

Investigation of the Self-Association Behavior of a Thermosensitive Copolymer with Lower Critical Solubility Temperature near Human Heat by Dynamic Laser Light Scattering

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ABSTRACT: The special thermosensitive copolymer with lower critical solubility temperature near human heat of 37°C in aqueous solution has been prepared successfully by copolymerizing *N*-isopropylacrylamide (NIPAM) and *N*-isopropylmethacrylamide (NIPMAM) randomly at an appropriate monomer ratio. The self-association behavior of the poly[NIPAM-*ran*-NIPMAM] copolymer has been studied by dynamic laser light scattering (DLS) at a concentration higher than the critical concentration of the self-associ-

ation calculated from viscosity data according to cluster theory. The effect of the concentration and temperature on the self-association behavior in water has been discussed respectively. © 2005 Wiley Periodicals, Inc. *J Appl Polym Sci* 96: 583–588, 2005

Key words: copolymerization; thermosensitive copolymer; self-association; cluster theory; viscosity; dynamic laser light scattering

INTRODUCTION

The self-association behavior of macromolecules in solution is a very important fundamental topic in both theory and application. But it is still a contestable problem about the critical condition of the self-association behavior taking place in solution. Recently, Cheng et al.^{1,2} proposed the cluster theory to claim that the critical concentration of the self-association should be consistent with the dynamic contact concentration C_s , which is a concept proposed by Qian et al.,^{3,4} in addition to the static contact concentration, i.e., overlap concentration, C^* . It is believed that the chain segments of the polymer coil start to feel the repulsive interaction between segments of neighboring coils, and the coil starts to shrink in dimension when the solution concentration $C > C_s$. The value of C_s lies in the concentration region of 10^{-3} to 10^{-4} g/mL, being much lower than the theoretically estimated static overlap concentration, C^* . The existence of C_s in the sense of coil shrinking in polymer solution

was confirmed later by precise size exclusion chromatogram⁵ and dynamic light scattering.⁶ Therefore it is reasonable to believe that the solution just contains real isolated polymer chains when the concentration of the solution is lower than C_s , on the contrary, when the concentration is higher than C_s , the interactions among the chains become operative to form multichain particles by self-association.

Recently, thermosensitive polymers have been given special attention for their potential applications. Many published works have dealt with both fundamental aspects and practical applications. Poly(*N*-isopropylacrylamide) (PNIPAM),^{7–13} which consists of both hydrophilic and hydrophobic groups in each repeat unit, has a lower critical solubility temperature (LCST) of around 32°C in the aqueous solution. When the so-called coil-to-globule transition takes place, the hydrodynamic volume of the polymer would decrease tremendously, which has great potential application, such as in drug delivery,¹⁴ separation,¹⁵ energy transduction,¹⁶ and catalysis.¹⁷ However, many biological reactions, especially in the human body, have to be carried out exactly at 37°C, so the thermosensitive carrier was required to have a transition temperature near human body temperature.

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Poly(*N*-isopropylmethacrylamide)^{18–21} has a LCST of around 45–48°C in water, whose hydrodynamic volume decreases in a similar way to that of PNIPAM. Recently, an attractive experiment was reported by Djokpé and Vogt²² in which a simple quantitative relationship between the phase transition temperature of poly[NIPAM-*ran*-NIPMAM] copolymers and the composition and the phase transition temperature could be adjusted between 32 and 48°C by controlling the composition of the two monomers in the copolymer, so a new thermosensitive sample with a phase transition temperature near the human temperature of 37°C in water could be made.

In this article, the self-association behaviors of the special thermosensitive copolymer sample in aqueous solution have been investigated by dynamic laser light scattering. For its special thermosensitivity, the self-association behavior should be related to not only the concentration, as mentioned above, but also the temperature, so the effect of the two influencing factors, concentration and temperature, on the self-association behavior have been studied respectively.

