

Comparison of Thermosensitive Properties of Poly(amidoamine) Dendrimers with Peripheral *N*-Isopropylamide Groups and Linear Polymers with the Same Groups

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Thermosensitive polymers, which exhibit changes in water solubility at a specific temperature, are highly attractive for the production of functional or intelligent materials.^[1] Although a number of polymers are known to exhibit thermosensitive properties, the most thoroughly studied is poly(*N*-isopropylacrylamide) (PNIPAAm).^[2,3] This polymer is highly soluble in water at low temperature. It becomes water-insoluble at temperatures greater than 31–32 °C: the polymer chains become dehydrated and form aggregates.^[3] This temperature is designated as the lower critical solution temperature (LCST).

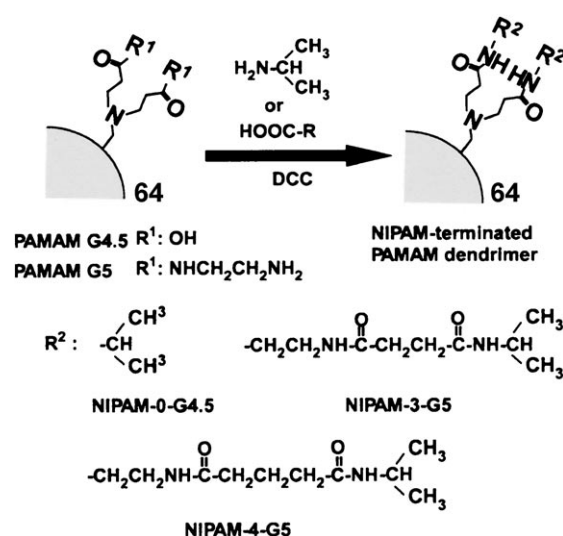
The thermosensitive polymers known so far generally possess a linear structure. However, recently we developed a new type of thermosensitive polymer with a globular structure. These polymers were obtained by modification of the chain terminals of poly(amidoamine) (PAMAM) or poly(propyleneimine) dendrimers with isobutyramide groups, which are common structural units in the thermosensitive polymer, poly(*N*-vinylisobutyramide).^[4] These modified dendrimers differ markedly from conventional thermosensitive polymers with a linear structure in terms of their molecular shape and location of alkyl amide groups, which play a crucial role in determining their thermosensitive properties.

A number of studies comparing dendritic, hyperbranched, and linear polymers have revealed that molecular shapes and topologies strongly influence their chemical and physical properties.^[5] In addition, the unique shape and characteristics of thermosensitive dendrimers may lead to the production of novel functional materials that are not obtainable with linear thermosensitive polymers. Therefore, we seek to elucidate how such differences affect their thermosensitive properties, which may lead to the generation of new applications for thermosensitive polymers.

In this study, we introduce the *N*-isopropylamide (NIPAM) group, which is a common structural unit with PNIPAAm, at every chain terminal of the PAMAM dendrimer through various spacers. The thermosensitive properties of the NIPAM-terminated dendrimers were compared with

those of NIPAM-bearing polymers with a linear structure, such as PNIPAAm and poly(*N*-isopropylacrylamide-*co*-acrylamide) [poly(NIPAAm-*co*-AAm)].

The PAMAM dendrimers with NIPAM, *N*-isopropylsuccinamide, and 4-isopropylcarbamoyl butyramide groups at every chain end, which are designated as NIPAM-0-G4.5, NIPAM-3-G5, and NIPAM-4-G5, respectively, were synthesized by reacting a corresponding amine or carboxylic acid with the carboxyl-terminated PAMAM G4.5 or amine-terminated PAMAM G5 dendrimer, according to a previously reported method (Scheme 1).^[4,6] Analysis by ¹H and ¹³C NMR spectroscopy demonstrated that essentially every chain terminal of these dendrimers was connected to the corresponding terminal groups of the surface-modified dendrimers (see the Supporting Information).



Scheme 1. Synthetic route for NIPAM-terminated dendrimers. DCC = 1,3-dicyclohexylcarbodiimide. The number 64 next to the gray surface refers to the number of terminal moieties.

Figure 1 depicts the temperature dependence of the light transmittance of solutions of various NIPAM-terminated dendrimers and linear polymers with NIPAM groups in a phosphate solution (10 mM, pH 9.0) at 500 nm. PNIPAAm exhibited a sharp decrease in transmittance at 32 °C, which indicates that this polymer underwent conformational transition from a hydrated coil to a dehydrated globule at this temperature. Poly(NIPAAm-*co*-AAm) also showed a sharp decrease in transmittance at 40 °C.

