The Effect of Salts on the Conformation and Microstructure of Poly(N-isopropylacrylamide) (PNIPA) in Aqueous Solution

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ABSTRACT: We present an experimental investigation of the conformation and microstructure of Poly(N-isopropylacrylamide) (PNIPA) in aqueous solution in the presence of salts. As a model, a strong salting-out salt (Na$_2$SO$_4$) and a strong salting-in salt (NaSCN) were chosen. Light scattering measurements show that Na$_2$SO$_4$ decreases the radius of gyration of PNIPA compared to its value in water, whereas NaSCN increases it. Moreover, the NaSCN solution was found to be a better solvent for PNIPA compared to water, whereas Na$_2$SO$_4$ solution is worse. Small-angle neutron scattering measurements of semidilute PNIPA solutions, at temperatures well below the phase-transition temperature, exhibit the behavior predicted by the model of dynamic concentration fluctuations characterized by a single correlation length. Excess scattering at low angles is observed in salt solutions at temperatures that are near, yet below, the phase-transition temperature. This may indicate intrachain heterogeneities on the scale of 6–8 nm. © 2004 Wiley Periodicals, Inc. J Polym Sci Part B: Polym Phys 42: 3713–3720, 2004

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INTRODUCTION

Stimuli-responsive polymers exhibit a phase transition in response to changes in environmental conditions such as temperature,\textsuperscript{1,2} pH,\textsuperscript{3} light,\textsuperscript{4} electrolyte concentration,\textsuperscript{5} and electromagnetic field.\textsuperscript{6,7} A well known instance of a temperature responsive polymer is poly(N-isopropylacrylamide) (PNIPA),\textsuperscript{8,9} which is soluble in water below the lower critical solution temperature (LCST) of about 33–35 °C, yet precipitates at temperatures above it. Phase transition also appears in crosslinked PNIPA hydrogels that exhibit a dramatic, reversible volume change at the LCST.\textsuperscript{10–18} Shibayama et al.\textsuperscript{19} presented a comparative evaluation of the microstructure of PNIPA solutions and gels with small-angle neutron scattering (SANS), as a function of temperature and concentration. In terms of conformation changes, one can associate the phase transition of soluble PNIPA with the coil-to-globule transition. This transition was predicted theoretically,\textsuperscript{20,21} and variations of characteristic lengths (e.g., radius of gyration) with temperature were observed experimentally.\textsuperscript{22–26} In particular, Wu and coworkers\textsuperscript{24,25} have identified intermediate states in the coil–globule transition that appear above the θ temperature, namely, a crumpled coil and a molten globule. This scenario for the transitions of PNIPA in water were further supported by...
Fourier transform infrared (FTIR) measurements.\textsuperscript{17} Adding cosolutes to the system manipulates the LCST of PNIPA solutions,\textsuperscript{27,28} as well as the transition temperature and swelling ratio of PNIPA gels.\textsuperscript{29,30} As a rule, increased electrolyte concentration decreases the solubility of the polymer and reduces the LCST (salting-out effect). However, there are a few salts that behave in an opposite way, that is, exhibit salting-in. The mechanism of this phenomenon is not fully understood, and is the focus of recent studies. Paz et al.\textsuperscript{31} studied the effect of salts on PNIPA in aqueous solution, and concluded that sulfate ions induce partial intrachain collapse, manifested by a relatively compact structure well below the LCST temperature. An empirical estimation of the salting-out effect can be done with the Hofmeister (lyotropic) series:\textsuperscript{32,33}

\[
\text{SO}_4^{2-} < \text{HPO}_4^{2-} < \text{CH}_3\text{COO}^- < \text{Cl}^- < \text{Br}^- < \text{ClO}_4^- < \text{SCN}^- \quad (1)
\]

In expression (1), \(\text{SO}_4^{2-}\) is ranked as a strong salting-out ion, \(\text{Cl}^-\) a mild salting-out ion, and \(\text{SCN}^-\) a strong salting-in ion.\textsuperscript{34}

The main focus of the work cited above was studying the variation of the phase-transition temperature in response to changing external conditions, e.g., the type and concentrations of cosolutes. Thus, there is a lack of quantitative information regarding the changes occurring in the microstructure of the system. In this article, we describe an experimental investigation of the conformation and microstructure of PNIPA in dilute and semidilute aqueous solution in the presence of cosolutes. As a model, a strong salting-out salt (\(\text{Na}_2\text{SO}_4\)) and a strong salting-in salt (\(\text{NaSCN}\)) with the same cation were chosen.