EXPERIMENTAL

Materials

NIPAM was purchased from ACROS, and NIPMAM was synthesized from methacryloylchloride and isopropylamine. The two monomers were recrystallized three times from a 65 : 35 (v/v) mixture of hexane and benzene before use. Azobisisobutyronitrile (AIBN) was recrystallized from methanol. The poly[NIPAM-*ran*-NIPMAM] copolymer sample was polymerized¹¹ by free radical reaction initiated by AIBN in methanol under nitrogen at 70°C. Its relative weight-average molar mass is 4.82×10^4 g/mol calculated from size exclusion chromatogram results, and the molecular weight distribution index (M_w/M_n) is 2.76.

Sample preparation

All of the solutions were prepared by being weighed and kept still at least 24 h at room temperature. They then were filtered through a Millipore filter with a 0.45- μ m pore diameter to remove dust before any measurements. Deionized water was used in all experiments. The weight concentration was converted to volume concentration (in g/mL) by applying the density correction for the later calculation.

Differential scanning calorimetry (DSC)

The copolymer solution was measured by a VP-DSC MicroCalorimeter (Microcal Inc). The cell volume was 0.5 mL. Temperature was increased or decreased

at the rate of 60°C per hour. The concentration of the copolymer solution was kept at 1.0×10^{-3} g/mL.

Viscosity measurement

A Ulldeband viscosimeter, with a capillary with a diameter of 0.413 mm and a length of 102.1 mm, was used to measure the change of the solution viscosity with the increasing concentration. The viscometer was thoroughly cleaned by repeatedly rinsing and soaking for 2 days with deionized water and finally drying at 110°C after every measurement.

Dynamic laser light scattering (DLLS)^{23,24}

A slightly modified spectrometer (ALV/DLS/SLS-5022F) equipped with a multi- τ digital time correlation (ALV5000) and a cylindrical 22 mW UNIPHASE He-Ne laser ($\lambda_0 = 632$ nm) as the light source was used. In DLLS, the Laplace inversion (the CONTIN procedure) of each measured intensity–intensity–time correction function $G^{(2)}(q,t)$ in the self-beating mode can be related to a line-width distribution $G(\tilde{\Delta})$. For a diffusion relaxation, $\tilde{\Delta}$ is related to the translation diffusion coefficient D by $(\tilde{\Delta}/q^2)_{c \rightarrow 0, q \rightarrow 0} \rightarrow D$, so that $G(\tilde{\Delta})$ can be converted into dynamic radius distribution $f(R_h)$ via Stokes–Einstein equation, $R_h = (k_B T / 6\pi\eta) D^{-1}$, where k_B , T , and η are the Boltzmann constant, the absolute temperature, and the solvent viscosity, respectively. Four aqueous solutions of copolymer samples at the concentrations of 0.922×10^{-3} , 1.30×10^{-3} , 3.25×10^{-3} , and 4.04×10^{-3} g/mL were prepared carefully and were measured by DLLS, respectively, at 25°C and a scattering angle of 90°. The copolymer solutions at the concentration of 3.25×10^{-3} g/mL were also measured by DLLS in the temperature range from 22 to 35.5°C and the scattering angle was also 90°. To guarantee reaching the equilibrium state at every temperature point, the temperature at every detected point was kept at least 30 min before measuring.

RESULTS AND DISCUSSION

According to Djokpé and Vogt's²² report, the poly[NIPAM-*ran*-NIPMAM] copolymer with LCST near human temperature in aqueous solution was prepared well, and the composition of the NIPMAM monomer in the mixture monomers before polymerization was about 25%. Figure 1 showed the DSC curve of the poly[NIPAM-*ran*-NIPMAM] aqueous solution. The heating and cooling cycle in the DSC experiment had been measured twice continuously, and the two results fitted with each other very well. From the heating curve in the DSC figure, the phase transition temperature was near 37°C, which was 2° higher than that from the cooling curve for kinetic reasons.

