 64, 1560–1567.

81) Y. Kohno, and H. Ohno,. "Temperature-responsive ionic liquid/water interfaces: relation between hydrophilicity of ions and dynamic phase change",. *Physical chemistry chemical physics* , **2012**, 14, 5063–5070.

82) Y. Kohno, and H. Ohno,. "Key Factors to Prepare Polyelectrolytes Showing Temperature-Sensitive Lower Critical Solution Temperature-type Phase Transitions in Water",. *Australian Journal of Chemistry*, **2012**, 65, 91–94.

83) R. Wang, W. Leng, Y. Gao, and L. Yu,. "Microemulsion-like aggregation behaviour of an LCST-type ionic liquid in water",. *RSC Advances*, **2014**, 4, 14055–14062.

84) H. Kang, D. E. Suich, J. F. Davies, A. D. Wilson, J. J. Urban, and R. Kostecki,. "Molecular insight into the lower critical solution temperature transition of aqueous alkyl phosphonium benzene

sulfonates",. *Communications Chemistry*, **2019**, 2, 1-11.

85) W. W. Li, and P. Wu,. "Unusual thermal phase transition behavior of an ionic liquid and poly(ionic liquid) in water with significantly different LCST and dynamic mechanism",. *Polymer Chemistry*, **2014**, 5, 5578-5590.

86) G. Wang, and P. Wu,. "In-depth study of the phase separation behaviour of a thermoresponsive ionic liquid and a poly(ionic liquid) in concentrated aqueous solution",. *Soft Matter*, **2015**, 11, 5253–5264.

87) J. J. Kim, H. Kang, Y. S. Choi, Y. A. Yu, and J. C. Lee,. "Thermo-responsive oligomeric poly(tetrabutylphosphonium styrenesulfonate)s as draw solutes for forward osmosis (FO) applications",. *Desalination*, **2016**, 381, 84–94.

88) X. Fan, H. Liu, Y. Gao, Z. Zou, V. S. J. Craig, G. Zhang, and G. Liu,. "Forward-Osmosis Desalination with Poly(Ionic Liquid) Hydrogels as Smart Draw Agents",. *Advanced Materials*, **2016**, 28, 4156–4161.

89) C.-H. Hsu, C. Ma, N. Bui, Z. Song, A. D. Wilson, R. Kostecki, K. M. Diederichsen, B. D. McCloskey, and J. J. Urban,. "Enhanced Forward Osmosis Desalination with a Hybrid Ionic Liquid/Hydrogel Thermoresponsive Draw Agent System",. *ACS Omega*, **2019**, 4, 4296–4303.

90) S. Y. Fan, Y. N. Hao, W. X. Zhang, A. Kapasi, Y. Shu, J. H. Wang, and W. Chen,. "Poly(ionic liquid)-Gated CuCo2S4 for pH-/Thermo-Triggered Drug Release and Photoacoustic Imaging",. *ACS Applied Materials and Interfaces*, **2020**, 12, 9000–9007.

91) Y. Men, X.-H. Li, M. Antonietti, and J. Yuan,. "Poly(tetrabutylphosphonium 4-styrenesulfonate): a poly(ionic liquid) stabilizer for graphene being multi-responsive",. *Polymer Chemistry*, **2012**, 3, 871-873.

92) R. Marcilla, M. L. L. Curri, P. D. D. Cozzoli, M. T. T. Martínez, I. Loinaz, H. Grande, J. A. A. Pomposo, and D. Mecerreyes,. "Nano-objects on a round trip from water to organics in a polymeric ionic liquid vehicle.",. *Small*, **2006**, 2, 507–512.

93) R. Marcilla, E. Ochoteco, C. Pozo-Gonzalo, H. Grande, J. A. Pomposo, and D. Mecerreyes,. "New Organic Dispersions of Conducting Polymers Using Polymeric Ionic Liquids as Stabilizers",. *Macromolecular Rapid Communications*, **2005**, 26, 1122–1126.

94) A. Tudor, J. Saez, L. Florea, F. Benito-Lopez, and D. Diamond,. "Poly(ionic liquid) thermo-responsive hydrogel microfluidic actuators",. *Sensors and Actuators B: Chemical*, **2017**, 247, 749–755.

95) A. Tudor, C. Delaney, H. Zhang, A. J. Thompson, V. F. Curto, G.-Z. Yang, M. J. Higgins, D. Diamond, and L. Florea,. "Fabrication of soft, stimulus-responsive structures with sub-micron resolution via two-photon polymerization of poly(ionic liquid)s",. *Materials Today*, **2018**, 21, 807–816.

96) Y. Kohno, D. L. Gin, R. D. Noble, and H. Ohno,. "A thermoresponsive poly(ionic liquid) membrane

enables concentration of proteins from aqueous media",. *Chemical Communications*, **2016**, 52, 7497–7500.

Chapter 2

Design and properties of poly(ionic liquid)s to show LCST-type phase transition with water

2.1 Introduction

Temperature-responsive polymers, which show an LCST-type phase transition with water, have been extensively studied as "intelligent polymers" or “smart polymers” in recent years and have been applied to a wide range of applications. Nevertheless, all of these intelligent polymers reported so far have been developed with mainly nonionic polymers represented by poly(*N*-isopropylacrylamide). Conventional temperature-responsive polymers have difficulty in both controlling the phase transition temperature and maintaining the temperature responsiveness. Therefore, their application has been limited to use at the phase transition temperature.

Meanwhile, PILs have been studied intensively as functional fluids for various applications. Interest has been focused recently on polyelectrolytes prepared by the polymerization of ILs. In 1998, Ito *et al,* first reported the polymerization of ILs composed of 1-alkyl-3-vinylimidazolium cations, giving rise to "polymerized ILs (PILs)."[1] PILs are readily prepared from IL monomers composed of ions having polymerizable groups such as vinyl groups. They have unique combinations of properties derived from the starting ILs, including exceptionally high affinity with carbon dioxide, low glass transition temperature, and controllable affinity with water. Many kinds of PILs have been designed and have found application as functional polymers, including as solid polymer electrolytes for electrochemical devices[2], as adsorbents or in selective permeation of gases[3], and as self-assembling materials.[4] Given the structural diversity of the component ions of IL monomers, the potential of these PILs is still not fully explored.

Ohno *et al.* has designed various ILs, including IL monomer, exhibiting unique phase behavior upon mixing with water. In 2012, they prepared a thermoresponsive IL monomer composed of tetrabutylphosphonium p-styrenesulfonate ([P_{4444}][SS]), and polymerized it to prepare poly([P_{4444}][SS]), which underwent a lower critical solution temperature (LCST)-type phase transition upon mixing with water.[5] This polyelectrolyte–water mixture was miscible at room temperature, but a turbid suspension was obtained upon heating it. It reversibly returned to a homogeneous mixture upon cooling. Although an LCST-type phase transition had already been observed in aqueous solutions containing some non-ionic polymers represented by poly(*N*-isopropylacrylamide)[6], the LCST-type phase transition of fully charged polymer–water mixtures was novel. The investigation of thermoresponsive IL monomer and PIL has started, and the knowledge is still insufficient.

In this chapter, the author demonstrated the LCST-type dynamic change in the PIL/water mixture. As a preliminary step, several IL monomers were prepared and their thermoresponsive behavior upon mixing with water was studied. Then, the prepared IL monomers were polymerized with radical polymerization, and thermo-responsivity of the resulting PIL/water mixture was evaluated. In addition, the aggregation state of PIL in water was evaluated, and the phase behavior of the PIL/water mixture was compared with that of the conventional non-ionic thermo-responsive polymer to deepen the understanding of this unique phenomenon.

It is noted that the effect of molecular weight on the phase transition temperature of PILs has been investigated using PILs exhibiting UCST, and it was found that the cloud point is independent of molecular weight even in the low molecular weight polymer, which is generally affected by molecular structure and initiator.[7] Therefore, the molecular weight of PILs was not specifically controlled in this study since the effect of molecular weight on the phase behavior and the phase transition temperature was regarded as negligible.

2.2 Experimental

2.2.1 Synthesis of IL monomer

Materials

Tetrabutylphosphonium hydroxide ([P_{4444}]OH)、tributylhexylphosphonium bromide ([P_{4446}]Br), Tributylvinylbenzylphosphonium chloride ([P_{444}VB]Cl) were provided by Hokko Chemical Industry Co., Ltd.. The following chemicals were purchased from Tokyo Chemical Industry: tributyloctylphosphonium bromide ([P_{4448}]Br), sodium styrenesulfonate (Na[SS]), potassium 3-sulfopropyl methacrylate (K[MC3S]), sodium 1-butanesulfonate (Na[C4S]), sodium 1-pentanesulfonate (Na[C5S]), sodium 1-hexanesulfonate (Na[C6S]), and 4-(chloromethyl) styrene. Potassium 3-sulfopropyl acrylate (K[AC3S]) was purchased from Sigma-Aldrich Co. LLC. Acetonitrile, hexane, and hydrochloric acid were purchased from Kanto Chemical Co., Inc. Tributylamine and 2,20-Azobis(2-methylpropionamidine) dihydrochloride (V-50) were purchased from Wako. Chem. Co. 2,2'-Azobis(isobutyronitrile) (AIBN) was purchased from Kanto Chemical Co. and was used with recrystallization with methanol. All materials without AIBN were used without further purification.

Synthesis of Tetrabutylphosphonium chloride ([P_{4444}]Cl)

A slight excess of HCl was added to the [P_{4444}]OH aqueous solution to neutralize it. After confirming that the solution turned acidic, water and small excess of HCl were removed by evaporation. After that, it was re-dissolved in chloroform, and an appropriate amount of potassium hydrogen carbonate was added to that and stirred overnight. Potassium hydrogen carbonate was then removed by filtration, and chloroform was removed by evaporation. After confirming that the pH of the aqueous solution of this salt was neutral, the [P_{4444}]Cl was dried by heating and vacuum drying.

Synthesis of [N_{444}VB]Cl

Tributylamine was placed in a vial, dissolved in acetonitrile, and added dropwise a 0.9-fold mole of 4 chloromethyl styrene. The vial was heated in an oil bath at 50 °C for 2 hours to allow the quaternization reaction to proceed. After the acetonitrile was removed from the solution by an evaporator, it was added dropwise to an excess of hexane and purified. After that, the white solid was taken out by suction filtration and was dried to obtain the target [N_{444}VB]Cl.

Synthesis of IL monomer via an anion exchange reaction

ILs containing following anions; [SS]$^-$, [MC3S]$^-$, [AC3S]$^-$, [C4S]$^-$, [C5S]$^-$, or [C6S]$^-$ were prepared by anion exchange reaction of [P_{4444}]Cl, [P_{4446}]Br, [P_{4448}]Br, [P_{444}VB]Cl, and [N_{444}VB]Cl with the corresponding alkali salts, according to the literature.[8,9] The products were extracted with dichloromethane, and the dichloromethane layer was washed with water several times. Since the polymerization reaction of the IL containing [P$_{444}$VB]$^+$ is likely to proceed by light or heat, stirring and evaporation were carried out under dark, and stirring was carried out in an ice bath. After evaporation, the products were dried in a vacuum at 70 °C for at least 24h. The water content of all the ILs prepared was confirmed to be less than 0.2 wt% with Karl Fischer titration method (Kyoto Electronics MKC-510N). Structure of the ILs was confirmed with both ^{1}H NMR spectrometry (JEOLECX-400, 400 MHz) and elemental analysis (Elementar vario EL III).

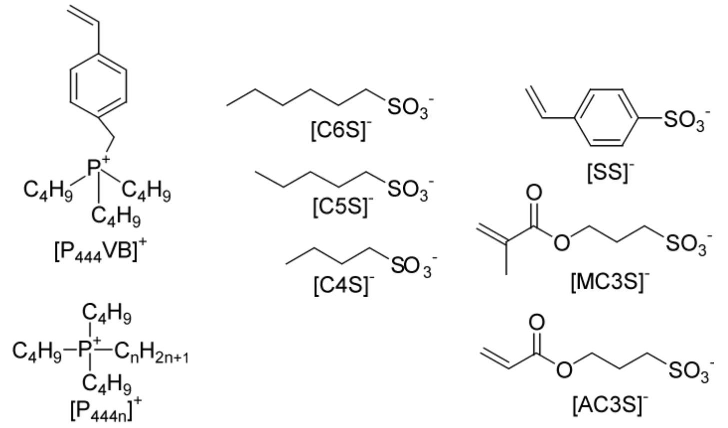


Figure 2-1 Structure of cations and anions used in this study.

Characterization of IL

[P$_{4444}$][SS]

^{1}H NMR (400 MHz, CDCl$_3$, δ /ppm relative to TMS):0.93(t, 12H, CH$_3$, J = 4.59 Hz), 1.43-1.49(m, 16H, CH$_2$), 2.26(t, 8H, CH2, J = 3.12 Hz), 5.22(dd, 1H, CH), 5.76(dd, 1H, CH), 6.64-6.78(m, 1H, CH), 7.33 (d, 2H, CH), 7.85(d, 2H, CH).

[P$_{4444}$][MC3S]

^{1}H NMR (400 MHz, CDCl$_3$, δ /ppm relative to TMS): 0.88(t, 12H, CH$_3$, J = 6.80 Hz), 1.44(t, 16H, CH$_2$, J = 3.60 Hz), 1.82 (s,3H, CH$_3$), 2.20-2.27 (m, 8H, CH$_2$), 2.80(t, 2H, CH2, J = 7.80 Hz), 4.17 (t,2H, CH$_2$, J = 6.40 Hz), 5.43(dd, 1H, CH), 5.99(dd, 1H, CH).

[P$_{4446}$][SS]

^{1}H NMR (400 MHz, CDCl$_3$, δ /ppm relative to TMS): 0.90(t, 12H, CH$_3$, J = 6.87 Hz), 1.26(t, 4H, CH$_2$, J = 3.66 Hz), 1,43-1.48(m, 16H, CH$_2$), 2.25(t, 8H, CH$_2$, J = 2.98 Hz), 5.23(dd, 1H, CH) , 5.74(dd, 1H, CH), 7.35(d, 2H, CH) , 7.84(d, 2H, CH).

[P$_{4446}$][MC3S]

^{1}H NMR (400 MHz, CDCl$_3$, δ /ppm relative to TMS): 0.90(t, 3H, CH$_3$, J = 2.29 Hz), 0.97(t,9H, CH3, J = 6.86 Hz), 1.31-1.32(m, 4H, CH$_2$), 1.51-1.56(m, 16H, CH$_2$), 1.92(t, 3H, CH$_3$), 2.19-2.38(m, 11H, CH$_2$), 2.89-2.93(m, 2H, CH$_2$), 4.26(t, 2H, CH$_2$, J = 6.52 Hz), 5.52(dd, 1H, CH), 6.08(dd, 1H, CH).

*[*P*4446*][AC3S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.95(t, 12H, CH_3, *J* = 3.66 Hz), 1.31-1.33(m, 4H, CH_2), 1.52-1.54(m, 16H, CH_2), 2.19-2.35(m, 14H, CH_2), 2.89-2.92(m, 2H, CH_2), 4.28(t, 2H, CH_2, *J* = 6.64 Hz), 5.80(dd, H, CH), 6.36(dd, H, CH).

*[*P*4448*][MC3S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.80(t, 3H, CH_3, *J* = 8 Hz),0.98(t, 9H, CH_3, *J* = 8 Hz), 1.27(m, 8H, CH_2), 1.53(t, 16H, CH_2, *J* = 4 Hz), 1.92(s, 3H, CH_3), 2.19-2.38(m, 11H, CH_2), 2.91(t, 2H, CH2, *J* = 8 Hz), 4.26(t,2H, CH_2, *J* = 6 Hz), 5.52(dd, 1H, CH), 6.08(dd, 1H, CH).

*[*P*444VB*][MC3S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.92(t, 3H, CH_3, *J* = 6.80 Hz), 1.409-1.515(m, 12H, CH_2), 1.90(s, 3H, CH_3), 2.114-2.328 (m, 8H, CH_2), 2.95(t, 2H, CH_2, *J* = 7.60 Hz), 3.969(d, 2H, CH_2), 4.27(t, 2H, CH_2, *J* = 6.40 Hz), 5.29(dd, 1H, CH_2), 5.50(dd, 1H, CH), 5.75(dd, 1H, CH), 6.09(dd, 1H, CH), 6.63-6.71(m, 1H, CH), 7.32(d, 2H, CH), 7.40(d, 2H, CH).

*[*P*444VB*][AC3S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.89(t, 12H, CH_3, *J* = 7.10 Hz), 1.411-1.514(m, 12H, CH_2), 2.12-2.28 (m, 2H, CH_2), 2.93(t, 2H, CH_2, *J* = 7.60 Hz), 3.97(d, 2H, CH_2), 4.25(t, 2H, CH_2, *J* = 6.41 Hz), 5.40(dd, 1H, CH), 5.76(dd, 1H, CH), 6.06(dd, 1H, CH), 6.40(dd, 1H, CH_2), 6.62-6.72(m, 1H, CH), 7.31(d, 2H, CH), 7.40(d, 2H, CH).

*[*P*444VB*][C4S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.85(t, 3H, CH_3, *J* = 7.22 Hz), 0.92(t, 9H, CH_3, *J* = 7.10 Hz), 1.30-1.44 (m, 14H, CH_2), 1.82-1.90(m, 2H, CH_2), 2.40(t, 6H, CH_2, *J* = 13.8 Hz), 2.85(t, 2H, CH_2, *J* = 8.20 Hz), 4.00(d, 2H, CH_2), 5.20(dd, 1H, CH), 5.71(dd, 1H, CH), 6.70-6.75(m, 1H, CH), 7.35(d, 2H, CH), 7.44(d, 2H, CH).