**Experimental**

**Materials**

PNIPA was produced by Polymer Source, Inc. The molecular weight of PNIPA was 98,000 g/mol and the molecular weight distribution was \(M_w/M_n = 2.50\). \(\text{Na}_2\text{SO}_4\) (Mallinckrodt Baker) and \(\text{NaSCN}\) (Riedel-de Haen) were of analytical grade and used as received. All solutions were prepared with mili-Q water.

**Characterization**

Static light scattering (SLS) measurements were performed with a BI-200SM Research Goniometer System (Brookhaven Instruments Corp.). A Compass 415M solid-state laser (Coherent), generating monochromatic green light of 532 nm wavelength, was used. The detector assembly includes a selected photo-multiplier tube (PMT), dynode chain, and an integral amplifier/discriminator. Samples were placed in a 55 mm tall, 12 mm outside-diameter round glass cell with a Teflon cap, immersed in a glass vat containing decalin as the index matching fluid. A special effort was made to ensure that all samples were completely dust-free; otherwise, the dust particles would contribute to the scattering intensity and lead to erroneous results. For this reason, the analyzed solution was clarified before use by microfuge (0.2–0.5 mm porosity) pressure filtration. Windows-based Zimm Plot software Version 3.17 provided with the instrument was employed for data processing. The refractive index increment, \(dn/dc\), was measured separately with the BI-DNDC (Brookhaven Instruments Corp.) differential refractometer. For aqueous solutions of PNIPA, the refractive index increment was found to be equal to 0.25 mL/g. When we used this value for other systems there was good matching to the specified value of the PNIPA molecular mass.

SANS measurements were performed at the LOQ beamline of the ISIS spallation source [Rutherford Appleton Laboratory (UK)] with a two-dimensional position sensitive detector with time-of-flight measurement. Solutions were prepared in \(\text{D}_2\text{O}\) (Aldrich) and placed in quartz cuvettes (Hellma) 10 mm wide and 2 mm path length. Temperature was controlled \((\pm 0.2 \degree C)\) with a water circulator (Lauda). Data reduction to scattering vector \(q\), subtraction of scattering from an empty cell, and normalization with respect to a reference standard were performed with the Colette procedure.\textsuperscript{35} The measured SANS patterns exhibit a constant intensity at \(0.15 < q < 0.25\ \text{Å}^{-1}\) mostly due to solvent fluctuations and the incoherent scattering of hydrogen atoms of PNIPA. The former were measured separately and the latter were calculated from the solution composition. The constant background intensity was evaluated from the high-\(q\) region of the scattering curve and subtracted as well. This background level was within 20% of the background evaluated from measurements of the solvent scat-
tering to which the calculated incoherent scattering of PNIPA was added.

THEORETICAL BACKGROUND

SLS

The classical light scattering theory\textsuperscript{36,37} has shown that for a dilute polymer solution at a small scattering angle the weight-average molecular weight can be related to the excess time-average scattered light intensity (excess Rayleigh ratio), \( \Delta R_w \), by

\[
\frac{KC}{\Delta R_w} \approx \frac{1}{M_w} \left(1 + \frac{1}{3} (R_g^2 q^2) + 2A_2 C \right) \tag{2}
\]

where \( K \) is an optical constant, \( C \) is the polymer concentration (g/mL), \( M_w \) is the weight-average molecular weight (g/mol), \( R_g \) is the average radius of gyration, \( q = (4\pi n\lambda_c) \sin(\theta/2) \) is the scattering vector, \( n \) is the refractive index of the medium, \( \theta \) is the scattering angle, \( \lambda_c \) is the wavelength of the incident beam in vacuo, and \( A_2 \) is the second virial coefficient. A plot of \( KC/\Delta R_w \) versus \( q^2 + kC \), where \( k \) is an arbitrary constant, (known as a Zimm plot) is used to obtain the values of \( M_w, R_g, \) and \( A_2 \).