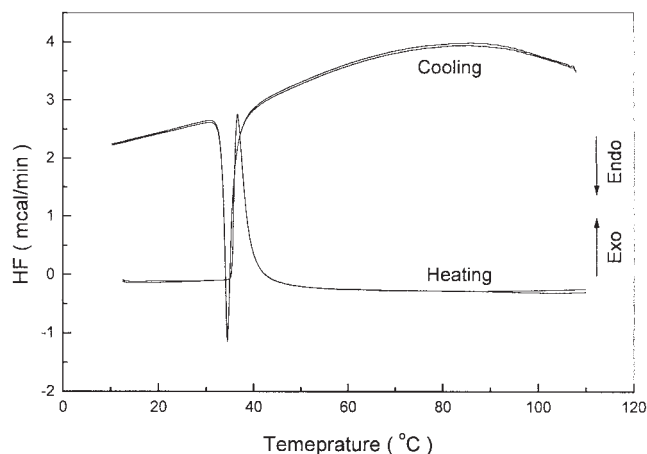


Figure 1 The temperature dependence of heat flow (HF) of poly[NIPAM-*ran*-NIPMAM] in water, where the heating and cooling rate of temperature is 60°C per hour.

The self-association behavior of the thermosensitive copolymer in water then was studied. According to cluster theory,^{1,2} the critical concentration of self-association in solution is consistent with the dynamic contact concentration C_s , and the C_s could be calculated directly from the viscosity data. On the basis of the cluster theory, the viscosity equation could be written as shown below

$$\frac{\eta_{sp}}{C} = [\eta] + 6K_m[\eta]C \quad (1)$$

wherein $[\eta]$ is the intrinsic viscosity and K_m is the apparent association constant.

K_m has a relationship with C_s , which is shown in eq. (2):

$$C_s = \frac{0.001}{2K_m} \quad (2)$$

Figure 2 showed the concentration dependence of the η_{sp}/C of the copolymer in water at 25°C. According to eq. (1), the $[\eta]$ and K_m could be calculated as 29.9 and 3.10 mL/g, respectively, and then, using eq. (2), C_s could be obtained, which gives a value of about 1.61×10^{-4} g/mL. Moreover, the C^* can also be estimated from the reciprocal of the intrinsic viscosity,²⁵ the value of which value is near 3.34×10^{-2} g/mL. From the cluster theory, it was believed that the copolymer coils began to contact each other to form associated bodies, i.e., multichain particles, when the concentration of the copolymer sample was higher than 1.61×10^{-4} g/mL.

The poly[NIPAM-*ran*-NIPMAM] aqueous solutions at the concentration range from 9.00×10^{-4} to 4.00×10^{-3} g/mL, which are all higher than the critical concentration of self-association but lower than C^* ,

were measured by DLLS, respectively, at 25°C. Figure 3 shows the concentration dependence of the hydrodynamic radius distribution $f(R_h)$ of the copolymer solutions by CONTIN simulation. At the temperature of 25°C, water is a good solvent for the copolymer. It was found that, from Figure 3, at the concentration of 0.922×10^{-3} g/mL, there was just one peak (Peak 1) in the curve visually, whose average size was near 7–8 nm, but with the concentration increasing, a new peak (Peak 2) appeared step by step, and the higher the solution concentration was, the more visible the Peak 2. It was believed that the two peaks in the curves of Figure 3 should stand for two different states of macromolecules in the solution. Figure 4 showed the concentration dependence of the average hydrodynamic radius and the percent of the two peaks' area in Figure 3. It could be seen from Figure 4(a) that, with the increase of the concentration, the position of Peak 1 changed very little, whereas that of Peak 2 increased obviously. It was believed that Peak 1 was related to a conformation independent of the solution concentration, whereas Peak 2 was related to a changeable conformation with the concentration, and the percent of the Peak 2's area would also increase synchronously according to Figure 4(b). Considering of the average size of the two peaks, the Peak 1 could be related to the single-chain state spontaneously, whereas Peak 2 was a multichain one by self-association.