*[*P*444VB*][C5S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.88(t, 3H, CH_3, *J* = 7.15 Hz), 0.93(t, 9H, CH_3, *J* = 7.00 Hz), 1.26-1.32(m, 4H, CH_2), 1.42-1.61 (m, 12H, CH_2), 1.82-1.95(m, 2H, CH_2), 2.36(t, 6H, CH_2, *J* = 14.6 Hz), 2.95(t, 2H, CH_2, *J* = 8.55 Hz), 4.10(d, 2H, CH_2), 5.33(dd, 1H, CH), 5.81(dd, 1H, CH), 6.65-6.83(m, 1H, CH), 7.28(d, 2H, CH), 7.36(d, 2H, CH).

*[*P*444VB*][C6S]*

^{1}H NMR (400 MHz, $CDCl_3$, δ /ppm relative to TMS): 0.85(t, 3H, CH_3, *J* = 7.20 Hz), 0.95(t, 9H, CH_3, *J* = 7.00 Hz), 1.26-1.30(m, 4H, CH_2), 1.36-1.51 (m, 12H, CH_2), 1.60(s, 6H, CH_2), 1.82-1.90(m, 2H, CH_2), 2.31(t, 6H, CH_2, *J* = 14.6 Hz), 2.85(t, 2H, CH_2, *J* = 8.40 Hz), 4.00(d, 2H, CH_2), 5.29(dd, 1H, CH), 5.78(dd, 1H, CH), 6.65-6.72(m, 1H, CH), 7.33(d, 2H, CH), 7.40(d, 2H, CH).

2.2.2 polymerization of ILs

Synthesis of PILs

There are two main procedures to obtain PILs; one is to polymerize IL monomers directly. This method is effective but requires several preparation and purification steps to obtain the IL monomers. Another way involves the polymerization of monomeric IL precursors followed by quaternization or an anion exchange reaction. In this study, PIL having $[P_{4444}]^+$,$[P_{4446}]^+$, $[P_{4448}]^+$, and $[P_{444}VB]^+$ were prepared by the former method i.e., radical polymerization. PIL comprised of $[P_{444}VB]$ or $[N_{444}VB]$ cation and [C4S], [C5S], or [C6S] anions were prepared by the latter method.

Synthesis of PILs comprised of $[P_{4444}]$,$[P_{4446}]$, $[P_{4448}]$ or $[P_{444}VB]$ cations.

A series of anionic PILs comprised of *n*-tributyl-*n*-alkylphosphonium cations ($[P_{444n}]^+$, n = 4, 6, 8) have been prepared *via* a free radical polymerization of corresponding IL monomer in water. Each IL monomer (20 mL) was dissolved in 80mL water and loaded into a 200mL three-necked round-bottom flask equipped with a magnetic stirrer bar. The flask was sealed, and the inner solution was degassed exhaustively by sonication. A water-soluble radical initiator (2,2'-azobis(isobutyronitrile), 0.076 g, 0.28 mmol (1.0 mol% to monomer)) was then added to the solution. The polymerization was conducted at 80 °C for 3 h. The resulting polymer solution was dialyzed in pure water thoroughly to remove monomers. The absence of the chloride anion was confirmed by the silver nitrate test. The solution was freeze-dried to obtain a white powder.

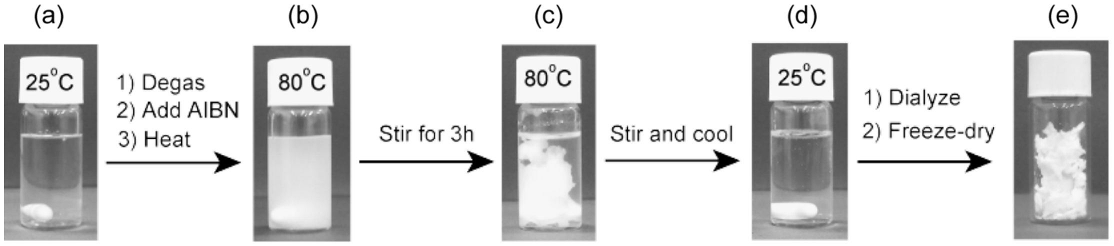


Figure 2-2 Scheme of preparation of PIL *via* radical polymerization.

Synthesis of PILs comprised of $[P_{444}VB]$ or $[N_{444}VB]$ cations and [C4S][C5S], or [C6S] anions.

Each PIL was prepared by the same procedure as above. The synthesis of $[P_{444}VB]$[C4S] is described here as an example. Tributyl (4-vinylbenzyl) phosphonium chloride (10.0 g, 28 mmol) was dissolved in 80mL water and loaded into a 200mL three-necked round-bottom flask equipped with a magnetic stirrer bar. The flask was sealed, and the inner solution was degassed exhaustively by sonication. A water-soluble radical initiator (2, 20-Azobis(2-methylpropionamidine) dihydrochloride, 0.076 g, 0.28 mmol (1.0 mol% to monomer)) was then added to the solution. The polymerization was conducted at 60 °C for 3 h. The resulting polymer solution was dialyzed in pure water thoroughly to remove monomer, and freeze-dried to obtain a white powder. Then, obtained poly($[P_{444}VB]$Cl) (0.47 g, 1.3

mmol) was mixed with 50mL water and 0.30 g (1.7 mmol) of sodium 1-butane sulfonate was added and stirred for 24 h. The resulting solution was dialyzed. The absence of the Cl anion was confirmed by silver the nitrate test. The solution was freeze-dried to obtain a white powder.

2.2.3 Evaluation of phase transition behavior

Phase transition behavior with water.

Temperature-dependent turbidity measurement (heating rate; 2 °C/min) was used to determine the phase transition temperature (T_c) of the IL or PIL/water mixtures by defining the T_c value as the temperature value at which the turbidity value became 90 % upon heating. The measurement was carried out using an ultraviolet-visible near-infrared spectrophotometer V-660 of Nippon Spectroscopy Co., Ltd. The plastic cell with an optical path length of 10 mm was used. The concentration of ILs or PILs was 10 wt% in water.

Light microscopic observation

Poly([P_{4446}][MC3S]) was added to 200 mM phosphate buffer and stirred at room temperature for overnight to give a clear homogeneous solution. A dye, coomassie brilliant blue R-250 (**Figure 2-3**), selectively soluble in the IL, was added to this solution and stirred to give a uniform solution. The resulting solution was centrifuged (10,000 rpm, EMD Millipore Corporation) to remove an excess dye. Only the upper layer of the solution after centrifugation was collected and heated at 2 ^{o}C/min from room temperature on a microscope cooling and heating device (LK -600, JAPAN HIGH TECH Co., Ltd.), and the change of the solution state as the function of temperature was observed by s optical microscope (BX -51, OLYMPUS).

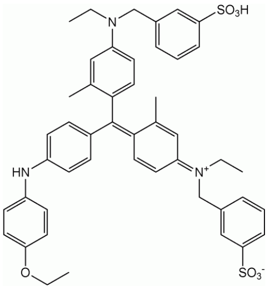


Figure 2-3 Structure of coomassie brilliant blue.

Transmission electron microscopy (TEM) observation

Surface-coated copper TEM grids were treated with JEOL HDT-400 to obtain hydrophilic surface. The aqueous solutions containing 0.1 w/w % polymer dispersions were then dropped onto the TEM grids. The grids were dried immediately by incubating them at 60 ^{o}C to avoid aggregation of polymers. TEM was measured with JEOL JEM-1400 operating at 120 kV.

Dynamic light scattering (DLS) measurement

DLS measurements were carried out using Fiber-Optical Dynamic Light-Scattering Spectrometer (FDLS-3000, Otsuka Electronics Co., Ltd.) equipped with 100 mW Diode-pumped Solid-state laser ($\lambda = 532$ nm) at the scattering angle of 90^{o}.

2.3 Phase transition of the IL monomers in water

First, I have prepared some IL monomers and studied their thermoresponsive behavior upon mixing with water. The thermoresponsive behavior of IL monomers was checked after mixing with 90 wt% of water, and the resulting solution was cooled to 0 °C and then heated gradually to 70 °C. Each IL monomer was classified into "Miscible", "Phase separation", and "LCST" according to the phase of the solution. When a clear homogeneous phase of the IL/water mixture was separated into two phase upon heating, this was classified as an LCST-type phase transition. It was found that some IL monomers underwent a temperature-sensitive LCST-type phase transition upon mixing with water. When a clear homogeneous solution of the $[P_{4446}]$[MC3S]/water mixture was heated, it became turbid, and then showed an apparent liquid-liquid phase separation between the IL and aqueous phase. The phase transition temperature (T_c) was determined with turbidity measurement for those mixtures showing the phase transition.

Table 2-1 shows the phase state and transition temperature of the ILs used in this study. It has already been reported that the phase behavior of ILs with water and the T_c value largely depends on the total hydrophilicity of the component ions.[10] To compare the phase behavior of ILs having $[P_{4444}]^+$ and $[P_{4446}]^+$, it was found that the [SS] anion is more hydrophobic than the [MC3S] anion. Both $[P_{4448}]$[MC3S] and $[P_{444}VB]$[MC3S] showed LCST-type phase transition, but their phase transition temperature suggested that $[P_{444}VB]^+$ was more hydrophobic than $[P_{4448}]^+$. The order of hydrophilicity of anions and cations used in this study are as follows. Since ILs were not prepared with the same cation, the exact order of hydrophilicity between $[SS]^-$ and $[C6S]^-$ was not clarified.

Table 2-1 Phase state and phase transition temperature of each IL monomer.

IL monomer	Phase state	T_c / °C (10 wt%)
$[P_{4444}]$[SS]	LCST	34
$[P_{4444}]$[MC3S]	Miscible	—
$[P_{4446}]$[SS]	Phase separation	—
$[P_{4446}]$[MC3S]	LCST	44
$[P_{4446}]$[AC3S]	Miscible	—
$[P_{4448}]$[MC3S]	LCST	20
$[P_{444}VB]$[C4S]	Miscible	—
$[P_{444}VB]$[C5S]	LCST	41
$[P_{444}VB]$[C6S]	Phase separation	—
$[P_{444}VB]$[MC3S]	LCST	7
$[P_{444}VB]$[AC3S]	LCST	19

Therefore, the order of hydrophilicity of the ions used in this study, as estimated from the phase transition temperature, is shown below.

Hydrophobic $[P_{444}VB]^+ < [P_{4448}]^+ < [P_{4446}]^+ < [P_{4444}]^+$ Hydrophilic
Hydrophobic $[SS][C6S]^- < [MC3S]^- < [AC3S]^- < [C5S]^- < [C4S]^-$ Hydrophilic

2.4 Phase transition of the PILs in water

Effect of polymerization and ionic structure on the phase transition behavior of PILs

The IL monomers showing LCST-type phase transition with water were then polymerized with radical polymerization. The resulting PIL was mixed with 90 wt% water, and the solution was cooled to 0 °C. and then heated to 70 °C. Each PIL was classified as "insoluble", "miscible", or "LCST" depending on the phase behavior of the solution. The PIL/water mixture exhibiting LCST was homogeneous at low temperatures, becoming turbid above the phase transition temperature. In the IL/water mixture system, phase separation immediately occurred above T_c. while in the PIL/water mixture system, the solution became turbid immediately above T_c, however the phase separation or precipitation were not observed even after a few minutes. It is suggested that the PIL showed incompletely phase separate above T_c are not hydrophobic, since a hydrophobic polymer would not completely dissolve in water. Otherwise, it is considered that the phase separation behavior becomes sluggish with an increase in the molecular weight of PIL caused by polymerization.

Table 2-2 shows phase behavior and T_c of each IL monomer and resulting PIL. The author expected that the PIL obtained by polymerizing an IL monomer showing LCST-type phase transition would also have termoresponsivity. However, some of PILs were a hydrophobic polymer. In addition, T_c of poly ($[P_{4444}][SS]$) and poly($[P_{4446}][MC3S]$) showing thermo-responsivity was different from that

Table 2-2 Phase transition temperature of each IL monomer and PIL.

IL	Monomer / T_c (10 wt%)	Polymer / T_c (10 wt%)
$[P_{4444}][SS]$	35	58
$[P_{4446}][MC3S]$	44	40
$[P_{444}VB][C5S]$	41	42 (*3 wt%*)
$[P_{4448}][MC3S]$	20	Insoluble
$[P_{444}VB][MC3S]$	7	Insoluble
$[P_{444}VB][AC3S]$	19	Insoluble
$[P_{4446}][SS]$	Phase separation	Insoluble
$[P_{444}VB][C4S]$	Miscible	Miscible (*3 wt%*)
$[P_{444}VB][C6S]$	Phase separation	Insoluble (*3 wt%*)

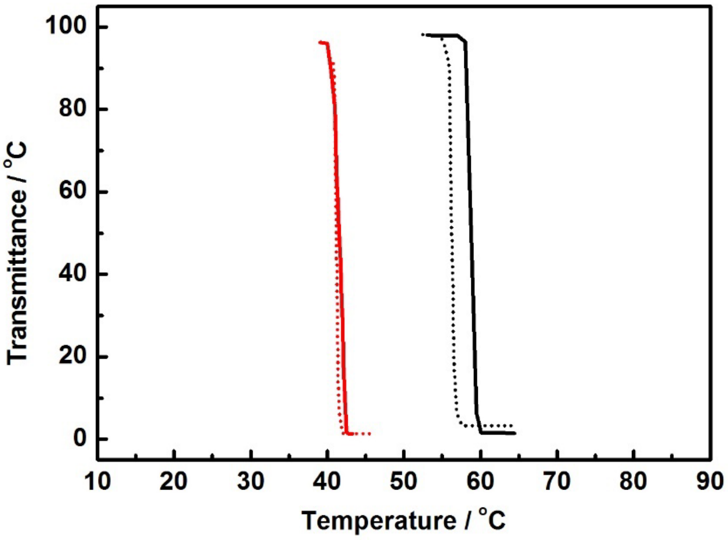


Figure 2-4 The change in transmittance of poly([P_{4444}][SS])/water mixture (red) and poly-([P_{4446}][MC3S])/water mixture (black). The straight line denotes the transmittance during heating and the dotted line denotes the transmittance during cooling.

of a monomer/water mixture having the same concentration. Former showed a higher phase transition temperature after polymerization. The change in transmittance of both poly([P_{4444}][SS]) and poly([P_{4446}][MC3S]) in water was highly sensitive upon temperature change and had small hysteresis (**Figure 2-4**).

Previous reports showed that the total hydrophilicity of component ions was an essential factor in controlling the LCST behavior of ILs.[10] A similar trend was found in the resulting PIL. For example, comparing poly([P_{4446}][MC3S]) and poly([P_{4448}][MC3S]) having the same main chain, poly([P_{4448}][MC3S]) is more hydrophobic be due to the total hydrophilicity of the component ions. However, a comparison of poly([P_{4444}][SS]) and poly([P_{4446}][MC3S]) indicated that poly([P_{4444}][SS]) was more hydrophobic polymer because of its higher phase transition temperature. The phase transition temperature revealed that [P_{4444}][SS] was more hydrophilic than [P_{4446}][MC3S]. This result indicated that the effect of the charges on main chain was strengthened after polymerization. Here, focusing on the ions constituting the main chain of the two polymers, [SS] anion is more hydrophobic than [MC3S] anion, as indicated by the hydrophilic order of the ions as mentioned above.

When both anionic and cationic monomers were copolymerized like [P_{444}VB][MC3S] or [P_{444}VB][AC3S], many ionic interactions are formed between the polymer chains after polymerization. As a result, the polymer chains strongly aggregated to form polyion complexes, which are not dissolved in water. Thus, the direct polymerization of ILs having a polymerizing group in both anions and cations is not suitable for the preparation of IL-derived polymers.

The poly([P_{444}VB][C5S]) obtained by polymerizing [P_{444}VB][C5S] also maintained the temperature response but showed a very high viscosity when dissolved in water. This result may be due to a strong interaction between polymers. Therefore, the phase transition temperature could not be measured at 10 wt%. At a lower concentration, the mixture of 3 wt% polymers showed T_c at 42 °C. From these results, IL monomers that do not exhibit LCST might be PIL showing LCST only when it got a suitable hydrophobicity after polymerization.

Next, the other method was mentioned for obtaining PIL by polymerizing a precursor of an IL followed by the counterion exchange. In detail, [P_{444}VB]Cl or [N_{444}VB]Cl, precursors of IL, were polymerized and then were ion-exchanged with a sodium salt having a target anion. **Table 2-3** shows the phase behavior of each PIL/water mixture prepared by this methods. PILs containing [P_{444}VB] cation exhibited the same behavior in water as the PIL containing [P_{444}VB] cation prepared by radical polymerization. In PILs containing [N_{444}VB] cation, the difference of the phase behavior and T_c value depended on the alkyl length of anion species. Poly([N_{444}VB][C4S]), which has the shortest alkyl chain length of anions in this study, was a hydrophilic polymer that was always compatible with water over a wide temperature range. On the other hand, poly([N_{444}VB][C5S]) and poly([N_{444}VB][C6S]) showed the LCST type phase transitions, and their phase transition temperature decreased with the elongation of alkyl chain length of the anions. By comparing PILs in terms of the same number of alkyl chains, poly([N_{444}VB][C5S]) and poly([P_{444}VB][C5S]) underwent LCST behavior, but the Tc value of poly([N_{444}VB][C5S]) was higher than that for poly([P_{444}VB][C5S]). This results are consistent with previous reports on ILs in that ammonium-type ILs are more hydrophilic than phosphonium-type ones[11], suggesting that the PILs prepared in this study inherited the basic physico-chemical properties of the corresponding pristine ILs. It should be noted that poly([N_{444}VB][C6S])

Table 2-3 Phase transition temperature of each PIL prepared by polymerizing a precursor of an IL and then performing counterion exchange.