SANS

A semidilute polymer solution is usually characterized by a single length scale that is known as the correlation length, \( \xi \).\textsuperscript{38,39} The small-angle scattering curve \( I(q) \) of such a solution is described by the Ornstein–Zernike equation (Lorentzian form):

\[
I(q) = \frac{I(0)}{1 + q^2 \xi^2} \quad \text{for} \quad q\xi < 1 \tag{3}
\]

Recently Norisuye et al.\textsuperscript{40} derived eq 3 for a semidilute polymer solution following the treatment by Panyukov and Rabin\textsuperscript{41,42} of dynamic thermal concentration fluctuations, relating \( \xi \) to the polymer volume fraction \( \phi \) and the Flory parameter of polymer–solvent interactions \( \chi \):

\[
\xi = a (12\phi (1 - 2\chi + \phi))^{-\frac{1}{2}} \tag{4}
\]

where \( a \) is the monomer length. They also showed that eq 4 represented well the neutron scattering from an aqueous PNIPA solution.\textsuperscript{40}

The scattering from heterogeneities in the solution microstructure, which can be assumed to be of an arbitrary shape with a constant density and a sharp interface to the solvent, is fit by the Debye–Anderson–Brumberger equation, usually used to describe the scattering curve of random two-phase structures\textsuperscript{43}:

\[
I(q) = \frac{I(0)}{(1 + q^2 \xi_D^2)^2} \quad \text{for} \quad q\xi_D > 1 \tag{5}
\]

If the volume fraction of the heterogeneities is small, \( \xi_D \) is their characteristic dimension. In the limit of a large \( q \) eq 5 yields Porod’s law \( [I(q) \sim q^{-4}] \).

RESULTS AND DISCUSSION

Temperature changes are known to have a similar effect on PNIPA hydrogels and on their individual chains in dilute solution, that is, gel shrinking and reduction in the chain dimension were observed at the same temperature. As already discussed, it was found that the presence of salts changes the equilibrium volume of PNIPA gels compared to gels immersed in pure water. Yet, the question whether an analogous change in the chain dimensions occurs as well is still open. Light scattering experiments provide a direct means to answer this question. SLS measurements were performed for three series of solutions: PNIPA in water, PNIPA in 2 wt % NaSCN solution (salting-in conditions), and PNIPA in 2 wt % Na2SO4 solution (salting-out conditions). We chose to conduct the experiment in a temperature well below the LCST: 18 °C for PNIPA in water and in a NaSCN solution, and 8 °C for PNIPA in a Na2SO4 solution.

Figure 1 shows a typical Zimm plot obtained from SLS experiments of an aqueous solution of PNIPA with NaSCN. Similar plots were built for PNIPA solutions in water and with Na2SO4. Zimm plots enable a straightforward calculation of the average radius of gyration, \( R_g \), the second virial coefficient, \( A_2 \), and the average molecular mass of the polymer, \( M_w \), based on eq 2. The results of the calculation for the studied systems are shown in Table 1.
The results shown in Table 1 clearly demonstrate that PNIPA chains expand in an NaSCN solution compared to pure water, whereas they contract in an Na₂SO₄ solution. Thus, an analogy between gel shrinking or swelling and changes in chains dimensions does exist. Moreover, the values of the second virial coefficient $A_2$ are also affected by the salts, indicating changes in the quality of the solvent.

Livney et al. 34, who studied the influence of salts on the swelling and osmotic pressure of dextran, suggested a mechanism to explain their findings. According to this work, SO₄⁻/H₂O, which has a high surface charge density, forms a complex of tightly bound water molecules. If this complex is considered as a solvent for the polymer, because of both entropic and enthalpic considerations, the mixing free energy of the polymer in a salt solution will be higher compared with that in pure water. Contrary to this, SCN⁻ ions tend to reduce the free energy of mixing because of both attractive interactions of the anions with the polymer and the combinatorial entropy of their hydrated counter ions. 34 Our results suggest that a similar mechanism may be used to describe the effect of these salts on PNIPA as well, since the $A_2$ values listed in Table 1 indicate that an aqueous solution of NaSCN is a better solvent for PNIPA than water, whereas Na₂SO₄ solutions are worse solvents.