However, as mentioned above, the concentration of the four solutions are all higher than the dynamic contact concentration, but there was just one peak visually in the hydrodynamic radius distribution curve at the concentration of 0.922×10^{-3} g/mL in Figure 3. Since the polymer coils began to form associated bodies of multichains at the concentration up to C_s , the single peak in the curve at 0.922×10^{-3} g/mL

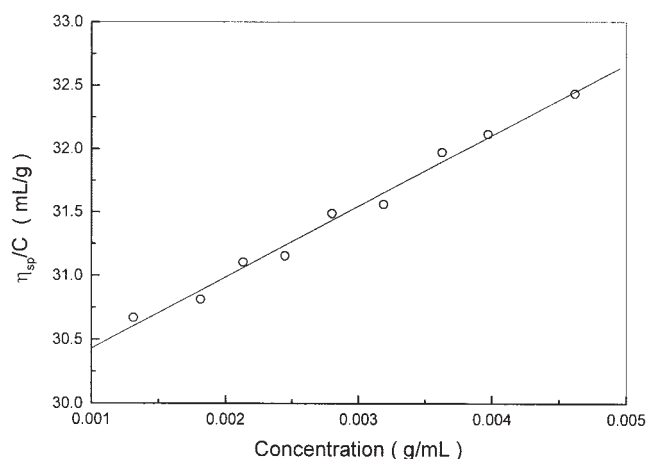


Figure 2 The concentration dependence of the η_{sp}/C of poly[NIPAM-*ran*-NIPMAM] in water, where the temperature is 25°C.

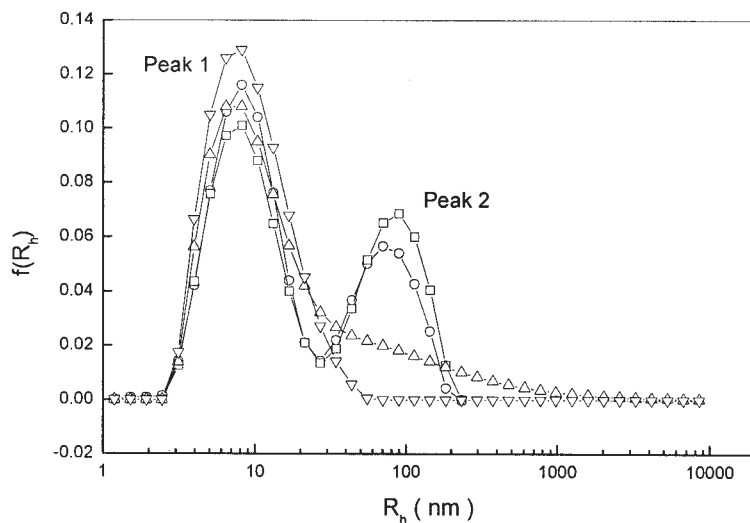


Figure 3 The concentration dependence of the hydrodynamic radius distribution $f(R_h)$ of poly[NIPAM-*ran*-NIPMAM] in water at 25°C and with a scattering angle of 90°, where the copolymer concentrations are 0.922×10^{-3} g/mL (∇), 1.30×10^{-3} g/mL (Δ), 3.25×10^{-3} g/mL (\circ), and 4.04×10^{-3} g/mL (\square), respectively.

resulted from not only a single-chain state, but also a multichain one. From the hydrodynamic radius distribution curve in Figure 3, the contribution of the multichain particles has been fully enshrouded by that of single-chain particles. It could be ascribed to two facts: one is the polydistribution of the sample itself, the other is that, despite the concentration of the solution being higher than C_s (but still at a very low value), the formed associated bodies are just made of a few chains, such as dimers or trimers and so on, in which the size still is very small. Moreover, the fraction of the associated bodies was also very low, so it was difficult to distinguish the multichain particles from the single-chain particles in the curve of their hydrodynamic radius distribution.

However, a new problem arose immediately following the above discussions, which was how to measure

the real distribution of the molecular weight of polymer sample and how much concentration should be used during the experiments of DLLS, GPC, and so on to detect their real distribution.