PIL	Polymer / T_c
Poly([N_{444}VB][C4S])	Miscible (*3 wt%*)
Poly([N_{444}VB][C5S])	64 (*3 wt%*)
Poly([N_{444}VB][C6S])	23 (*3 wt%*)
Poly([P_{444}VB][C4S])	Miscible (*3 wt%*)
Poly([P_{444}VB][C5S])	42 (*3 wt%*)
Poly([P_{444}VB][C6S])	Insoluble (*3 wt%*)

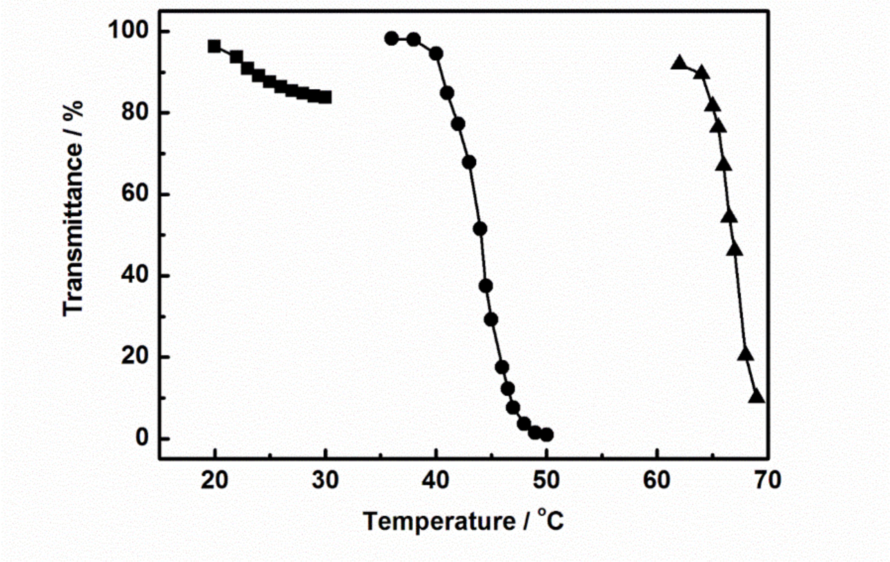


Figure 2-5. Effect of PIL species on transmittance change upon heating; (■): poly([N_{444}VB][C6S]), (●): poly([P_{444}VB][C5S]), (▲): poly([N_{444}VB][C5S])

exhibited rather anomalous behavior compared with other PILs showing the LCST-phase transition with water. The transmittance of the poly([N_{444}VB][C6S]) mixture fell below 90% at 23°C but did not drop to less than 80% even further increasing temperature (Figure 2-5). The aggregation state and aggregation size of poly([N_{444}VB][C6S]) in water should be different from those of poly([N_{444}VB][C5S]). Detailed analysis of this will be discussed later. As compared with the poly([P_{444}VB][C5S])/water mixture, the T_c of poly([N_{444}VB][C5S]) was 22 °C higher than that of poly([P_{444}VB][C5S]).

Sol-gel transition behavior of Poly([P_{4446}][MC3S])/water mixture

For IL-water mixtures exhibiting LCST behavior, the separated IL phase contain relatively more significant amounts of water than in highly hydrophobic ILs undergoing static phase separation with water[10]. The same phenomena were observed in the poly([P_{4446}][MC3S])-aqueous systems. To confirm this, four different mixtures, which contain 20 wt% of poly([P_{4446}][MC3S]) and phosphate buffer having different salt concentration, were heated above 45 °C and maintained without perturbation. **Figure 2-6** shows the phase change of the poly([P_{4446}][MC3S])/buffer mixture when buffer concentration was 200 mM. The homogeneously mixed solution turned turbid immediately upon heating at 45 °C, and poly([P_{4446}][MC3S]) was precipitated from the aqueous phase in 15 minutes. The precipitated polymer was dissolved again after cooling into 25 °C under stirring. Similar polymer phases were observed in all other mixtures with different buffer concentration. In the case of water, precipitation occurred after keeping it at 45 °C for more than 2 h due to the relatively high T_c value.

This suggests that the salt in phosphate buffer promotes the phase separation of PIL and water. It was also found that the salt concentration in the buffer had little effect on the phase separation rate.

After removing the upper aqueous phase, the thermal behavior of the resulting polymer-rich phase was studied. The precipitated polymer-rich phases showed a highly temperature-sensitive LCST transition (**Figure 2-6 right**). The turbid gel-like polymer phase immediately became a transparent viscous liquid upon cooling to 25 °C. This liquid began to turn turbid at 26 °C, and the gel-like phase was observed again upon maintaining it at 35 °C for 30 seconds. This implies that the polymer phase underwent a reversible liquid-to-gel transition over a small temperature change. Although some papers have reported this type of phase transition for a series of polymer-solvent mixed systems (e.g., amphiphilic block copolymers,[12,13] surfactants,[14], and even ILs[15]), there has been no previous report of the liquid-to-gel transition of polymers prepared in this manner that undergo LCST-type phase separation with water.

Visual observation of the polymer-rich phase of the lower phase confirmed that it was clearly larger than the volume of PIL existed in the mixture. To analyze this in detail, the water content of the separated polymer-rich phase (W_{aq}/wt%) was determined. The W_{aq} value was calculated using the weight loss of the samples measured by the TG/DTA method. As shown in **Table 2-4**, the polymer-rich phases contained a large amount of water even after phase separation. It was also found that the W_{aq} was almost constant regardless of the buffer concentration. In general, hydrophobic polymers that are phase-separated from water may swell slightly in water, but are substantially insoluble in water. It is not common for a hydrophobic polymer to contain 59 wt% water like poly([P_{4446}][MC3S]). This difference between hydrophobic polymer and poly([P_{4446}][MC3S]) might be due to the degree of hydrophobicity, PIL after the phase transition is insufficiently hydrophobic. It is also possible that the PIL may form a water containing structure during aggregation. In IL/water mixtures, similar trends have been reported. When the water content of hydrophobic IL and that of temperature-responsive IL after phase transition were evaluated, the number of water molecules per one ion pair of IL was the function of polymer structure. In addition, the water content of the hydrophobic IL was independent from temperature, but the water content of the temperature-responsive IL increased with decreasing temperature.

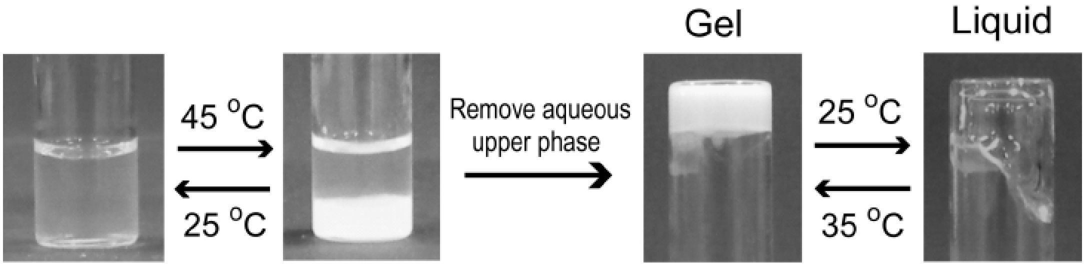


Table 2-4 The T_c value of poly([P_{444}VB][C5S]) in pure water determined by turbidity measurement.

The salt concentration of phosphate buffer	W_{aq}/wt%
0	59±3
100	51±2
200	60±1
300	58±2

2.5 Changes in aggregation state of PIL in water

The phase transition mechanism and the phase state of the thermoresponsive PIL in water are still not clear. The previous discussion suggested that the aggregation state and aggregation size of PIL depended on the ionic structure because poly([P_{4446}][MC3S])/water mixtures showed a rapid change in transmittance above the T_c, while the change in transmittance of poly([N_{444}VB][C6S])/water mixture was much slower. In order to design PILs showing highly temperature sensitive phase transition with water, it is necessary to understand the aggregation states of PILs showing a rapid change in transmittance above the T_c. Therefore, microscopic observation and dynamic light scattering measurements were carried out in order to observe changes in the aggregation state of several PILs in water with temperature change.

Effect of polymer concentration

After adding a dye (CBB), which is selectively soluble in IL, to a poly([P_{4446}][MC3S])/water mixture, the change in the solution state upon increasing the temperature was observed by light microscopy (× 400) (**Figure 2-7**). The poly([P_{4446}][MC3S])/water mixture with polymer concentration of 20 wt%, was a homogeneous solution below the phase transition temperature. However, the sea-island structure appeared in the solution when heated up to near the phase transition temperature (36 °C). Since there was no blue color derived from the dye in the droplet and the surrounding liquid phase was colored blue, it was strongly suggested that the droplet was water phase and formed a water-in-polymer type (W/P) emulsion. Generally, in a sea-island structure, a minor volume component of the dispersed phase has a smaller contact area with the continuous phase, and the interfacial free energy is lowered so that the minor volume component is dispersed in the larger volume component. However, it is challenging to consider that water is a small volume component, even if the specific gravity of the resin is generally more significant than that of water. Taking into account the water content of the polymer-rich phase in the sol-gel transition as mentioned above, it was inferred that the polymer phase contained a large amount of water immediately after the phase transition. In the case of turbidity measurement, the transmittance of the solution was 0 up to 37 °C, while in the observation of the solution, the solution state continued to change, and the water phase gradually increased above 37 °C. The aqueous phase was large and about 50 μm in diameter at 35 °C, whereas it was about 100 μm in diameter at 45 ° C. This result was explained by the gradual release of the water contained in the polymer phase. It was suggested that the PIL/water phase separation proceeds even above the phase transition temperature, and the amount of water contained in the polymer phase decreases with increasing temperature.

A different behavior was observed in a poly([P_{4446}][MC3S])/water mixture having a polymer concentration of 10 wt% (**Figure 2-8**). The formation of a multi-phase emulsion was observed in this mixture above the phase transition temperature (**Figure 2-8 d**). In contrast to the 20 wt% solution, the size of each phase was smaller. Since no clear color was observed in either phase, it was hard to clarify the PIL phase from water phase.

Effect of alkyl chain length of counter anion

Here I mention the effect of structural difference of constituent ions on the aggregation state. Poly([P_{444}VB][C4S]), poly([P_{444}VB][C5S]), and poly ([P_{444}VB][C6S]), which are PILs having different alkyl chain lengths of anions, were mixed with water at room temperature to obtain polymer concentrations of 0.10 wt%, respectively. Clear homogeneous solutions were obtained for both poly([P_{444}VB][C4S]) and poly([P_{444}VB][C5S]), whereas poly([P_{444}VB][C6S]) partially dissolved in water but a small amount of coagulum was seen in the mixture. This clearly indicates that a small extension of the alkyl groups of sulfonate anions drastically affects the hydrophilicity of the

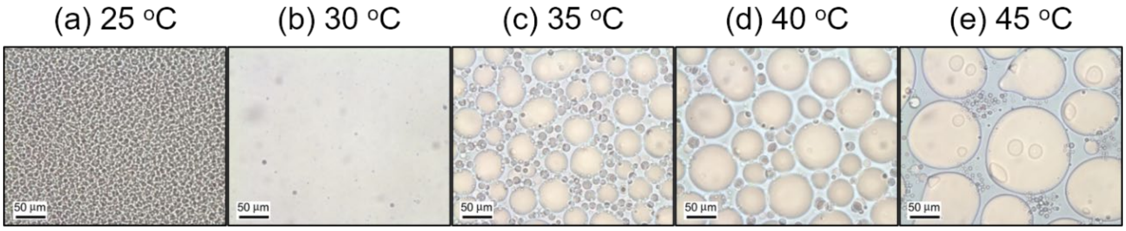


Figure 2-7 Optical microscope images of poly([P_{4446}][MC3S]) with pure water upon heating. The polymer concentration was 20 wt%.

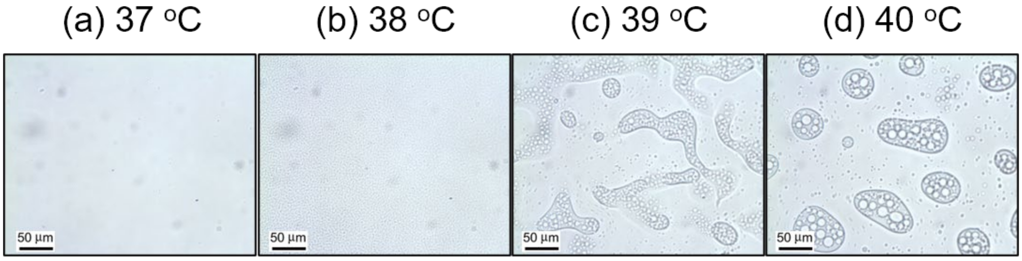


Figure 2-8 Optical microscope images of poly([P_{4446}][MC3S]) with pure water upon heating. The polymer concentration was 10 wt%.

corresponding PILs. After filtration of these solutions, transmission electron microscopy (TEM) was applied to determine the morphology of the PILs in water. As shown in **Figure 2-9**, polydispersed aggregates were seen for all PILs but their shapes and sizes depended strongly on the alkyl chain length. Amorphous aggregates with a size less than 100 nm were observed in the case of poly([P_{444}VB][C4S]). On the other hand, spherical aggregates were found in both poly([P_{444}VB][C5S]) and poly([P_{444}VB][C6S]). Some poly([P_{444}VB][C5S]) aggregates with slightly larger sizes (over 100 nm) than those of poly([P_{444}VB][C6S]) were also found. Although these TEM samples were completely dried and their sizes might be different from that in an aqueous solution, it proved that spherical charged nanoparticles were formed by the PILs with the anions having suitable alkyl chains.

Change in aggregation state before and after phase transition of LCST type observed by DLS

In order to offer further insight into the phase change of thermoresponsive PIL, a temperature-dependent dynamic light scattering (DLS) study was conducted on a 3.0 wt% aqueous poly([P_{444}VB][C5S]) mixture (**Figure 2-10**). At and below 35 ^{o}C, a broad scattering profile was observed, and the measurement could not fix the average particle size. On the other hand, a different scattering intensity was observed at both 40 and 45 ^{o}C. The particle sizes observed at 45 ^{o}C were 70 and 1000 nm. The solid and monodisperse scattering around 1000 nm affected the color change of the solution. This result strongly suggests that poly([P_{444}VB][C5S]) nanoparticles fused into micrometer-sized unimodal aggregates upon heating.

Then microscopy was carried out to support this result (**Figure 2-11**). Below the T_c, there was no object seen in the mixture. On the other hand, a number of unimodal and spherical particles were seen above the T_c. The particles disappeared upon lowering the temperature below the T_c.

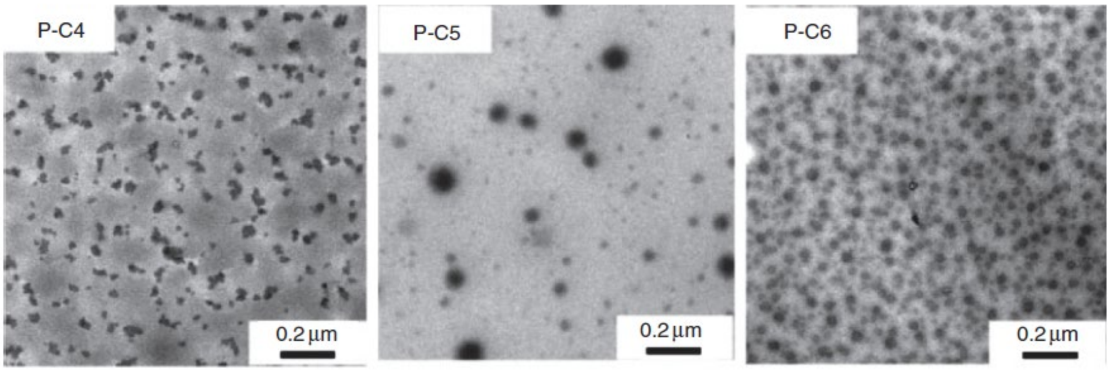


Figure 2-9 Transmission electron microscopy images for aqueous dispersions of poly([P_{444}VB][C4S]) (left)、poly([P_{444}VB][C5S]) (center)、poly([P_{444}VB][C6S]) (right).

According to this microscopic analysis, poly([P_{444}VB][C5S]) was found to display rapid growth into microspheres upon an LCST-type phase transition, which resembles coacervate formation. The formation of coacervates by the LCST-type phase transition is often seen in some non-ionic polymers[16,17].

However, our IL-derived polyelectrolytes in this study are unique beyond the classical thermoresponsive polymers since they form spherical nanoparticles in water even below the T_c, and the particle size can be tuned by solution temperature. This result means that PILs are not completely soluble in water below the phase transition temperature like PNIPAM, suggesting that the phase transition behavior mechanism is completely different from that of conventional nonionic temperature-responsive polymers. In addition, the property of PILs that the size of spherical nanoparticles can be controlled by temperature below the T_c may lead to new applications.