Once we have demonstrated the influence of salts on the PNIPA chain dimensions in dilute solutions, it is also interesting to study their effect on the conformation in a semidilute solution. In Figure 2, one can see the SANS patterns of aqueous solutions of PNIPA (0.5, 1, 2, and 3.5 wt %) at 8 °C. The scattering patterns from aqueous Na₂SO₄ solutions (2 wt %) of the same PNIPA concentrations, at 8 °C, are shown in Figure 3. When water with NaSCN (2%) was used as a solvent, the SANS patterns from the PNIPA solutions were essentially identical to those from salt-free solutions, and are thus not presented. The solid lines in Figures 2 and 3 are fits for eq 3.

The correlation length of the semidilute polymer solution, $\xi$, evaluated from the fit of eq 3, is shown as a function of concentration in Figure 4 for PNIPA aqueous solutions with and without Na₂SO₄. Considering a power-law for the measured data yields an exponent of about 0.6, which is lower than predicted by scaling theory for good solvent conditions: $\xi \sim c^{-3/4}$ 44,45, and is shown as a solid line in Figure 4. It is much lower than the scaling law expected at $\Theta$ solvent conditions: $\xi$

### Table 1. The Radius of Gyration and Second Virial Coefficient of PNIPA in Aqueous Solutions with and without Salts, Measured by Light Scattering

<table>
<thead>
<tr>
<th>System Composition</th>
<th>$A_2$, cm$^3$ mol/g$^2$</th>
<th>$R_g$, nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>PNIPA</td>
<td>$6.67 \times 10^{-4} \pm 6 \times 10^{-10}$</td>
<td>$30.4 \pm 0.3$</td>
</tr>
<tr>
<td>PNIPA:NaSCN</td>
<td>$1.18 \times 10^{-8} \pm 2 \times 10^{-10}$</td>
<td>$32.8 \pm 0.3$</td>
</tr>
<tr>
<td>PNIPA:Na₂SO₄</td>
<td>$4.4 \times 10^{-4} \pm 1 \times 10^{-9}$</td>
<td>$23.1 \pm 0.2$</td>
</tr>
</tbody>
</table>
1.46 This may indicate that at 8 °C the polymer experiences good solvent conditions even in the presence of the sulfate ions. However, a much better fit of the measured data is obtained with the Shibayama equation (9), with the monomer dimension $a = 0.812$ nm, as shown by the dashed line in Figure 4. The Flory interaction parameter obtained from the fit, $\chi = 0.445$, is reasonable for this rather hydrophobic polymer even at a temperature significantly below its cloud point, and is consistent with the low values of $A_2$ reported for dilute solutions at 18 °C in Table 1.

When the temperature was raised to 18 °C, the SANS patterns from the PNIPA solutions in water with and without NaSCN were not significantly changed. However, enhanced excess scattering at low angles was observed in the scattering from PNIPA solutions in water with Na$_2$SO$_4$, as demonstrated in Figure 5 for the 2 wt % solution. The excess scattering at low angles is much larger than expected from the increase in temperature only, as reported by Shibayama et al. for aqueous PNIPA solutions below yet near the cloud point. Furthermore, a much poorer fit of eq 3 to the low-$q$ part of the SANS patterns is obtained. We suggest that a more plausible model relates the excess scattering to inhomogeneities in the structure of the polymer coil that can form local regions of higher polymer concentration, consistent with the crumpled coil model of Wu and Wang. The excess scattering at low-$q$ is thus fit with eq 5, as shown by the solid lines in Figure 6, yielding a characteristic dimension of $7.8 \pm 0.2$ nm for all concentrations measured. Eq 5 is valid if the system is described as a random two-phase structure with sharp interfaces that may not be strictly true for the crumpled coil.