As we all know, even the ideal unidistribution polymer sample still displays a distribution of the conformation in solution, and the distribution of the conformation $w(R)$ could be followed by the Gaussian function, which is shown as follows:

$$w(R) = \left(\frac{3}{2\pi_{1/2}Nb_2} \right)^{3/2} \exp\left(-\frac{3R_2}{2Nb_2} \right) \quad (3)$$

where N , b , and R are bond number, bond length, and end-to-end distance, respectively. As for polydistribution polymer samples, the distribution of the conformation could be written as

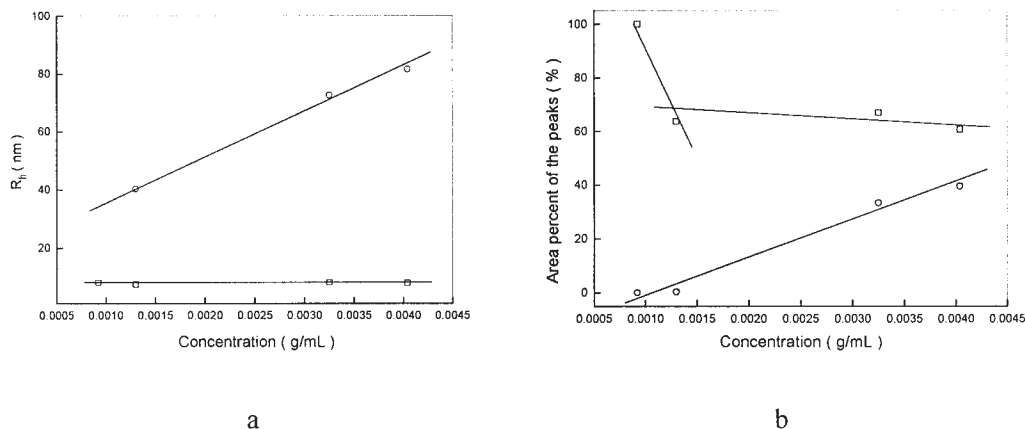


Figure 4 (a) The concentration dependence of the average hydrodynamic radius of the two peaks in Figure 3, where \square is related to peak 1 and \circ is relative to peak 2. (b) The concentration dependence of the percent of the two peaks area in Figure 3, where \square and \circ are the same as in a.

$$w(R) = \sum_N \left[W(N) \times \left(\frac{3}{2\pi_{1/2} N b_2} \right)^{3/2} \exp\left(-\frac{3R_2}{2N b_2} \right) \right] \quad (4)$$

where $W(N)$ is the weight fraction of the different compositions in sample with different molecular weights; if the concentration of the solution is higher than C_s , self-associated bodies would be formed and contribute to the distribution of the conformation, which could be described simply as follows:

$$w(R) = f(j, N, R) \quad (5)$$

in which j is the number of the chains to form the associated bodies of j -mer, and $f(j, N, R)$ is the contribution function related to j and N . It could be seen that eq. (5) is too complicated to give a distinct expression. It was believed that the apparent conformation distribution of polymer sample, measured by normal methods such as GPC, DLLS, and so on, could not reflect its real distribution for the polydistribution sample at a concentration higher than C_s . So it could be concluded that the conformation distribution of the sample must be measured at an appropriate concentration to prevent the effect of self-association from disturbing the real distribution.

Otherwise, the poly[NIPAM-*ran*-NIPMAM] copolymer is a thermosensitive sample in aqueous solution. The self-association behavior is also influenced by temperature. When the concentration of the sample is higher than C_s , the hydrodynamic radius distribution curve is more complicated. Figure 5 showed the temperature dependence of $f(R_h)$ of the copolymer range from 22 to 35.5°C, which was all lower than the transition temperature of 37°C, and the concentration of the copolymer solution was 3.25×10^{-3} g/mL. The temperature dependence of the average hydrodynamic radius and the percent of the two peaks' area was drawn in Figure 6. From Figure 6, the average hydrodynamic radius and the percent of the Peak 1 area decreased slowly at the beginning; however, when the temperature exceeded 29.2°C, the percent of the Peak 1 area decreased rapidly; at 34.9°C, near the transition temperature, Peak 1 disappeared abruptly. On the contrary, the average hydrodynamic radius and the percent of the Peak 2 area increased rapidly at the high temperature range. It was well known that the quality of the solvent water of the copolymer changed from good to bad with the increase in temperature; the polymer coils have shrunk and aggregated together to form bigger-size particles.