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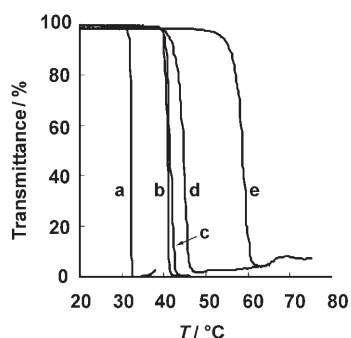


Figure 1. Effect of temperature on transmittance of a) PNIPAAm, b) poly(NIPAAm-co-AAm), c) NIPAM-0-G4.5, d) NIPAM-4-G5, and e) NIPAM-3-G5 dissolved in 10 mM phosphate solution (10 mg mL⁻¹, pH 9.0).

Similarly, solutions of NIPAM-terminated dendrimers showed a sharp decrease in turbidity at a certain temperature: the LCSTs for the solutions of NIPAM-0-G4.5, NIPAM-3-G5, and NIPAM-4-G5 dendrimers were estimated as 41, 56, and 43 °C, respectively. This result further confirms that the introduction of a common structural unit to the surface of dendrimers can impart temperature-sensitive properties to them.^[4,7] Although these dendrimers have the same number of NIPAM groups on the periphery, they exhibit different LCSTs. As their molecular structures show (Scheme 1), the length of the spacer moiety between the tertiary amine at the outermost branching point and the terminal NIPAM group increases in the order of NIPAM-0-G4.5 < NIPAM-3-G5 < NIPAM-4-G5 dendrimers. The increase in spacer length might decrease the density of NIPAM groups in the dendrimer periphery. Such a situation could depress the occurrence of phase transition of the dendrimer.^[4] However, the hydrophobicity of the spacer moiety might increase with increasing spacer length, which enhances the occurrence of the phase transition. These opposing effects of the spacer length could cause the deviation of the LCSTs of the dendrimers.

Next, we characterized the transitions of the NIPAM-bearing polymers and dendrimers by differential scanning calorimetry (DSC; Figure 2).^[2,8] PNIPAAm and poly(NIPAAm-co-AAm) showed endothermic peaks centered at 31.4

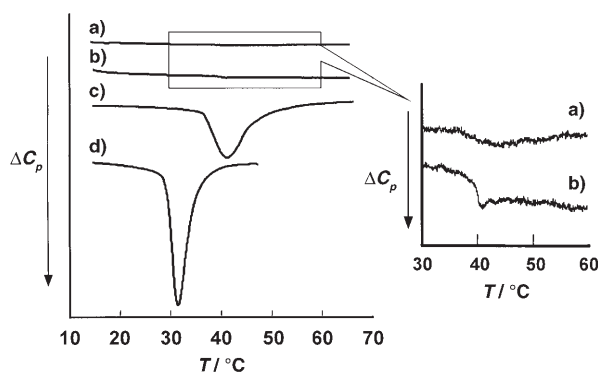


Figure 2. DSC thermograms of a) NIPAM-0-G4.5, b) NIPAM-4-G5, c) poly(NIPAAm-co-AAm), and d) PNIPAAm. The heating rate was 0.5 K min⁻¹. C_p = heat capacity.

and 41.0 °C, respectively, which agree with their LCSTs (Figure 1). Similarly, the NIPAM-terminated dendrimers exhibited an endothermic peak around the LCST. However, a striking difference is apparent between the DSC curves for the NIPAM-terminated dendrimers and the linear polymers: the heat of transition for the former is extremely small.

The cloud points and transition enthalpies (ΔH) for these thermosensitive polymers and dendrimers are summarized in Table 1. The phase separation or precipitation of thermosen-

Table 1: Transition enthalpies of various thermosensitive polymers.