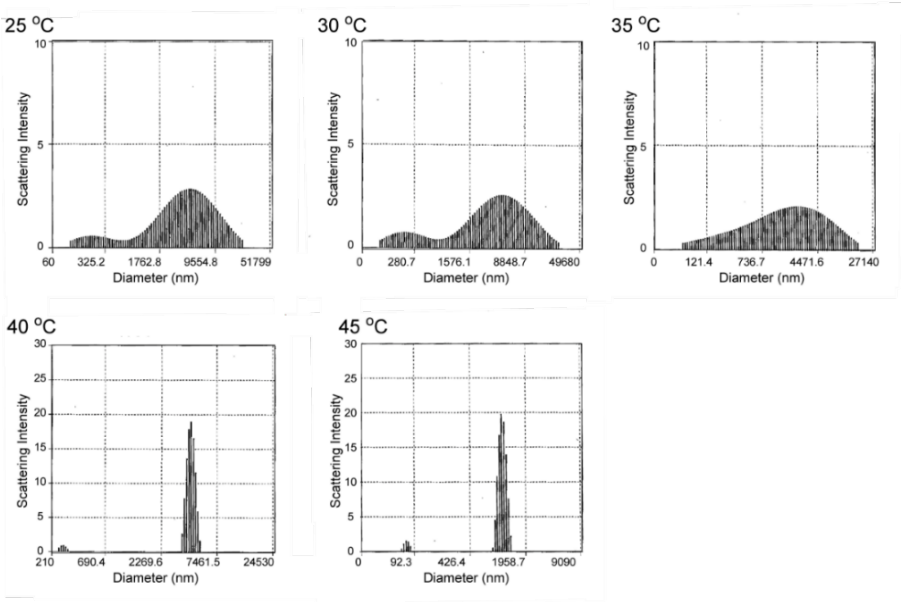


Figure 2-10 DLS profiles for poly([P_{444}VB][C5S]) (3.0 wt%) upon increasing temperatures.

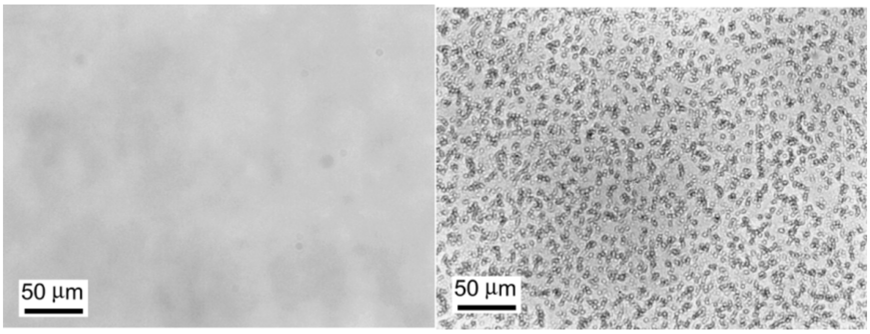


Figure 2-11 Optical microscope images for poly([P_{444}VB][C5S]) (5.0 wt %) at 30 °C (left) and after heating at 40 °C (right) at a magnification of 400 times.

2.6 Conclusion

In this chapter, some thermoresponsive IL monomers and PIL were prepared and evaluated. Some IL monomers showing LCST-type phase transition with water were polymerized in order to obtain PIL. Most of PIL prepared using ILs showing LCST-type phase transition with water remained thermo-responsivity. The T_c value of PILs was strongly affected by hydrophobicity of component ion as well as IL showing LCST-type phase transition with water. The PIL/water mixture showing LCST-type phase transition became turbid above the T_c. In the case of poly([P_{4446}][MC3S]), when the aqueous mixture was kept above the T_c for 15 min, the mixture separated into two phases and a sol-like PIL-rich phase was obtained. Phosphate buffer accelerated the phase separation rate of the PIL/water mixture. The resulting polymer-rich phase underwent reversible sol-gel transition with a slight change in temperature. The polymer rich phase contained 59 wt% water, suggesting that PIL after phase transition was not completely hydrophobic. Changes in the aggregation state of several PILs in water with temperature change was also evaluated. TEM measurements suggested poly([P_{444}VB][C5S]) showing LCST-type phase transition with water forms aggregates even below the phase transition temperature in water. According to DLS and microscopic measurement, poly([P_{444}VB][C5S]) was found to display rapid growth into microspheres upon an LCST-type phase transition, which resembles coacervate formation.

2.7 Reference

1) H. Ohno, and K. Ito,. "Room-Temperature Molten Salt Polymers as a Matrix for Fast Ion Conduction",. *Chemistry Letters*, **1998**, 27, 751–752.

2) W. Lu, A. G. Fadeev, B. Qi, E. Smela, B. R. Mattes, J. Ding, G. M. Spinks, J. Mazurkiewicz, D. Zhou, G. G. Wallace, D. R. MacFarlane, S. A. Forsyth, and M. Forsyth,. "Use of ionic liquids for π-conjugated polymer electrochemical devices",. *Science*, **2002**, 297, 983–987.

3) S. Supasitmongkol, and P. Styring,. "High CO2 solubility in ionic liquids and a tetraalkylammonium-based poly(ionic liquid)",. *Energy & Environmental Science*, **2010**, 3, 1961–1972.

4) J. Yuan, S. Soll, M. Drechsler, A. H. E. Müller, and M. Antonietti,. "Self-assembly of poly(ionic liquid)s: Polymerization, mesostructure formation, and directional alignment in one step",. *Journal of the American Chemical Society*, **2011**, 133, 17556–17559.

5) Y. Kohno, and H. Ohno,. "Key Factors to Prepare Polyelectrolytes Showing Temperature-Sensitive Lower Critical Solution Temperature-type Phase Transitions in Water",. *Australian Journal of Chemistry*, **2012**, 65, 91–94.

6) H. G. Schild,. "Poly(N-isopropylacrylamide): experiment, theory and application",. *Progress in Polymer Science*, **1992**, 17, 163–249.

7) H. Yoshimitsu, A. Kanazawa, S. Kanaoka, and S. Aoshima,. "Well-Defined Polymeric Ionic Liquids with an Upper Critical Solution Temperature in Water",. *Macromolecules*, **2012**, 45, 9427–9434.

8) K. J. Fraser, D. R. MacFarlane, K. J. Fraser, and D. R. MacFarlane,. "Phosphonium-Based Ionic Liquids: An Overview",. *Australian Journal of Chemistry*, **2009**, 62, 309–321.

9) K. Tsunashima, and M. Sugiya,. "Physical and Electrochemical Properties of Room Temperature Ionic Liquids Based on Quaternary Phosphonium Cations",. *Electrochemistry*, **2007**, 75, 734–736.

10) Y. Kohno, and H. Ohno,. "Temperature-responsive ionic liquid/water interfaces: relation between hydrophilicity of ions and dynamic phase change",. *Physical chemistry chemical physics : PCCP*, **2012**, 14, 5063–5070.

11) Y. Kohno, H. Arai, S. Saita, and H. Ohno,. "Material design of ionic liquids to show temperature-sensitive LCST-type phase transition after mixing with water",. *Australian Journal of Chemistry*, **2011**, 64, 1560–1567.

12) S. Sugihara, K. Hashimoto, S. Okabe, M. Shibayama, S. Kanaoka, and S. Aoshima,. "Stimuli-Responsive Diblock Copolymers by Living Cationic Polymerization: Precision Synthesis and Highly Sensitive Physical Gelation",. *Macromolecules*, **2004**, 37, 336–343.

13) A. Blanazs, R. Verber, O. O. Mykhaylyk, A. J. Ryan, J. Z. Heath, C. W. I. Douglas, and S. P. Armes,. "Sterilizable Gels from Thermoresponsive Block Copolymer Worms",. **2012**,.

14) Z. Chu, and Y. Feng,. "Thermo-switchable surfactant gel.",. *Chemical communications (Cambridge, England)*, **2011**, 47, 7191–3.

15) J. C. Ribot, C. Guerrero-Sanchez, R. Hoogenboom, and U. S. Schubert,. "Thermoreversible ionogels with tunable properties via aqueous gelation of an amphiphilic quaternary ammonium oligoether-based ionic liquid",. *Journal of Materials Chemistry*, **2010**, 20, 8279.

16) D. L. Nettles, A. Chilkoti, and L. A. Setton,. "Applications of elastin-like polypeptides in tissue engineering",. *Advanced Drug Delivery Reviews*, **2010**, 62, 1479–1485.

17) H. Miyazaki, and K. Kataoka,. "Preparation of polyacrylamide derivatives showing thermo-reversible coacervate formation and their potential application to two-phase separation processes",. *Polymer*, **1996**, 37, 681–685.

Chapter 3

Control of phase transition temperature of poly(ionic liquid) in water

3.1 Introduction

Through the design of their ion structure, I have prepared thermoresponsive PIL/water mixtures. PILs showing LCST-type-phase transition with water have a great potential to be used for various applications such as purification and extraction due to their temperature sensitive responsivity. In order to apply the temperature-responsive PIL for various purpose, it is necessary to control the phase transition temperature. As described in chapter 2, phase behavior and T_c value of PIL strongly depended on the total hydrophobicity of component ions. However, it should be limited to finely control the T_c value by only designing their component ions.

There are a few ways known to control the T_c value in the IL/water mixture system. For example, Ohno *et al.* reported that the T_c value of the thermoresponsive IL/water mixture can be control by the concentration of IL.[1] Mixing ILs with different hydrophobicity was also found to be effective in controlling the T_c value. It is expected that the T_c value of the PIL/water mixture could be controlled in the same way. In this chapter, I would like to discuss how to finely control the T_c value of PIL by the methods which were effective in the IL/water system. In addition, the difference between copolymer and blend was pointed out.

3.2 Experimental

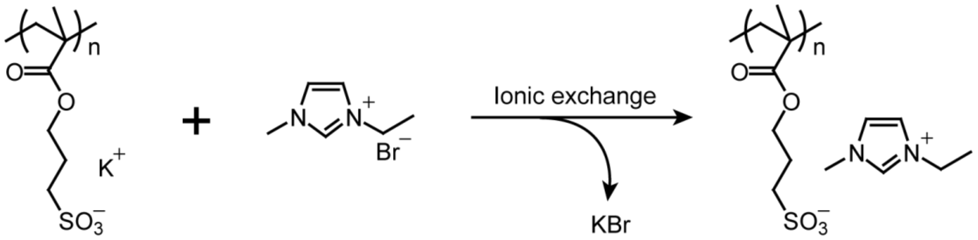
Preparation of PILs

The PILs which composed of single IL monomer species such as (poly(tetrabutylphosphonium 3-sulfopropyl-methacrylate) (poly([P_{4444}][MC3S])), poly(tributylhexylphosphonium 3-sulfopropyl-methacrylate) (poly([P_{4446}][MC3S])), poly(tributylvinylbenzylphosphonium 3-sulfopropyl-methacrylate) (poly([P_{444}VB][C5S])), and poly(tributyloctylphosphonium 3-sulfopropylmethacrylate) (poly([P_{4448}][MC3S])) were prepared according to the methods as described in chapter 2.

Copolymer of [P_{4446}][MC3S] and [P_{4448}][MC3S] was prepared using solution in which corresponding IL monomers are mixed at different molar ratio, and polymerized. Polymerization is the same procedure as that in the chapter 2. The name of the copolymer was described as poly([P_{4446}][MC3S]$_{1-x}$-*co*-[P_{4448}][MC3S]$_x$); x = 0, 0.1, 0.2, 0.3, 0.4.

Poly(1-methyl-3-ethyl imidazolium 3-sulfopropylmethacrylate) (poly([C_2mim][MC3S])) was prepared by ionexchange method. First, 3-sulfopropylmethacrylate potassium (K[MC3S]) was polymerized to obtain poly(K[MC3S]). Then, 1-methyl-3-ethyl imidazolium bromide ([C_2mim]Br) was synthesized by quaternization reaction of 1-methyl imidazole and ethyl bromide. Then 1.35 fold mole of [C_2mim]Br was added to the poly(K[MC3S]) aqueous solution and stirred vigorously for more than one night to allow the ion-exchange reaction. The prepared poly([C_2mim][MC3S]) was purified by dialysis and freeze-drying.

One or two types of the obtained PILs were mixed at different molar ratios in pure water, salt solution, or buffer and stirred for more than 48 hours to give a homogeneous solution. The resulting solution was filtered using a Tefron® filter (Hole Diameter 0.20 µm, DISMIC -13 HP045AN, Toyo Roshi Kaisha Ltd.).



Scheme 3-1 Ionic exchange between poly(K[MC3S]) and [C_2mim]Br in order to obtain poly-([C_2mim][MC3S]).

Preparation of NaCl solution

NaCl purchased from Kanto Kagaku Co., Ltd. was used without purification. First, 7.01 g of NaCl was dissolved in 200 mL of pure water to prepare a 600 mM NaCl aqueous solution. Then the solution was diluted to prepare 300, 150, and 75 mM NaCl aqueous solution.

Preparation of Phosphate Buffer

KH_2PO_4 was purchased from Wako Pure Chemical Industries, Ltd. and K_2HPO_4 was purchased from Kanto Kagaku Co., Ltd. Each reagent was used without purification. Each solution of KH_2PO_4 and K_2HPO_4 were prepared and mixed to a prepare phosphate buffer of the desired pH. The pH was measured with a pH meter (InLab ® Routine Pro, METTLER TOLEDO International Inc.).

GPC measurement

The weight-average molecular weight (M_w) and the number-average molecular weight (M_n) of the prepared PIL was measured by gel permeation chromatography (GPC) measurement using water/acetonitrile mixed (water/acetonitrile = 6/4) containing 50 mM LiCl as an eluent. The GPC curve was recorded by Shimadzu instruments equipped with Shimadzu SPD-20A UV-vis detector (λ = 215 nm), LC-20AD solvent delivery unit, and a poly(vinyl alcohol) gel column (Shodex Asahipak GF-7M HQ, 0.4 ml min^{-1}). The column was heated at 40 ^{o}C using a column oven (Shimadzu CTO-20AC). A calibration curve was prepared using PSS as a standard sample. Polydispersity index (PDI) is defined as M_w/M_n and is an indicator of the wideness of the molecular weight distribution.

Table 3-1 M_n value, M_w value, and PDI of PILs used in this study.

	M_n	M_w	PDI
Poly([P_{4444}][MC3S])	2.6×10^5	4.1×10^5	1.54
Poly([P_{4446}][MC3S])	7.9×10^4	3.6×10^5	4.50
Poly([P_{4448}][MC3S])	3.8×10^5	8.7×10^5	2.29
Poly([C_2mim][MC3S])	1.7×10^5	9.9×10^5	5.78

Evaluation of phase transition behavior

The phase transition behavior of the PIL/water mixtures was evaluated by the turbidity change of the solution. For turbidity measurement, an ultraviolet-visible V-660 of Japan Spectroscopy Co., Ltd. was used. The measurement was performed using a plastic cell with an optical path length of 10 mm, under following conditions; a wavelength of 600 nm, and a bandwidth of 2 nm. The phase transition temperature (T_c) was defined as the temperature at which the transmittance decreased to 90% under the heating speed of 2 ^{o}C/min.

3.3 Factors affecting phase transition temperature

3.3.1 Concentration of PIL

A concentration dependence of the phase transition behavior of PIL was first evaluated. **Figure 3-1** shows changes in the transmittance of aqueous mixtures containing different amounts of poly([P_{4446}][MC3S]), determined by UV-vis spectroscopy. The T_c value was determined as the temperature at which the transmittance of the mixture fell below 90%. The transmittance of the mixture fell immediately when temperature close to the T_c. This sharp change in transmittance was one of the characteristic properties of suitably designed PILs. The T_c value decreased from 54 to 36 °C upon increasing the concentration of poly([P_{4446}][MC3S]) from 2.5 to 20 wt%, The T_c value of monomeric and polymerized [P_{4446}][MC3S] was 44 (see chapter 2 **Table 2-1**) and 40 °C (water content was 90 wt%), respectively(**Table 3-2**). The T_c value of poly([P_{4446}][MC3S])/water mixture were inversely proportional to the natural logarithm of the concentration of poly([P_{4446}][MC3S]).

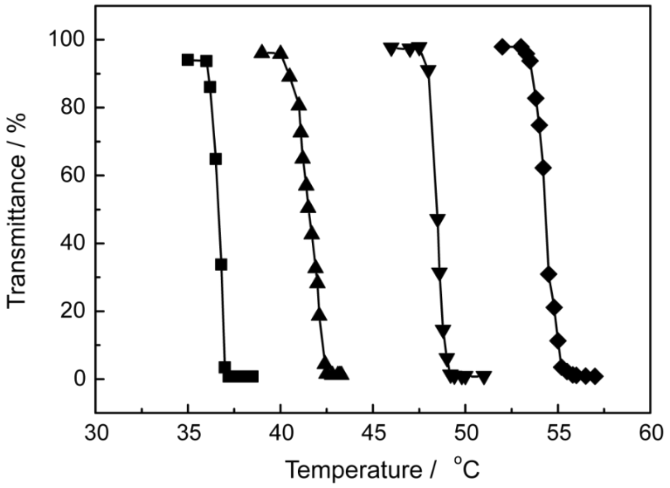


Figure 3-1 Temperature-dependent change in transmittance of poly([P_{4446}][MC3S]) in pure water upon heating. The polymer concentration was 20 (■), 10 (▲), 5 (▼), and 2.5 wt% (◆).

Table 3-2 The T_c value of poly([P_{4446}][MC3S]) in pure water determined by turbidity measurement and DSC measurement.

Polymer concentration / wt%	T_c / turbidity	T_c / DSC
2.5	54	-
5	48	48
10	40	42
20	36	37

The phase transition temperature of PNIPAM remains constant in the solution concentration range of 0.01 to 1 wt%, but phase change was slow when the concentration was lower. This is due to the fact that the coil-globule transition of PNIPAM takes around 30 microseconds, while the phase separation of aqueous solutions takes much longer as several milliseconds to several hundred milliseconds. That is, the phase separation is the rate-determining step of the entire process of temperature response. As the distance between molecules decreases with increasing concentration, the polymers aggregates earlier . On the other hand, this tendency was not observed in poly([P_{4446}][MC3S])/water mixture. The T_c value changed with PIL concentration, but the temperature responsiveness did not change. It suggested that the phase transition of PIL differs from that of PNIPAM. The phase transition of PIL takes longer than the phase separation of aqueous solution and that phase transition would be the rate controlling process of the whole process. In addition, the findings that the phase transition of PILs is affected by the intermolecular distance suggested that the phase transition does not occur with a single pair of side chains and counterions. It is likely that the side chains and counterions interact with each other in a wide range of intermolecular distances.