It could be seen that, when the temperature rose, the hydrodynamic radius distribution also changed a lot for the self-association behavior synchronously from Figure 5. And the detected conformation distribution curve of the sample was farther away from its real one.

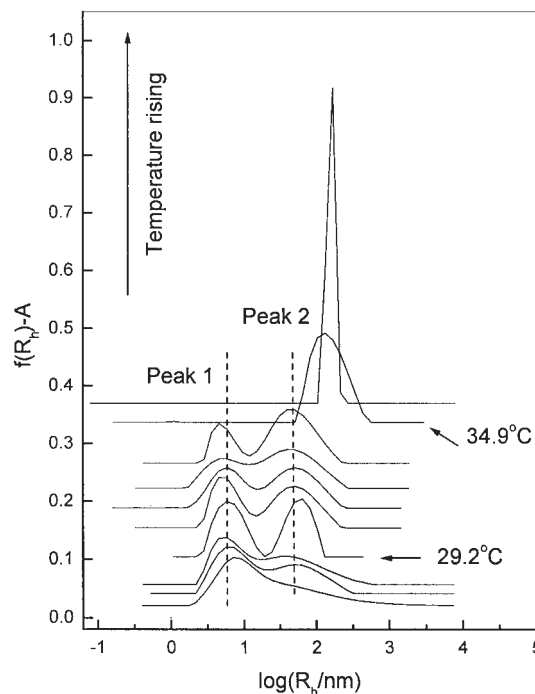


Figure 5 The temperature dependence of the hydrodynamic radius distribution $f(R_h)$ of poly[NIPAM-*ran*-NIPMAM] in water, where the copolymer concentration is 3.25×10^{-3} g/mL, the temperature is range from 22 to 35.5°C, and the scattering angle is 90°.

So the observed conformation distribution of the sample in solution should be corrected by a temperature factor α_T yet:

$$w(R) = \alpha_T \times f(j, N, R) \quad (6)$$

In comparison with eq. (5), eq. (6) seemed more complicated.

CONCLUSIONS

Above all, the self-association behavior of the thermosensitive polymer compared with the normal polymer samples was influenced by not only the concentration, but also the temperature. When the concentration was higher than the critical value, which could be calculated from viscosity data according to cluster theory, the self-association behavior had taken place, which gave great effect on the conformation distribution of the sample in solution. As for the factor of temperature, it was found that the coil shrinking also affected the copolymer conformation in solution with the quality of the solvent change into bad.

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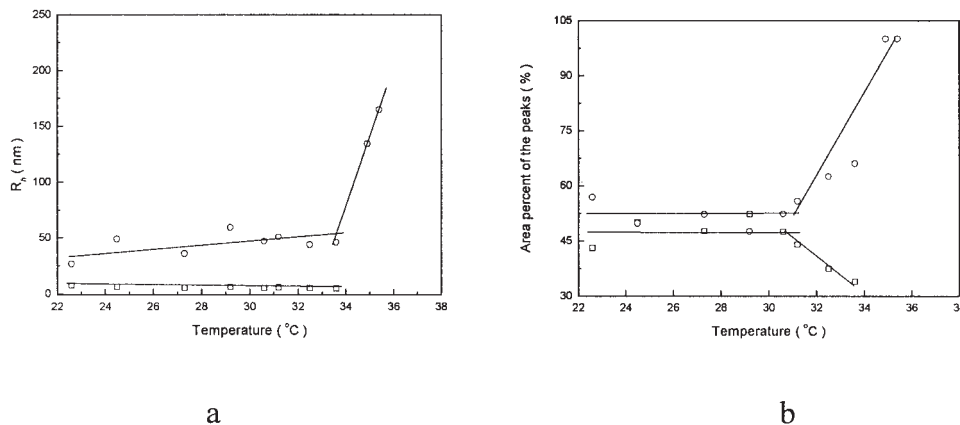


Figure 6 (a) The temperature dependence of the average hydrodynamic radius of the two peaks in Figure 5, where \square is related to peak 1 and \circ is relative to peak 2. (b) The temperature dependence of the percent of the two peaks area in Figure 5, where \square and \circ are the same as in a.

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