Polymer	Cloud point [°C]	T_{max} [°C] ^[a]	ΔH [J g ⁻¹]	ΔH [kJ mol ⁻¹ of NIPAM unit]
PNIPAAm	32.2	31.4	34.3	3.88
poly(NIPAAm-co-AAm)	40.2	41.0	21.1	2.79
NIPAM-0-G4.5	40.6	43.8	0.3	0.07
NIPAM-4-G5	43.2	40.4	0.2	0.08

[a] Evaluated by DSC.

sitive polymers has been explained from the viewpoint of entropy effects in the polymer solution.^[2,9] At low temperatures, strong hydrogen bonding between hydrophilic amide groups and water exceeds the unfavorable free energy related to the exposure of hydrophobic isopropyl groups to water. With increasing temperature, hydrophobic interaction between isopropyl groups is enhanced, whereas hydrogen bonding is weakened. Therefore, at temperatures higher than the LCST, interaction between hydrophobic groups becomes dominant, thereby resulting in the entropy-driven polymer collapse and concomitant release of structured water around the hydrophobic groups, which requires the absorption of heat.^[2,9]

As shown in Table 1, the transition enthalpy of poly(NIPAAm-co-AAm) is somewhat lower than that of PNIPAAm because hydration of hydrophobic groups decreases with increasing temperature.^[9] However, compared to these linear polymers, the dendrimers exhibit transition enthalpies that are two orders of magnitude lower, even though these polymers and dendrimers include the same hydrophobic groups. Indeed, the weight percent of NIPAM groups in the whole molecule differs among these polymers and dendrimers. Consequently, the transition enthalpy is normalized by the number of NIPAM groups in a molecule, but much smaller values are still apparent for the dendrimers after the normalization compared with those of the linear polymers (Table 1).

The release of the structured water might take place not only from NIPAM groups, but also from the polymer backbone for the linear polymers. However, a larger fraction of hydrophobic carbon atoms exists in the NIPAM groups. These side groups play a crucial role in the thermosensitive properties of the polymers. Therefore, hydration or dehydration of NIPAM groups before and after the transition might be responsible for the large difference between the transition enthalpies of the linear polymers and globular dendrimers.

The dendrimer surface should exhibit a more hydrophobic nature if NIPAM groups in the periphery of the dendrimer are

less hydrated than the NIPAM-bearing linear polymers. Therefore, we examined the hydrophobicity of these NIPAM-bearing polymers below the LCST using 1-pyrene-carboxaldehyde (PyCHO), which changes the wavelength of the emission maximum ($\lambda_{\max}$) depending on the polarity of the solvent.^[10,11]

Figure 3 shows the dependence of $\lambda_{\max}$ on the concentration of NIPAM-terminated dendrimers, PNIPAAm, and poly(NIPAAm-co-AAm) below the LCST. The result for the

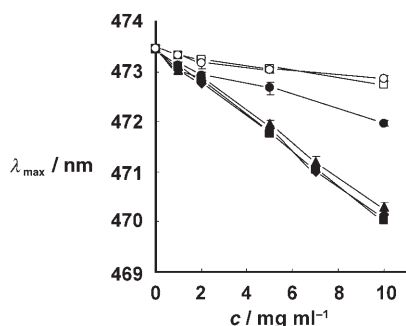


Figure 3. Emission maxima of PyCHO (1 μM) as a function of the concentration of NIPAM-0-G4.5 ($\blacksquare$), NIPAM-3-G5 ($\blacktriangle$), NIPAM-4-G5 ($\blacklozenge$), OH-terminated G5 ($\bullet$), PNIPAAm ($\square$), and poly(NIPAAm-co-AAm) ($\circ$) dissolved in 10 mM phosphate solution (pH 9.0) at 20 °C. $\lambda_{\text{ex}} = 365.5$ nm.

OH-terminated PAMAM G5 dendrimer is also given, as a dendrimer without NIPAM groups in the periphery. The presence of PNIPAAm or poly(NIPAAm-co-AAm) only slightly affected $\lambda_{\max}$ even at a high concentration of 10 mg mL^{-1} , which indicates that these linear polymers do not form domains with a hydrophobic nature. However, in the presence of NIPAM-terminated dendrimers, a blue shift of $\lambda_{\max}$ was observed with increasing concentration of the dendrimers. The OH-terminated PAMAM-G5 dendrimer caused a blue shift of $\lambda_{\max}$ which was much less than that caused by NIPAM-terminated dendrimers. These dendrimers contain the same interior. Therefore, the marked change of $\lambda_{\max}$ in the presence of the NIPAM-terminated dendrimers might be attributable to the peripheral NIPAM groups. Probably, the dense packing of NIPAM groups in the dendrimer periphery enhances dehydration around these groups.