The concentration dependence of the phase behavior was also evaluated using poly([P_{444}VB][C5S]). A poly([P_{444}VB][C5S])/water mixture was prepared at a lower polymer concentration than the poly([P_{4446}][MC3S])/water mixture because of its high viscosity. As a result, the solutions with PIL concentration of 5 and 4 wt% showed a rapid change in transmittance at temperature near T_c. However, the decrease in transmittance became slow as the concentration of PIL

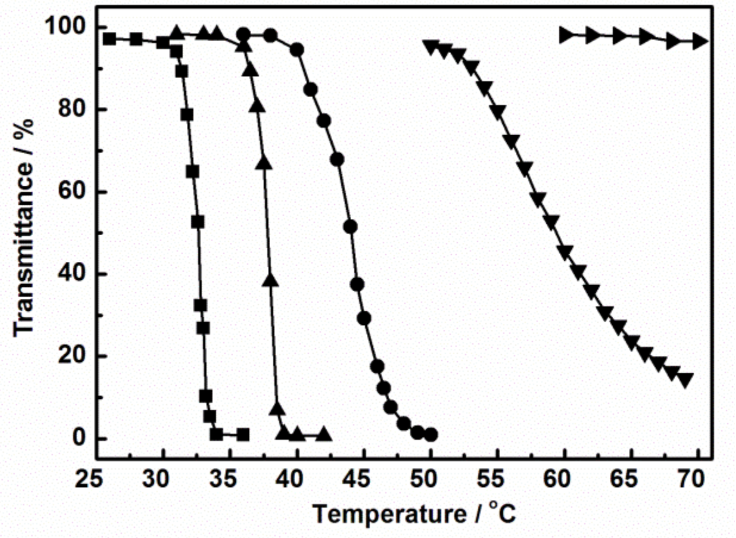


Figure 3-2 Temperature-dependent transmittance change ($\lambda = 600$ nm) of poly([P_{444}VB][C5S])/water mixtures upon heating. The polymer concentration (wt%) was 5 (■), 4 (▲), 3 (●), 2 (▼) and 1(►). Heating rate was 1 °C/min.

decreased to 3 wt% (**Figure 3-2**). The turbidity of the solution with a PIL concentration of 1 wt% was hardly changed (as seen in **Figure 3-2**; ▲) even heated up to 70 ^{o}C. The concentration-dependent in phase transition temperature of poly([P_{444}VB][C5S]) was more distinct than that of poly-([P_{4446}][MC3S]) (**Table 3-3**). It was also found that poly([P_{444}VB][C5S]) had a large concentration dependence on the phase transition behavior of PIL. The phase transition behavior of poly([P_{444}VB][C5S]) is likely to be affected by the average intermolecular chain distance. Also in ([P_{444}VB][C5S]) system, The T_c value of PIL/water mixture were inversely proportional to the natural logarithm of the concentration of PIL as well as poly([P_{4446}][MC3S])/water mixture. It was found that there is a fundamental property that the natural logarithm of the PIL concentration is inversely proportional to the T_c value of PIL/water mixture.

Figure 3-3 is photographs of a poly([P_{444}VB][C5S])/water mixture with a PIL concentration of 3 wt%. As the solution is heated/cooled, the turbidity of the solution was slightly changed. Above 45 ^{o}C, the phase transition temperature, the solution became opaque and bluish. Upon cooling to 35 °C, the solution turned transparent again. No precipitation was observed in the solution within the temperature range analyzed. Reversibility of phase behavior was also confirmed by transmittance measurements.

Table 3-3 The T_c value of poly([P_{444}VB][C5S]) in pure water determined by turbidity measurement.

Polymer concentration / wt%	T_c / turbidity
1.0	Not detected
2.0	53
3.0	41
4.0	37
5.0	32

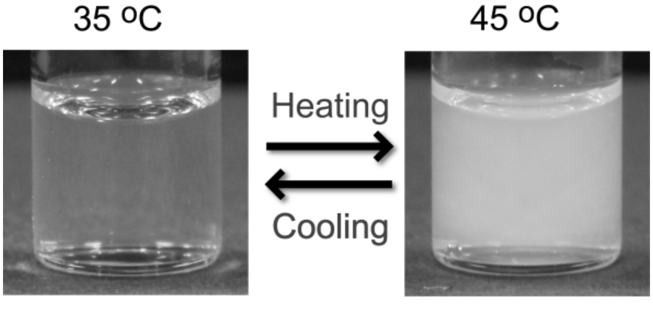


Figure 3-3 Visual appearances of poly([P_{444}VB][C5S]) (3 wt% in pure water) upon varying temperatures from 35 ^{o}C to 45 ^{o}C. The transmittance of solution was c.a. 50 % at 45 ^{o}C.

3.3.2 Salt strength

The effect of adding salt to the PIL/water mixture on its phase transition behavior was evaluated using NaCl and phosphate buffer. Firstly poly([P_{4446}][MC3S]) was dissolved in NaCl aqueous solution containing various concentration of NaCl. The PIL concentration of the solution was fixed to 5 wt%. The transmittance of the solution was measured upon heating (**Figure 3-4**). In the range of salt concentration of 0 ~ 600 mM, all the solutions showed the LCST-type phase transition, and the transmittance decreased with increasing temperature. The transmittance rapidly decreased to almost 0% at a temperature change of only about 2 °C near T_c. There are no salt strength dependence in the decreasing manner. The T_c value increased with increasing NaCl concentration in the solution. In 600 mM NaCl aqueous solution, equivalent to seawater, showed 6 °C higher T_c than that in pure water.

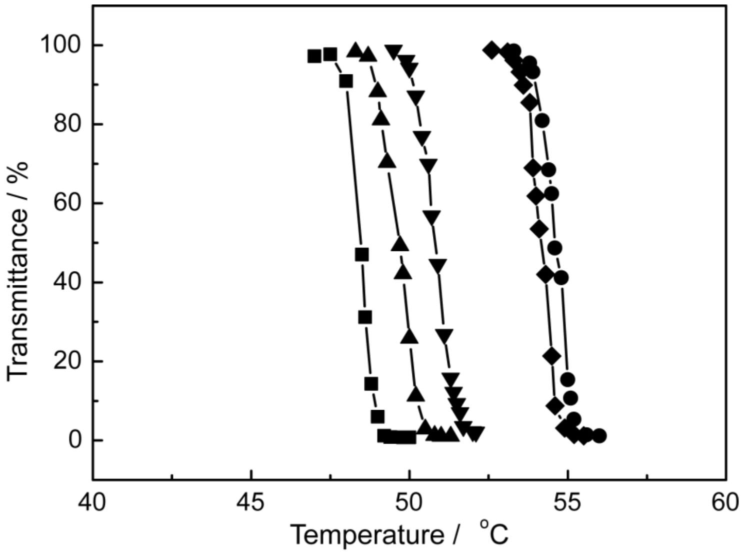


Figure 3-4 Change in transmittance of 5 wt% poly([P_{4446}][MC3S]) after mixing with phosphate buffer having differing salt concentrations; salt concentration was 0 (■), 75 (▲), 150 (▼), 300 (◆), and 600 mM (●).

Table 3-4 The T_c value of poly([P_{4446}][MC3S]) in NaCl solution having different salt concentrations determined by turbidity measurement.

Salt concentration	**T_c / turbidity**
0	48
75	49
150	50
300	54
600	54

In anticipation of its use in the biomaterials field, phosphate buffer containing KH_2PO_4/K_2HPO_4 was essentially used (pH = 7.0, polymer concentration 20 wt%). The T_c value of poly([P_{4446}][MC3S])/buffer mixture was much lower than that in pure water while maintaining the same sensitivity of the phase transition (**Figure 3-5**). **Table 3-5** shows that T_c was 33, 27, and 23 °C when poly([P_{4446}][MC3S]) was mixed with phosphate buffers at salt concentrations of 100, 200, and 300 mM, respectively. The buffer concentration and the T_c value of poly([P_{4446}][MC3S])/buffer mixture were simple proportional. This trend was different from the relationship between the PIL concentration and the T_c value of PIL/water mixture. It is considered that the mechanism by which buffer concentration controls the T_c value of mixture is different from that of PIL concentration.

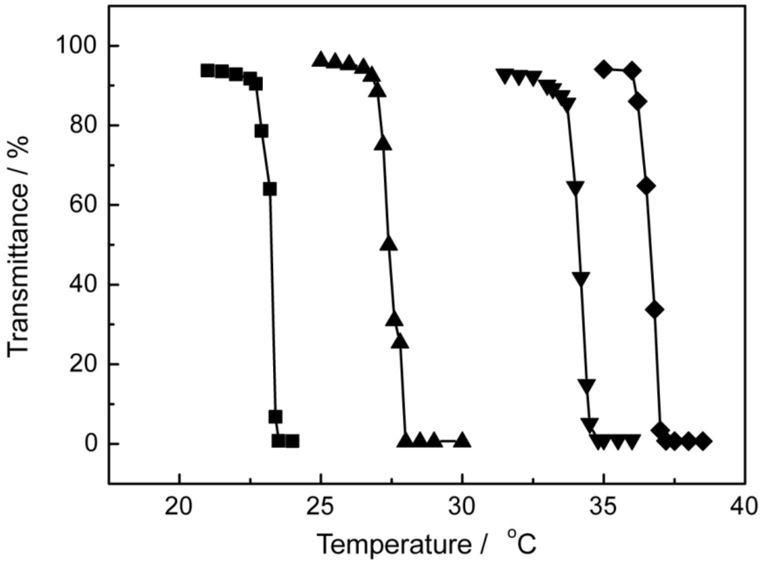


Figure 3-5 Change in transmittance of 20 wt% poly([P_{4446}][MC3S]) after mixing with phosphate buffer having difference salt concentrations; salt concentration was 0 (◆), 100 (▼), 200 (▲), and 300 (■).

Table 3-5 The T_c value of poly([P_{4446}][MC3S]) in phosphate buffer having different salt the concentration determined by turbidity measurement.

Salt concentration	***T*c / turbidity**
0	36
100	33
200	27
300	23

Yuan *et al.* reported that the T_c value for poly([P_{4444}][SS]) increased dramatically upon adding KBr.[2] This trend is opposite to that observed upon adding KH_2PO_4/K_2HPO_4 salts, implying that the kosmotropicity of the salts strongly influences the T_c value. KH_2PO_4/K_2HPO_4 become kosmotropic ions in water and the ions order the structure of water. Accordingly a salting-out effect leads to the dehydration of PIL which cause the phase transition temperature of PIL getting lower. On the contrary, KBr and NaCl become caotropic ions in water and cause a salting-in effect which increase the phase transition temperature of PIL. therefore, the T_c value of the aqueous PIL solution was proportional to the concentration of the added salt. The result that the T_c value was the same when NaCl concentration was 300 mM and 600 mM was probably because the salting-in effect reached the upper limit.

3.4 Control of Phase Transition Temperature through Copolymerization of Ionic Liquid Monomers with Different Hydrophobicity

It is necessary to precisely control the total hydrophilicity of component ions in order to design ILs that showing the LCST-type phase transition with water. It has also been suggested in the previous chapter that the total hydrophilicity of component ion of PIL affects their LCST-type phase transition with water in the same way. However, it is difficult to precisely control their hydrophilicity only by the ion design and to tune the phase behavior and T_c. In monomeric IL systems, Ohno et al. has achieved successive control of T_c by mixing ILs with different hydrophobicity. Based on this finding, I have attempted to control the phase behavior and T_c of PIL by copolymerization.

First of all, [P_{4446}][MC3S] and [P_{4448}][MC3S] were mixed at the desired ratio and then polymerized. The obtained PIL described as poly([P_{4446}][MC3S]$_{1-x}$-*co*-[P_{4448}][MC3S]$_x$ (x = 0, 0.1, 0.2, 0.3, or 0.4)) (**Figure 3-6**). The copolymer was mixed with pure water to obtain a PIL concentration of 5 wt%, and the phase behavior with temperature change was analyzed. All the prepared copolymer solutions showed the temperature response and became turbid with increasing temperature. As shown in **Figure 3-7**, contrary to expectations, the phase transition sensitivity of each solution was the same regardless of the copolymerization ratio. It was suggested that even when PILs which are not temperature-responsive were copolymerized, they would undergo a phase transition in response to temperature if the total hydrophilicity of entire PILs was in the appropriate range. This result also supported the previous argument that the side chains and counterions interact over a wide range of intermolecular distances. Such sensitivity was not observed in the conventional non-ionic thermo-responsive polymers which copolymerized with hydrophobic monomer in order to control the T_c value[3,4]. The T_c value of each PIL/water mixture decreased steadily with increasing the copolymerization ratio (**Table 3-6**). It was suggested that the hydrophobicity of component ions of IL monomer affected the phase transition temperature of PIL/water mixture as well as the IL/water mixture and the T_c value of PIL can control by the copolymerization ratio of IL monomers having different hydrophobicity.

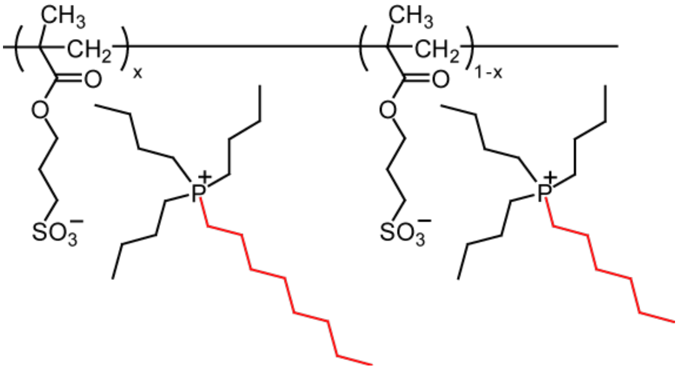


Figure 3-6 Structure of poly([P_{4446}][MC3S]$_{1\text{-}x}$-*co*-[P_{4448}][MC3S]$_x$).

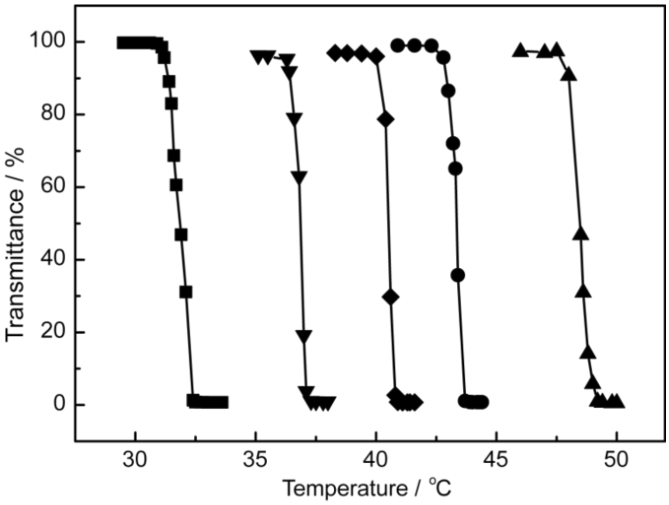


Figure 3-7 Change in transmittance of poly([P_{4446}][MC3S]$_{1\text{-}x}$-*co* -[P_{4448}][MC3S]$_x$) in water upon heating. The x value was 0 (▲), 0.1 (●), 0.2(◆), 0.3(▼), and 0.4(■).

Table 3-6 The T_c value of of poly([P_{4446}][MC3S]$_{1\text{-}x}$-*co*-[P_{4448}][MC3S]$_x$) (x = 0, 0.1, 0.2, 0.3, or 0.4) in pure water.

x value	***T*$_c$ / turbidity**
0	48
0.1	43
0.2	40
0.3	36
0.4	31

3.5 Control of Phase Transition Temperature by blending Ionic Liquid-Derived Polyelectrolytes having Different Hydrophobicity

The PIL is composed of polyions and counterions, and the thermal responsiveness is affected by affinity and hydrophobicity, as described in the previous chapter. When several IL monomers are copolymerized, ion exchange may occur on the polymer chains. If this assumption is true, the T_c can be controlled very easily just by mixing. To verify the assumption, poly([P_{4446}][MC3S]) and poly([P_{4448}][MC3S]) were prepared and mixed. The PILs were mixed in desired molar ratios and were dissolved in water to give 5 wt% solution. When each solution was heated up to 50 °C, all solutions showed temperature response and became turbid (**Figure 3-8**). The change in transmittance with the increasing temperature of the solution was measured. The transmittance of the solution decreased rapidly near T_c. When the T_c value of the resulting mixture was compared with that of the aforementioned copolymer, both the copolymer and the mixture exhibited approximately the same T_c value when the ratios of [P_{4446}][MC3S] were the same(**Table 3-7**).

The T_c value was the function of the hydrophobic PIL fraction in the mixture. This result clearly indicated that counterion exchange is occurred among poly([P_{4446}][MC3S]) and poly([P_{4448}]-[MC3S]) in the mixture. Otherwise, the T_c value of the mixture should increase as the concentration of poly([P_{4446}][MC3S]) decreases with the increase in the amount of poly([P_{4448}][MC3S]) added. It is also suggested that the T_c value of the PIL/water mixture depends on the total hydrophobicity of the PILs in the solution and that T_c value can be controlled by only mixing PILs, if the hydrophobicity is appropriately adjusted.