To confirm the low extent of hydration of the dendrimer NIPAM groups, we further investigated the influence of urea on the LCST, because urea is known to modify hydration around hydrophobic solutes in aqueous solutions and to reduce hydrophobic interactions.^[12] The presence of urea did not affect the LCST of the PNIPAAm solution (Figure 4a). Fang et al. reported that the presence of urea at a concentration of 3 M did not change the LCST of PNIPAAm, although swelling of the compact conformation of the polymer chain was caused by urea at temperatures higher than the LCST.^[13] In contrast, a marked difference is apparent for the NIPAM-terminated dendrimer, which exhibited a considerable increase of LCST with increasing urea concentration.

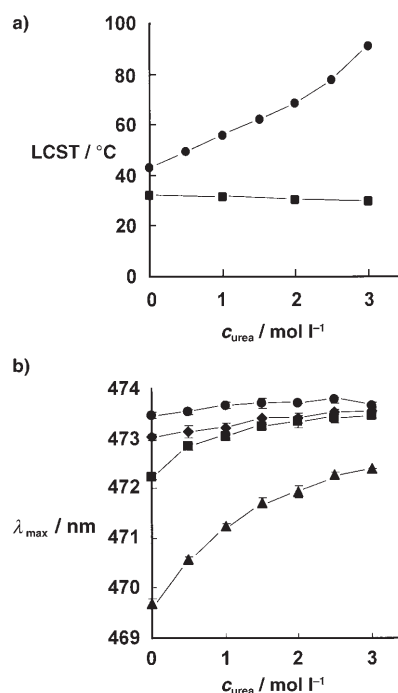


Figure 4. a) Influence of urea on LCST of NIPAM-4-G5 ($\bullet$) and PNIPAAm ($\blacksquare$) dissolved in 10 mM phosphate solution (10 mg mL^{-1} , pH 9.0). b) Influence of urea on emission maxima of PyCHO (1 μM) in 10 mM phosphate solution in the absence ($\bullet$) or presence ($\blacklozenge$) of PNIPAAm, PAMAM-OH G5 ($\blacksquare$), and NIPAM-4-G5 ($\blacktriangle$) at 20 °C.

The effects of urea on the hydrophobicity of the NIPAM-bearing dendrimer and linear polymers were also examined using PyCHO (Figure 4b). Although the effects on PNIPAAm and OH-terminated PAMAM G5 dendrimer were negligible, the NIPAM-terminated dendrimer displayed a considerable increase in $\lambda_{\max}$ with increasing urea concentration. Probably, urea molecules disturb the hydrophobic interaction of NIPAM groups and reduce their dehydration in the surface of the dendrimer.

The different hydration states around NIPAM groups that arise from the structural features of these linear and globular polymers are illustrated schematically in Figure 5. For NIPAM-bearing linear polymers, their backbone has a large conformational freedom, which enables efficient hydration of NIPAM groups below the LCST and efficient association of NIPAM groups above the LCST. For that reason, a large

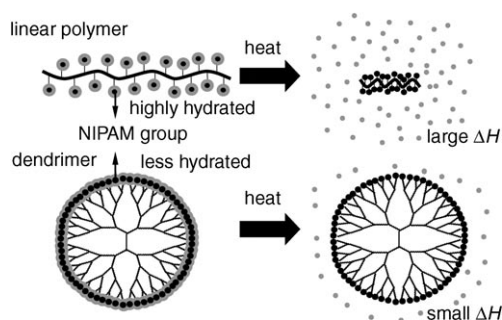


Figure 5. Schematic illustration of the phase transition of NIPAM-bearing polymer with a globular or a linear structure.

amount of structured water that solvates around NIPAM groups can be released upon the transition, thus yielding a large transition enthalpy. However, for the NIPAM-terminated dendrimers, the low conformational freedom that arises from their highly branched structure might cause dense packing of NIPAM groups in the periphery. Such a situation should lead to inefficient hydration around NIPAM groups below the LCST and inefficient dehydration of the NIPAM groups above the LCST, thereby resulting in the extremely small transition enthalpy.

In conclusion, we have demonstrated that marked differences exist in transition enthalpy, hydrophobicity, and sensitivity to urea between NIPAM-bearing dendrimers and linear polymers, which arise from their structural features. The thermosensitive dendrimers could undergo a sharp transition by dehydration of the peripheral moiety without a large conformational change of the whole molecule. Such properties, as well as the globular shape of the thermosensitive dendrimers, might be attractive for their use as intelligent nanocapsules for drug delivery and catalysis. The findings obtained through this study increase the understanding of thermosensitive polymers and expand their range of application.

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