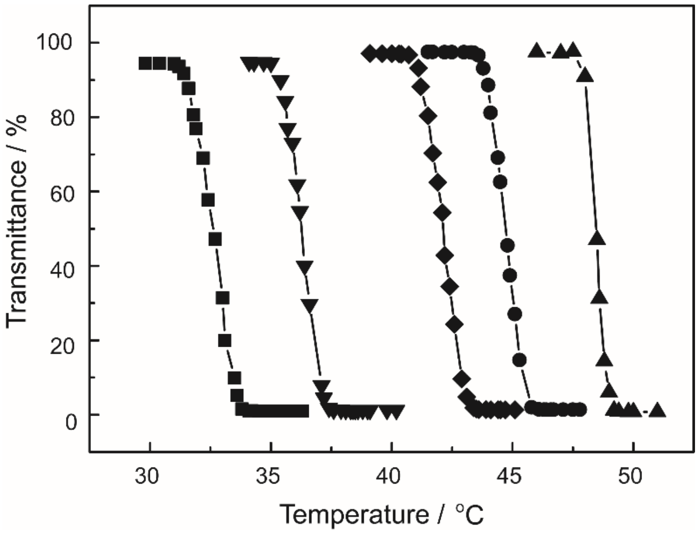


Figure 3-8 Change in transmittance of poly([P_{4446}][MC3S])/ poly([P_{4448}][MC3S]) blends in water upon heating. the mole fraction of poly([P_{4448}][MC3S]) was 0 (▲), 0.1 (●), 0.2(◆), 0.3(▼), and 0.4(■).

Table 3-7 The T_c value of of poly([P_{4446}][MC3S]$_{1-y}$/[P_{4448}][MC3S]$_y$) (y = 0, 0.1, 0.2, 0.3, or 0.4) in pure water.

y value	**T_c / turbidity**
0	48
0.1	44
0.2	41
0.3	35
0.4	31

Based on the above results, hydrophilic PIL (poly([P_{4444}][MC3S]) and hydrophobic PIL (poly([P_{4448}][MC3S])) were mixed to obtain the mixture showing LCST-type phase transition. **Figure 3-9** shows a transmittance change of the resulting PILs/water mixtures. When the molar ratio varied from equimolar to 4 : 6 or 6 : 4, all the mixture remains LCST-type phase transition. The T_c value of the solutions increased with increasing the ratio of hydrophobic PIL (**Table 3-8**). The sharpness of the phase transition of PILs/water was as sharp as that of poly([P_{4446}][MC3S])/water mixture. This result clearly demonstrates the importance of the total hydrophobicity of ion composing PIL in the solution for showing the LCST-type phase transition.

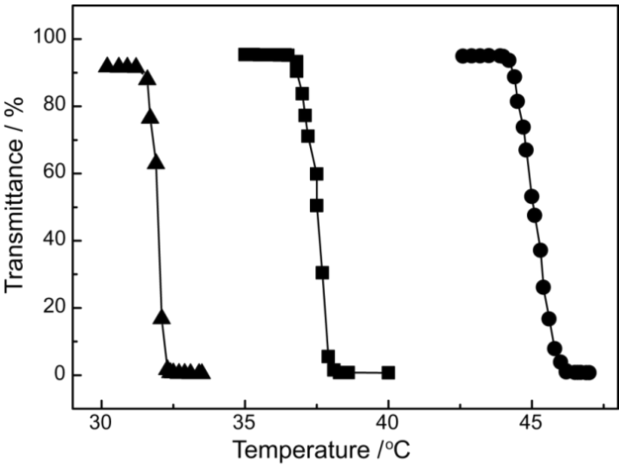


Figure 3-9 Change in transmittance of poly([P_{4444}][MC3S])/poly([P_{4448}][MC3S]) blends in water upon heating. the mole fraction of poly([P_{4448}][MC3S]) was 0.4(▲), 0.5(■), and 0.6(●).

Table 3-8 The T_c value of of poly($[P_{4444}][MC3S]_{1-z}/[P_{4448}][MC3S]_z$) (z = 0.4, 0.5, 0.6) in pure water determined by turbidity measurement.

The z value	***T*$_c$ / turbidity**
0.4	44
0.5	37
0.6	32

Finally, it should be remarked that the fine-tuning of LCST behavior, i.e., by blending hydrophobic and hydrophilic PILs, was observed not only in PILs with *n*-butyl-*n*-alkylphosphonium cations but also in PILs blended with highly hydrophilic poly($[C_2mim][MC3S]$), in which $[C_2mim]^+$ was used as the counter cation. When an equimolar amount of poly($[C_2mim][MC3S]$) was mixed with poly($[P_{4448}][MC3S]$), the resulting blended PIL assumed underwent the LCST behavior and showed the T_c value at 64 °C. This value was much higher than that observed for any other blended PILs, reflecting the hydrophilic nature of the $[C_2mim]^+$-based PIL more extensive than that of $[P_{444n}]^+$-based ones. These results clearly suggest that LCST-type phase transition is possible even in hydrophilic alkylimidazolium-based PILs upon suitably blending them with hydrophobic PILs. The property of thermoresponsive PILs make it possible to easily add further functions to the aqueous mixture by simply blending other functional PILs.

3.6 Conclusion

In summary, I have mentioned several methods to control the T_c value of the PIL/water mixture. The T_c value changed with PIL concentration, but the temperature responsiveness did not change. The natural logarithm of the PIL concentration is inversely proportional to the T_c value of PIL/water mixture. It suggested that the phase transition mechanism of PIL differs from that of PNIPAM, and the phase transition of PIL takes longer than the phase separation of aqueous solution and that phase transition would be the rate controlling process of the whole process. The findings that the phase transition of PILs is affected by the intermolecular distance suggested that the phase transition does not occur among only single pair of side chains and counterions. Adding salt is also effective and the T_c value greatly changed with increasing salt concentration. This result suggests that the ionic strength of the solution affect the phase transition behavior of the PIL. The kosmotropicity of the salts also strongly influenced the T_c value. Subsequently, when monomers having different hydrophilicity were copolymerized, the T_c value of thus prepared copolymer tended to decrease with an increase in the proportion of hydrophobic fraction of the PIL. However, unlike the case of the conventional temperature-responsive polymer, the phase transition behavior did not become low sensitive even if the ratio of the hydrophobic moiety increased. This result also supported the previous argument that the side chains and counterions interact over a wide range of intermolecular distances. At last, I showed that aqueous mixtures containing thermoresponsive poly([P_{4446}][MC3S]), which underwent the LCST-type phase transition, and water-immiscible poly([P_{4448}][MC3S]) exhibited analogous LCSTs compared to those of the copolymerized PILs. This result clearly indicated that counterion exchange is occurred among poly([P_{4446}][MC3S]) and poly([P_{4448}][MC3S]) in the mixture. I also show that the T_c values of the blended PILs strongly depended upon the total hydrophilicity of the component ions.

3.7 Reference

1) Y. Kohno, H. Arai, S. Saita, and H. Ohno,. "Material design of ionic liquids to show temperature-sensitive LCST-type phase transition after mixing with water",. *Australian Journal of Chemistry*, **2011**, 64, 1560–1567.

2) Y. Men, X.-H. Li, M. Antonietti, and J. Yuan,. "Poly(tetrabutylphosphonium 4-styrenesulfonate): a poly(ionic liquid) stabilizer for graphene being multi-responsive",. *Polymer Chemistry*, **2012**, 3, 871-875.

3) Y. G. Takei, T. Aoki, K. Sanui, N. Ogata, T. Okano, and Y. Sakurai,. "Temperature-responsive bioconjugates. 2. Molecular design for temperature-modulated bioseparations",. *Bioconjugate Chemistry*, **1993**, 4, 341–346.

4) H. Feil, Y. H. Bae, J. Feijen, and S. W. Kim,. "Effect of comonomer hydrophilicity and ionization on the lower critical solution temperature of N-isopropylacrylamide copolymers",. *Macromolecules*, **1993**, 26, 2496–2500.

Chapter 4

Design of thermoresponsive hydrogel derived from ionic liquids

4.1 Introduction

Stimuli-responsive hydrogels that undergo reversible changes in their physicochemical properties have been broadly investigated for a variety of applications, such as “smart materials”. The hydrogel prepared using non-ionic thermoresponsive polymers represented by PNIPAM are well-known as thermoresponsive materials that show a LCST-type phase transition. The phase transition of thermoresponsive hydrogels are expected to be accompanied by a large volume change, the “smart gels” have been exploited specifically for biomedical fields such as drug delivery,[1] tissue engineering,[2,3] actuators, [4] enzyme immobilizations,[5] and material separation systems.[6]

In spite of their potential for a variety of intriguing applications,[7–9] little effort was paid to fabricate thermoresponsive hydrogels composed of wholly charged polymers. In fact, there was no report on hydrogel composed of charged polymers undergoing the LCST-type phase transition until the author began to prepare smart gel derived from ILs. Since polyelectrolytes have a higher affinity for water than nonionic polymers, charged polymer gels such as sodium polyacrylate can swell more water and are widely used as superabsorbent materials. If the water content of fully charged hydrogels can be reversibly controlled by temperature, the superabsorbent material can be reused in various application fields, making it an environmentally friendly material. It is also expected that the large water content change of the charged hydrogel can be utilized to further develop applications where conventional non-ionic thermoresponsive polymer hydrogels have been used.

In this chapter, I have examined the composition and preparation condition of the hydrogel using IL monomer which shows LCST-type phase transition with water. In addition, I have investigate necessary condition to prepare PIL hydrogels that reversibly undergo absorb/desorb water molecules *via* the LCST-type phase transition.

4.2 Experimental

Materials

Tributylhexylphosphonium 3-sulfopropylmethacrylate ([P_{4446}][MC3S]), tributyloctylphosphonium 3-sulfopropylmethacrylate ([P_{4448}][MC3S]), and poly([P_{444}VB][MC3S]) were prepared according to the methods described in chapter 2. Ethylene glycol dimethacrylate (EGM) and triethylene glycol dimethacrylate (TEGDM) were purchased from Tokyo Chemical Industry Co. 1,6-Hexylene dimethacrylate (C_6M), 2,2'-azobis[2-(2-imidazoline-2-yl)propane] disulfate dehydrate (VA-046B), and ammonium persulfate (APS) were purchased from Wako Chemical Industry Co. 1,14-Tetradecanediol dimethacrylate (C_{14}M) and poly(ethylene glycol) dimethacrylate with the number-average molecular weight of 550 (PEGDM) were purchased from Sigma-Aldrich Co. 2,2'-Azobis(isobutyronitrile) (AIBN) and tetramethylene diamine (TEMED) was purchased from Kanto Chemical Co. AIBN was recrystallized with ethanol before use. All of other chemicals and solvents were used as received.

Preparation of hydrogel by chemical crosslinking

Both [P_{4446}][MC3S] and pure water were mixed to exhibit a monomer concentration of 70 wt %, and crosslinker was added. The resulting mixtures were individually placed into a vial or plastic syringe (1 mL syringe; TERUMO Co.) and degassed by ultra-sonication. A free radical initiator was then dissolved in the mixture and the vial or syringe was sealed under N_2 atmosphere. To induce gelation, the vial was heated up to 80 °C for a method where AIBN was used as an initiator, and the syringe was heated up to 50 °C for VA-046B, depending on the 10-hour half-life temperature (T_{10}) of each initiator, i.e., AIBN with T_{10} of 65 °C and VA-046B with T_{10} of 46 °C. When APS/TEMED was used, the gelation reaction was carried out at room temperature. After the gelation reactions, the resulting PIL hydrogels were removed off from the vials or syringe and soaked in pure water. After the equilibrium soaking of water into the hydrogel, the visual appearances of the resulting PIL hydrogels were confirmed. When the hydrogels did not crack after water absorption, temperature-dependent water pumping behavior was further investigated by heating the gels to induce the LCST-type phase transition

Preparation of hydrogel by electrostatic interaction

Poly([P_{444}VB][MC3S]) was used as a matrix and [P_{4446}][MC3S] was used as a monomer. AIBN was used as the polymerization initiator at 1 mol% to IL monomer. The other procedure is the same as described above.

Preparation of hydrogel using both covalent bonding and electrostatic interaction

Poly([P_{444}VB][MC3S]) was used as a matrix and [P_{4446}][MC3S] was used as a monomer. PEGDM was used as the crosslinking agent, and AIBN was used as the polymerization initiator at 1 mol% to the IL monomer. The other procedure is the same as described above.

Determination of the phase transition temperature of PIL gels (T_c')

The T_c' values were determined from characteristic endothermic peaks of LCST-type phase transition using DSC during heating the PIL hydrogels. The heating rate was 1 °C min^{-1} from 20 to 60 °C.

Evaluation of water content of the hydrogel

The water content of the PIL hydrogel was determined by measuring the weight of water-saturated gel ($W_{water+polymer}$) and dry gel ($W_{polymer}$) with the following equation.

$(W_{water+polymer} - W_{polymer})/W_{polymer}$.

The $W_{polymer}$ was measured after the hydrogel was freeze-dried for at least 48 hours to completely remove water.

4.3 Effect of preparation method on phase transition behavior of hydrogels

4.3.1 Crosslinking method

There are such three main methods to prepare polymer networks as (i) a method for forming a chemical crosslinking through covalent bonding (chemical gel), (ii) a method for physically forming a crosslink by electrostatic interaction or physical entanglement of a polymer (physical gel), and (iii) a method by combining above mentioned ones. PIL gels prepared using these three methods were examined to study the temperature response, phase transition temperature, and appearance of the hydrogels.

(i) Gels formed by chemical crosslinking

First, I sought a suitable synthetic procedure to obtain PIL gels by varying conditions such as species of the crosslinkers. I firstly selected dimethacrylate-type crosslinker 1,14-tetradecanediol dimethacrylate ($C_{14}M$). When PIL gels were prepared by mixing IL monomers; $C_{14}M$ and AIBN, the resulting gels became so rigid that we could not handle them. Instead, IL monomers were mixed with pure water to reach the water content of 20 wt% and the resulting solution was degassed with ultra-sonication. Both 1 mol% AIBN and 3 mol% $C_{14}M$ were then added to the reaction mixture. Polymerization was carried out in glass vials at 80 ^{o}C for 1h under a nitrogen atmosphere. Self-standing gels were obtained and were easily detached from the vials (**Figure 4-1 left**). When a larger amount of crosslinking agent was added, the flexibility of the gel decreased and accordingly the gel was easily broken into pieces after taking from vial. (**Figure 4-1 right**). When the gels were heated, the entire gels gradually became white. This phenomenon was also observed when the PNIPAM hydrogel was heated above the phase transition temperature. Therefore, the PIL gel prepared by chemical crosslinking were found to maintains thermal responsiveness. On further heating, there was leaching out of the water with volumetric shrinkage. After holding them at 60 ^{o}C, the gels were broken down into small fragments by a large volume change. Improving the mechanical strength of the gel will prevent fragmentation, while adding too much crosslinking agent will cause the gel to become

Figure 4-1 PIL gels prepared with 1 mol % of $C_{14}M$ (right) and 3 mol % of $C_{14}M$ (left).

too rigid, leading to fragmentation and making it difficult to undergo a volume phase transition. In order to prepare PIL hydrogel with excellent temperature response and mechanical strength, it is important to optimize the type and amount of crosslinking agent. These considerations will be discussed later.

(ii) Gels formed by electrostatic interaction

It is known that physical gels show a sol-gel transition due to temperature change because the bonding of crosslinking points is generally reversible. Therefore, the physically crosslinked PIL gel may show a flexible volume change when the water is released. Meanwhile, conventional polyion complexes, which are complexes of polyanions and polycations, are widely used mainly as membranes for batteries, humidity sensors, antistatic coating, hemodialysis, and artificial lung. The polyion complex is generally a water-insoluble solid because it has many interaction points between opposite charges on the polymer chains. In fact, when [P_{444}VB][MC3S] was polymerized in Chapter 2, a water-insoluble polymer was obtained. Therefore, it is necessary to adjust the amount of interaction between polyanions and polycations in order to prepare flexible hydrogels.

For the above-mentioned purpose, poly([P_{444}VB][MC3S]), [P_{4446}][MC3S], and AIBN were mixed. The AIBN was used as a radical initiator at 1 mol% of IL monomer. Water was added to the mixture (30 wt%) and heated at 80 °C for 1 h to polymerize. A 5 mL glass vial was used for preparation. **Figure 4-2** shows photographs of the resulting gel where poly([P_{444}VB][MC3S]) was [P_{4446}][MC3S] in a ratio of the repeating units of 1: 10. The saturated swelling PIL gel was too soft to stand on. When the gel was heated, a large amount of water was released while the gel became turbid, suggesting that the thermoresponsiveness was maintained. However, when it was held at high temperatures the gel gradually failed to endure volume changes and fractured. The gel did not recombine even when it was soaked in water again. The volume change induced by the phase transition of the physically crosslinked gel was larger than that of the chemically crosslinked gel. This suggested that the

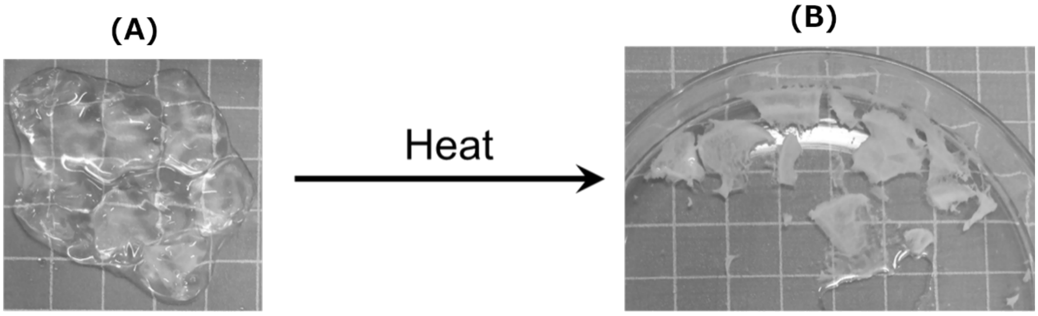


Figure 4-2 Visual appearance of PIL gel before (A) and after (B) heating.

flexibility of the physically-crosslinked gel facilitates the leaching out of water caused by the phase transition of the PIL. It was also found that the reformation of physical crosslinks after swelling was not carried out. In order to improve the strength of the gel by increasing the crosslinking point, the molar ratio of poly([P_{444}VB][MC3S]) to [P_{4446}][MC3S] was raised and mixed, however, the solution before polymerization did not become uniform and the gel could not be produced.

(iii) Gels formed using both covalent bonding and electrostatic interaction

The gel formed the crosslinking through electrostatic interaction was not strong enough, whereas the amount of water in the gel was very large. Therefore, it was attempted to form gels with a crosslinking points by both chemical and physical interaction. I have prepared gels containing linear polycations, network polyanions and monocations by the radical polymerization of poly([P_{444}VB][MC3S]), [P_{4446}][MC3S], and crosslinker.

First, poly([P_{444}VB][MC3S]) and [P_{4446}][MC3S] were mixed in a molar ratio of 1 : 10, and PEGDM was added (0.05 mol% to IL monomer). The resulting gel remained rigid after swelling with water. The gel became turbid by heating, suggesting that the thermoresponsivity was maintained. The gel continued to shrink when heated, causing cracks on the gel surface. Hence, the gel was prepared by changing the ratio of poly([P_{444}VB][MC3S]) to [P_{4446}][MC3S] as well as the amount of crosslinking agent. The optimum crosslinking ratio was proposal by the results. **Figure 4-3** shows photographs of the resulting hydrogels. As a result, the gel prepared by mixing poly([P_{444}VB][MC3S]) and [P_{4446}][MC3S] at a molar ratio of 1: 50 and adding 0.10 mol% of a chemical crosslinking agent to an ionic liquid monomer became a self-standing gel and had appropriate elasticity. When the gel was

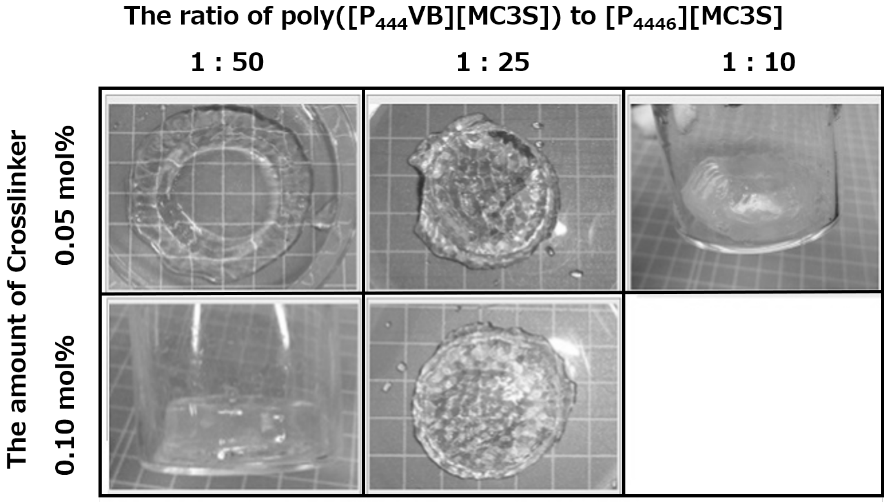


Figure 4-3 visual appearances of chemically and physically crosslinked PIL gels.

heated up to 60 ^{o}C, an LCST type phase transition occurred and the gel became turbid. However, water was not released even when the gel was heated further up to 70 ^{o}C. Therefore, a gel having both mechanical strength and showing reversible water absorption could not be obtained by this method.

4.3.2 Effect of crosslinker and radical initiator

It can be expected that not only the properties of the monomer used here but also the properties of the reagent to be added such as the polymerization initiator or crosslinking agent strongly affect the properties of the obtained hydrogel. Therefore, the effects of the crosslinking agent and the polymerization initiator on the physical properties of the gel were evaluated. the PIL gels were prepared using the method for chemically forming the crosslinked structure which showed relatively good properties in the previous section. **Figure 4-4** shows the structures of the crosslinking agent and the polymerization initiator used in this section. Five kinds of dimethacrylate crosslinking agents with different spacer lengths and repeating structures of spacers were used. Also time different polymerization initiators were used.

The effect of crosslinker species on the stability of the PIL hydrogels has first been investigated, where AIBN has been used as an initiator (**method 1 to 5**). Since AIBN is often used to prepare PNIPAM gels, it was also used in this study. AIBN is an insoluble polymerization initiator in water, but was soluble in aqueous IL monomer solutions. Both EGDM and TEGDM with relatively short spacers made the hydrogel fragile and stiff, resulting in complete fragmentation when soaking them in pure water. I then did not further investigate these hydrogels. The PIL hydrogels with C$_6$M and C$_{14}$M were stable and did not undergo fragmentation after soaking in pure water. However, once the PIL hydrogels were heated to induce

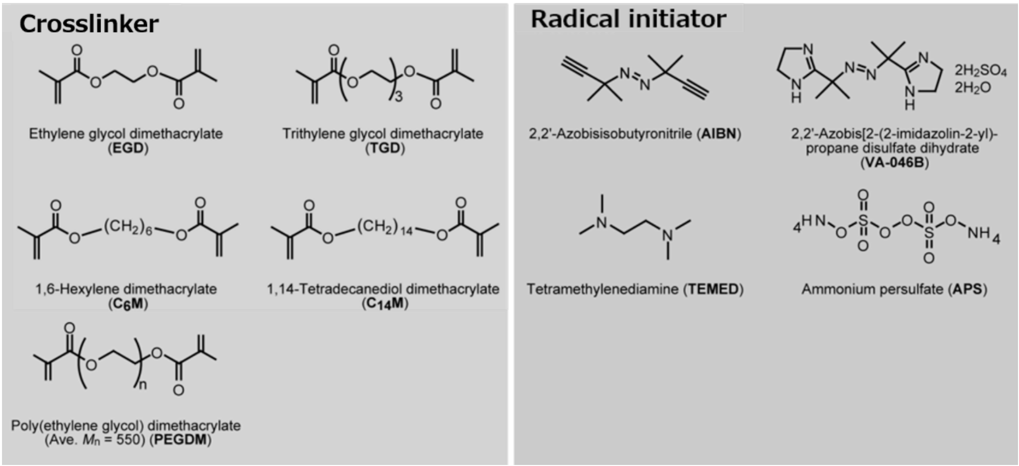


Figure 4-4 Chemical structures of crosslinker and radical initiator used here.

Table 4-1 List of synthetic methods and water absorption/desorption reversibility of thermoresponsive PIL hydrogels

Method #	Crosslinker		Initiator		Water pumping reversibility
	Name	Concentration (mol% to IL)	Name	Concentration (mol% to IL)	
1	EGDM	1.0 ~ 3.0	AIBN	0.1	–[a]
2	TEGDM	1.0 ~ 3.0	AIBN	0.1	–[a]
3	C_6M	1.0 ~ 3.0	AIBN	0.1	–[a]
4	$C_{14}M$	1.0 ~ 3.0	AIBN	0.1	–[a]
5	PEGDM	0.1	AIBN	0.1	+[b]
6	PEGDM	0.1	APS/TEMED	0.3/0.4	–[a]
7	PEGDM	0.1	VA-046B	0.1	+[b]

[a]These gels were fragile and did not show reversible absorption/desorption of water; [b]These gels reversibly absorbed/desorbed water.

the LCST-type phase transition, they were fragmented while desorbing water. Although the spacer lengths of C_6M and $C_{14}M$ were about doubly different, the effect of the spacer length in this range on the strength of the swollen gels seemed to be small. Then, I changed the crosslinker to PEGDM having sufficiently long ethylene glycol spacer. The spacer structure was changed from alkyl chains to ethylene glycol chains to improve the water affinity. However, the resulting PIL hydrogels with 1 mol % of PEGDM were also rigid and fragile. On the other hand, it is confirmed that relatively stable PIL hydrogels showing reversible water pumping behavior can be prepared when 0.1 mol% of PEGDM is used as a crosslinker (**method 5**). The PIL hydrogel reversibly absorbs/desorbs water upon temperature change, but after fifth absorption/desorption cycles, the hydrogel was fragmented. Since the polyethylene glycol chain is more hydrophilic than the alkyl chain, the gel can retain water during both swelling and shrinking. This is considered to reduces volume change of the gel and thus suppresses fragmentation. Note that when concentration of PEDMG was smaller than 0.1 mol%, the gel became too soft to self-standing. From the above results, it was found that the hydrophilicity and the amount of cross linker were important to control the mechanical strength of hydrogel, while the effect of their spacer length were little.

As $[P_{4446}][MC3S]$ shows LCST behavior with water, the monomer is phase-separated from water during polymerization when AIBN is used as an initiator due to active temperature (~ 60 °C). The polymerization, therefore, proceeds under heterogeneous conditions. Such heterogeneous gelation reaction was also exploited for Gemini-dicationic PIL systems by Yuan's group.[10] Although there is still no data on the mechanical stability of the gels upon temperature change, the heterogeneity of the reaction mixtures should be ascribed as one of the issues to make the hydrogels fragile, upon considering my current investigations.

AIBN was then replaced by two different types of initiators (i.e., APS/TEMED and VA-046B) for radical polymerization at lower-temperature so that the gelation was carried out under a homogeneous state. The PEGDM-type crosslinker (0.1 mol %) was used in all the following systems because it provided relatively stable PIL hydrogels based on the aforementioned results. The gelation was undertaken at room temperature for APS/TEMED system and at 50 °C for the VA-046B system, during which the reaction mixture remained a homogeneous state in both cases. When APS/TEMED was applied for the gelation (**method 6**), the resulting mixture was still liquid state and did not undergo gelation regardless of the initiator concentration tested. Against this, a mechanically stable, transparent, and thermoresponsive PIL hydrogel was obtained by employing VA-046B in the system (**method 7**). As **Figure 4-5** shows, the gel had excellent mechanical strength and showed little change in appearance even after 10 cycles of swelling and shrinking. Therefore, it was found that the reduction of the heterogeneity of the structure of the gel by polymerization in a homogenous state improves the resistance of the volume change of the gel.

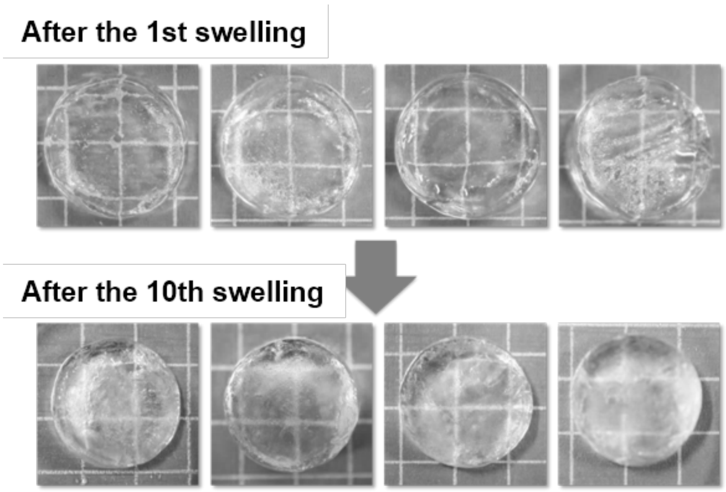


Figure 4-5 Visual appearances of PIL gels prepared by method 7 before and after 10 times swelling/shrinking process.

4.4 Control of phase transition temperature by copolymerization

The control of T_c value is very important when considering the wide application of PIL gel as a temperature-responsive material. In the previous chapter, the author showed that copolymerization of ionic liquid monomers with different hydrophobicity was effective in controlling the T_c value of PILs. It was also cleared that increasing the ratio of hydrophobic fraction in PILs tended to decrease the T_c value linearly. Therefore, in this section, the ionic liquid monomers, [P_{4446}][MC3S] and [P_{4448}][MC3S] having different hydrophobic cations, were copolymerized and evaluated the effect of the obtained PIL gel on T_c.

The two types of IL monomer, [P_{4446}][MC3S] and [P_{4448}][MC3S], were mixed in a molar ratio of 10: 0, 8: 2, and 6: 4, and polymerize them according to the **method 4** or the **method 7** (see in **Table 4-1**) to prepare PIL gels. The obtained gel was soaked in pure water for 24 hours or more to saturate and then, the T_c value was determined by DSC measurement. The phase transition temperatures of each prepared PIL gel are shown in **Table 4-2**. In the gel prepared by the method 4, the T_c value changed almost linearly with the fraction of [P_{4448}][MC3S], similar to the linear polymer system as described in the previous chapter. On the other hand, in the PIL gel prepared by method 7, endothermic peaks attributable to phase transitions could not be identified during DSC measurement. It might be because the saturated water content of the gel was very high and the amount of polymer in the gel was small when it was separated into the sample pan. However, it was confirmed that the phase transition occurred because the whole gel was turbid when heated. When the gels using only [P_{4446}][MC3S] as monomers were compared, the T_c value differed by approximately 20 °C. The gels prepared by the **method 7** showed higher T_c value than that by the **method 4**. This suggests that the polymerization initiator and crosslinker also affect the T_c value of PIL gels and the effect is not small. On the other hand, when the molar ratio of [P_{4446}][MC3S] to [P_{4448}][MC3S] was 6:4, the phase

Table 4-2 The T_c values of PILs gel

[P_{4446}][MC3S][P_{4448}][MC3S]	**T_c / °C**
AIBN, C_{14}M (method 4)	
10 : 0	47
8 : 2	44
6 : 4	37
VA-046B, PEGDM (method 7)	
10 : 0	c.a. 65 *1
8 : 2	c.a. 55 *1
6 : 4	c.a. 35 *1

*1 visually determined value

transition temperatures were almost the same in spite that the measurement methods were different. This result indicates that the structure of polymerization initiator and crosslinker also contribute to the T_c value.

The change of saturated water content in PIL gels prepared with VA-046B and PEGDM was found to be proportional to the $[P_{4446]}[MC3S]$ fraction (**Table 4-3**). As the hydrophilicity of the polymer chains increases, more amount of water retained. The amount of water in poly($[P_{4446}][MC3S]$) gel which calculated from water content was 99.6 wt%. In the previous chapter, I found that the Tc value of PIL/water mixture can be controlled by PIL concentration. For example, poly($[P_{4446}][MC3S]$)/water mixture showed the T_c value at 54 °C when a polymer concentration was 2.5 wt%, and the T_c value of poly($[P_{4446}][MC3S]$)/water mixture were inversely proportional to the logarithm of the concentration of poly($[P_{4446}][MC3S]$). According to the calibration curve calculated by the results, the T_c value when a polymer concentration was 0.4 wt% should be 70 °C. Considering this value, the T_c value of 65 °C for the poly($[P_{4446}][MC3S]$) gel is low. It was suggested that the T_c value of PIL is affected by polymer structure. This property is particularly unique for PILs showing the LCST-type phase transitions with water. In the case of PNIPAM, polymer structure has little effect on the phase transition temperature. The T_c value of hydrogel and linear polymer in water are almost the same, around 32 °C. In addition, the T_c value of the poly($[P_{4446}][MC3S]_{0.6}$-*co*-$[P_{4448}][MC3S]_{0.4}$) gel was much lower than that of poly($[P_{4446}][MC3S]$) gel, although the amount of water in the gel, 99.5 wt%, was almost the same as poly($[P_{4446}][MC3S]$) gel. It is found that the T_c value of PIL gel is much more sensitive to water content than that of PIL/water mixture.

Table 4-3 Water content of PIL gel which is a fully swollen state.

$[P_{4446}][MC3S][P_{4448}][MC3S]$	Water content (g/g)
VA-046B, PEGDM	
10 : 0	275
8 : 2	221
6 : 4	191

4.5 Semi Reversible Changes of Water Content in PIL gels

In PIL gels, polymer chains are entangled by a crosslinked structure. In addition, the internal structure of PIL gels is more complex than that of IL monomer/water mixtures and that of PIL/water mixtures. Therefore, the volume phase transition caused by the phase transition (phase separation in the solution system) is expected to be slow. Moreover, since the polymerization initiator and crosslinker affected the phase transition temperature, they may also affect the behavior of the volume phase transition. Therefore, I evaluated the phase transition behavior by measuring the temperature dependence of the water content of PIL gels. The prepared PIL gels were kept at 6.5 °C for more than 72 hours to reach a saturated water swollen state, and then weighed after 30 minutes at every temperature. Five samples were used for each evaluation, and the average value was used for discussion.

Figure 4-6 shows the water content of the PIL gels ([P_{4446}][MC3S]:[P_{4448}][MC3S] = 6:4) prepared by **method 4**. Compared with the PIL/water mixture system reported in the previous chapter, the water content of the PIL gel changed slowly over a wide temperature range. This slow and wide response is thought to be due to the inhomogeneous network structure of the gel. It was suggested in the previous chapter that the intermolecular distance is important because the polymer concentration affects the phase transition temperature. When there is a variation in the intermolecular distance of the

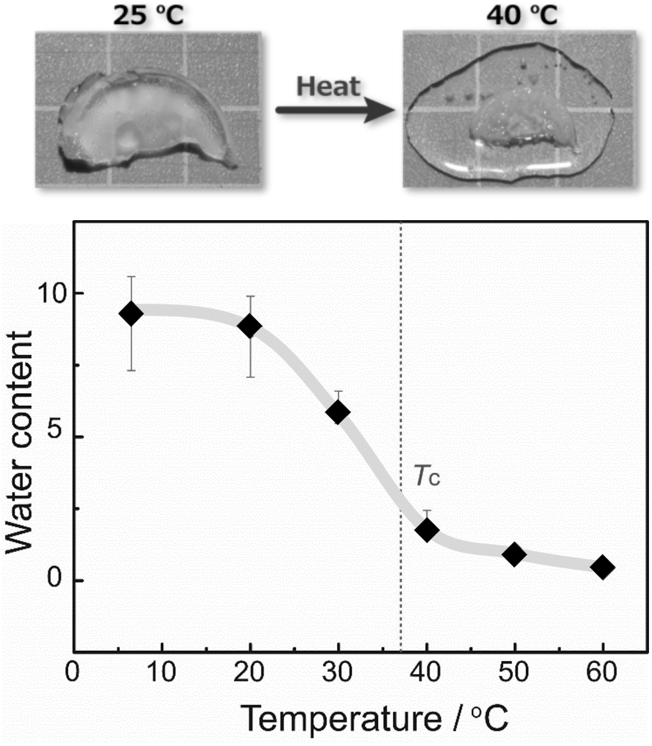


Figure 4-6 Change in water content of poly([P_{4446}][MC3S]$_{0.6}$-*co*-[P_{4448}][MC3S]$_{0.4}$) gel prepared by method 4 upon heating for 30 min.

gel, the phase transition will occur over a wide temperature range. The change in water content was not constant between 6.5 °C to 60 °C, and the largest change in water content was observed around the T_c value as seen in the figure. The gel released about 9 times its own weight of water when the temperature was raised from 6.5 °C to 60 °C. The water content of the gel at 60 °C was less than 1wt%. Thus, the water content of the gel decreased by 90% by heating. The large volume change associated with the phase transition was thought to be the cause of the cracks in the gel.

The PIL gel prepared by **method 7** ([P_{4446}][MC3S]:[P_{4448}][MC3S] = 5 : 5) had higher water content at all temperatures than the gel prepared by **method 4**. The gel gradually released water over a wide temperature range (**Figure 4-7**). When the hydrogel was heated from 6.5 °C to 60 °C, the amount of released water was about 110 times its own weight, but it still held 170 times its own weight in water even at 60 °C. This means that the volume change of the gel prepared by **method 7** was small as compared to the gel prepared by **method 4**. This small volume change made PIL gel able to adsorb and desorb water repeatedly.

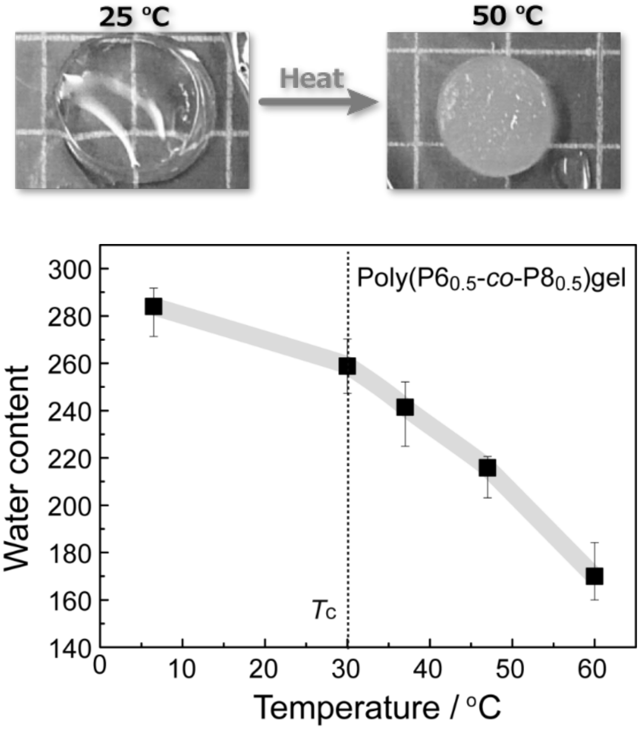


Figure 4-7 Change in water content of poly([P_{4446}][MC3S]$_{0.5}$-*co*-[P_{4448}][MC3S]$_{0.5}$) on heating for 30 min.

Since the PIL gels prepared by method 7 did not show any crack during shrinkage, the water absorption behavior of the PIL gels was subsequently evaluated. The change in water content of the gel with time is shown in **Figure 4-8**. The gel began to absorb water slowly when it was immersed in water at room temperature, and reached a saturated state after about 150 minutes. The swelling seems to be caused by the natural diffusion of water in polymer network chain, whereas the shrinkage was induced by the repulsive force generated by phase transition.

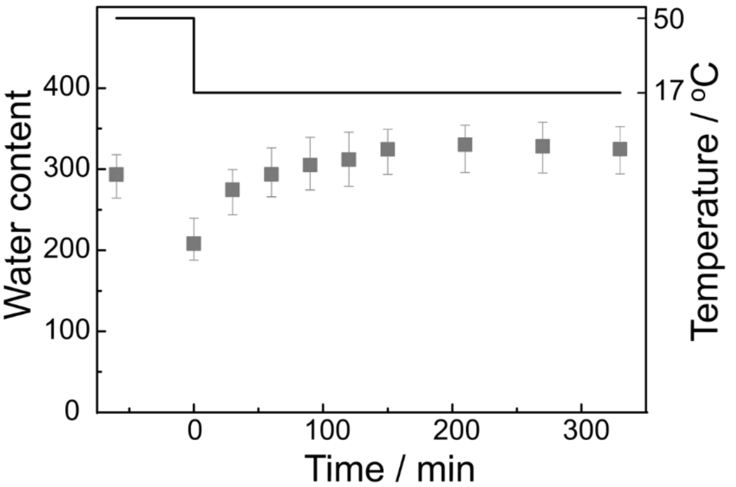


Figure 4-8 Swelling profile of poly([P_{4446}][MC3S]$_{0.6}$-*co*-[P_{4448}][MC3S]$_{0.4}$) at room temperature.

4.6 Repeated Changes of Water Content in the PIL gel

Finally, the continuous change in water content of the PIL gel in response to temperature was evaluated. **Figure 4-9** summarizes the change of the water content of poly([P_{4446}][MC3S]$_{0.5}$-*co*-[P_{4448}][MC3S]$_{0.5}$) gels prepared by method 7 by temperature cycling. The water content of the water-saturated hydrogel was 280. The gel oozed a certain amount of water at 50 °C. The average water content was calculated to be 242 after desorption at 50 °C. The gel oozed more water by further heating, but the reversibility of water pumping was evaluated at a temperature between 5 and 50 °C. The change in water content during swelling and shrinking was almost the same during the 10 cycles. In addition, this hydrogel was found to be stable without generating any crack even after absorption/desorption cycles for more than 10 times. It can be claimed, therefore, that suitably prepared PIL hydrogels in this study can undergo reversible water pumping without degradation. It should be noted that the water content in the hydrogel was sometimes fluctuating as shown in **Figure 4-9**. Before measuring the water content of the hydrogel, the excess amount of the oozed water on the gel was wiped off. This process might cause experimental errors.

As described above, I succeeded in fabricating PIL gels that reversibly adsorbs and desorbs water. The water pumping mentioned here would provide a new platform to fabricate a wide variety of hydrogels based on these PILs.

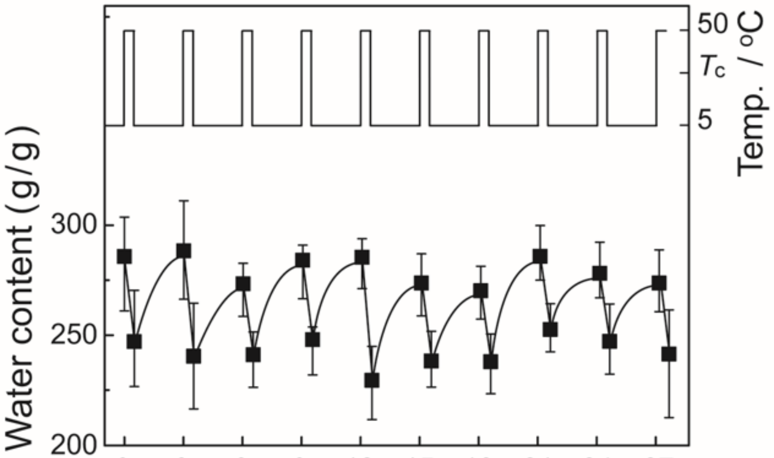


Figure 4-9 Swelling/shrinking behavior of poly([P_{4446}][MC3S]$_{0.5}$-*co*-[P_{4448}][MC3S]$_{0.5}$) gel upon temperature change between 5 °C and 50 °C.

4.7 Conclusion

This chapter mentions the formation method of crosslinked gel structure and the control of physical properties targeting long life PIL hydrogels. Then, the excellent temperature response of the obtained PIL gel was evaluated. Through careful selection of both radical initiators and crosslinkers, as well as the reaction procedures, some mechanically stable thermoresponsive PIL hydrogels that reversibly absorb/desorb water molecules by moderate temperature change have been prepared. It was confirmed that the gels that continuously absorb/desorbed water have smaller volume changes during saturated swelling and shrinkage than the gels easily fragmented after first swelling. Such materials capable of inducing changes of the chemical environment with small temperature change. They have potential for various applications.

4.8 Reference

1) F. Eeckman, a J. Moës, and K. Amighi,. "Poly(*N*-isopropylacrylamide) copolymers for constant temperature controlled drug delivery.",. *International journal of pharmaceutics*, **2004**, 273, 109–119.

2) Z. Tang, Y. Akiyama, and T. Okano,. "Temperature-Responsive Polymer Modified Surface for Cell Sheet Engineering",. *Polymers*, **2012**, 4, 1478–1498.

3) Z. Tang, Y. Akiyama, and T. Okano,. "Recent development of temperature-responsive cell culture surface using poly(*N*-isopropylacrylamide)",. *Journal of Polymer Science Part B: Polymer Physics*, **2014**, 52, 917–926.

4) M. E. Harmon, M. Tang, and C. W. Frank,. "A microfluidic actuator based on thermoresponsive hydrogels",. *Polymer*, **2003**, 44, 4547–4556.

5) L. C. Dong, and A. S. Hoffman,. "Thermally reversible hydrogels: III. Immobilization of enzymes for feedback reaction control",. *Journal of Controlled Release*, **1986**, 4, 223–227.

6) C. Sakamoto, Y. Okada, H. Kanazawa, E. Ayano, T. Nishimura, M. Ando, A. Kikuchi, and T. Okano,. "Temperature-and pH-responsive aminopropyl-silica ion-exchange columns grafted with copolymers of *N*-isopropylacrylamide."., *Journal of Chromatography A,* **2004**, 1030, 247–253.

7) J.-F. Lutz, O. Akdemir, and A. Hoth,. "Point by point comparison of two thermosensitive polymers exhibiting a similar LCST: is the age of poly(NIPAM) over?",. *Journal of the American Chemical Society*, **2006**, 128, 13046–7.

8) L. Ayres, M. R. J. Vos, P. J. H. M. Adams, I. O. Shklyarevskiy, and J. C. M. van Hest,. "Elastin-Based Side-Chain Polymers Synthesized by ATRP",. *Macromolecules*, **2003**, 36, 5967–5973.

9) R. Hoogenboom, H. M. L. Thijs, M. J. H. C. Jochems, B. M. van Lankvelt, M. W. M. Fijten, and U. S. Schubert,. "Tuning the LCST of poly(2-oxazoline)s by varying composition and molecular

weight: alternatives to poly(*N*-isopropylacrylamide)?",. *Chemical communications*, **2008**, 5758–5760.

10) Y. Y. Men, H. Schlaad, A. Voelkel, and J. Yuan,. "Thermoresponsive polymerized gemini dicationic ionic liquid",. *Polymer Chemistry*, **2014**, 5, 3719-3724.

Chapter 5

General conclusion

This thesis mentioned the design of polymerized ionic liquids (PILs) to show temperature-sensitive lower critical solution temperature (LCST)-type phase transition in water and the chemical control of their transition Based on the obtained results, the preparation method of the thermoresponsive hydrogel was summarized. Through the physicochemical evaluation of its temperature-responsive behavior, the physicochemical propertied of PIL hydrogel were also discussed.

Chapter 1 gave a general introduction of conventional non-ionic polymer showing LCST-type-phase transition with water, themoresponsive IL, and PIL. Then the objective of this thesis was also outlined.

In Chapter 2, the phase behavior of the ILs monomer and the resulting PILs was evaluated. In addition, the aggregation state of PIL in water was analyzed by various methods. Some IL monomers showed LCST-type phase transition with water and the thermoresponsivity was confirmed to be maintained after polymerization. Consequently, the polymer was found to aggregate in the water even in the under the phase transition temperature.

In Chapter 3, the factors to control the phase transition temperature of PIL/water mixtures were discussed. It was confirmed that the phase transition temperature of the PIL/water mixture can be controlled by some methods such as adjusting the PIL concentration, adding an inorganic salt, and copolymerizing monomers having different hydrophilicity. In addition, it was clarified that the phase behavior with water could be controlled by blending PILs with different hydrophilicity. It was clearly confirmed that it is possible to give a temperature response to PILs that are not showing the LCST-type phase transition by adjusting the hydrophilic and hydrophobic properties within an appropriate range by blending.

In Chapter 4, adequate cross-linking methods and additives were explored for the purpose of obtaining a thermoresponsive PIL hydrogel capable of continuous use. The gel prepared by chemical cross-linking was the easiest to handle. The phase behavior of the PIL hydrogel with temperature change was evaluated. This gel retained water 280 times its weight below the phase transition temperature and discharged 14% of the water upon heating above the phase transition temperature. In addition, water was continuously adsorbed and desorbed for 10 times or more by the temperature change.

All the results discussed in this thesis lead to a conclusion that PILs showing the LCST-type phase transition with water provides new class of thermoresponsive polymer as thermoresponsive polymers which easily control their phase transition temperature with temperature sensitivity. These materials surely expand both scientific interest and advanced applications. Particularly, thermally phase transition of water/charged species is extremely useful for different applications and different temperature range from non-ionic systems. Dynamic change of hydration state of the ionic environment may induce additional switching of many functions *via* change of electrostatic interaction force derived from the change in hydration state. I believes that diverse functional applications can be designed with this PIL showing LCST-type phase transition with water.

List of Publications

1. Y. Kohno, Y. Deguchi, and H. Ohno,
"Ionic liquid-derived charged polymers to show highly thermoresponsive LCST-type transition with water at desired temperatures"
Chemical Communication 2012, **48**, 11883-11885. [Chapter 2, 3]

2. Y. Kohno, Y. Deguchi, N. Inoue, and H. Ohno,
"Temperature-driven and reversible assembly of homopolyelectrolytes derived from suitably designed ionic liquids in water"
Australian Journal Chemistry 2013, **66**, 1393–1398. [Chapter 2]

3. Y. Deguchi, Y. Kohno, and H. Ohno,
"Design of ionic liquid-derived polyelectrolyte gels toward reversible water absorption/desorption system driven by small temperature change"
Australian Journal of Chemistry 2014, **67**, 1666-1670. [Chapter 3, 4]

4. Y. Deguchi, Y. Kohno, and H. Ohno,
"A fine tuning of LCST-type phase transition of poly(ionic liquid)s in water."
Chemistry letters 2015, **44**, 238-240. [Chapter 3]

5. Y. Deguchi, Y. Kohno, and H. Ohno,
"Reversible water uptake/release by thermoresponsive polyelectrolyte hydrogels derived from ionic liquids"
Chemical Communication 2015, **51**, 9287-9290. [Chapter 4]

6. Y. Deguchi, N. Nakamura, and H. Ohno,
"Thermoresponsive ionic liquid/water mixtures for separation and purification technologies"
Separation and Purification Technology 2020, **251**, 117286. [Chapter 3, 4]

The author also contributed to the following papers.

7. Y. Deguchi, J S. Moreno, S. Panero, B. Scrosati, H. Ohno, and G.B. Appetecchi,
"An advanced ionic liquid-lithium salt electrolyte mixture based on the bis(fluoromethane-sulfonyl) imide anion"
Electrochemistry communications 2014, **43**, 5-8.

8. J. S. Moreno, Y. Deguchi, S. Panero, B. Scrosati, H. Ohno, E. Simonetti, and G. B. Appetecchi,
"*N*-Alkyl-*N*-ethylpyrrolidinium cation-based ionic liquid electrolytes for safer lithium battery systems"
Electrochimica Acta 2016, **191**, 624-630.

Acknowledgements

The author summarized this thesis at Tokyo University of Agriculture and Technology during 2012-2015 and 2019-2022 under the direction of Professor Dr. Hiroyuki Ohno and Professor Dr. Nobufumi Nakamura. The thesis is concentrated on the design of polymerized ionic liquids showing LCST-type phase transition with water.

First of all, I would like to express my cordial gratitude to Professor Dr. Hiroyuki Ohno for his encouragement, pointed advice, and continuous support. His powerful mind to pursue various novel and exciting science made me more passionate about my research.

I wish to thank Professor Dr. Nobufumi Nakamura for his kind and continuous support and helpful discussions.

I am very grateful to Dr. Yuki Kohno and Dr. Shohei Saita for their support and encouragement. I have realized the importance and worth of my research through discussion with them.

I gratefully acknowledge the work of past and present members of our laboratory for their assistance.

Finally, I express my deep gratitude to my parents for their support and my husband for powerful encouragement.

Yuki Deguchi-Nishida