Nevertheless, we maintain that the unique shape of the scattering patterns from PNIPA in aqueous Na$_2$SO$_4$ solutions at 18 °C, relative to those in water or with NaSCN, indicates that the crumpled coil appears and is stable even well below the

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**Figure 3.** SANS patterns from aqueous solutions of PNIPA containing 1.9 wt % Na$_2$SO$_4$ at 8 °C. Polymer concentration (wt %): (◯) 0.5, (∗) 1, (△) 2, (♦) 3.5.

**Figure 4.** The correlation length $\xi$ evaluated by fitting eq 3 to SANS patterns from solutions of PNIPA in water (♦) and 1.9% Na$_2$SO$_4$ (◯), at 8 °C. Solid line: the scaling law; $\xi \sim c^{-3/4}$; dashed line: fit of the Shibayama equation (4).

**Figure 5.** SANS patterns from 2 wt % aqueous solutions of PNIPA containing 1.9 wt % Na$_2$SO$_4$ at 8 °C (◯) and at 18 °C (∗). Lines show the fit to eq 3.
phase-transition temperature. Preliminary analysis with eq 5 provides an estimate of the characteristic dimension of the heterogeneities in the crumpled coil state, which is about 8 nm. It should be noted that a two correlation lengths model (i.e., a summation of eqs 3 and 5) could be easily fit to the experimental data. However, such a model requires six-fitted parameters and, therefore, the reliability of the fit is rather low.

Intense excess scattering at low $q$ was also observed from PNIPA solutions in water and in 1.1% NaSCN at 30 °C, as shown in Figure 7. As with the solutions in Na$_2$SO$_4$ at 18 °C, the shape of the curve with an apparent inflection point and a sharp increase in intensity at low angles may indicate a heterogeneous structure, as described by the crumpled coil model. Application of eq 5 to the low-$q$ scattering in Figure 7 yields characteristic dimensions of 6.9 ± 0.4 and 6.2 ± 0.2 nm for solutions in water and in 1.1% NaSCN, respectively, with an insignificant dependence on polymer concentration.

When the PNIPA solutions in water with Na$_2$SO$_4$ were heated to 30 °C, their opaque white appearance indicated that the cloud point temperature had been surpassed. This is in accord with a previous report by Paz et al., following earlier work by Inomata et al., that showed the striking effect of strong salting-out salts such as NaCl on lowering the transition temperature of PNIPA gels. The enhanced scattering at low-$q$ now exhibits the scaling of Porod’s law $I(q) \sim q^{-4}$ in Fig. 8, as expected from large-scale heterogeneities of compact polymer globules.

The SANS measurements of the microstructure in semidilute PNIPA solutions in water and in aqueous solutions of Na$_2$SO$_4$ and NaSCN are interpreted here as indicating three stages in transitions as the temperature is raised through the cloud point. At a low temperature, the ordinary behavior of a semidilute solution of polymer coils is observed. At a higher temperature, yet below the phase-transition temperature, small-scale heterogeneities appear in the coiled state, as
suggested by the crumpled coil model of Wu and Wang\textsuperscript{25} and supported by spectroscopic measurements by Paz et al.\textsuperscript{31} This interpretation differs from the classical interpretation of changes in the SANS patterns of PNIPA solutions with rising temperature made by Shibayama et al.\textsuperscript{19} They analyzed the SANS patterns with eq 4 and evaluated the increase in $\xi$ and $I(0)$ with temperature as a critical divergence as the spinodal temperature is approached. It seems to us that the shape of the SANS patterns that we attribute to the crumpled coil state, with an apparent inflection point and enhanced scattering at low-$q$, even at temperatures much below the cloud point, differs from what was reported by Shibayama et al.\textsuperscript{19} The origin of this discrepancy is not clear and should be investigated further. A possible explanation may be that at the higher concentrations studied by Shibayama et al., the more entangled state of the polymer coils hinders the formation of intrachain heterogeneities that characterize the crumpled coil state.

**CONCLUSIONS**

We have studied the conformation poly(N-isopropylacrylamide) (PNIPA) in aqueous solution under salting-out and salting-in conditions. Light scattering measurements showed that there is an analogy between gel shrinking or swelling and changes in chain dimensions: The salting-out salt (Na$_2$SO$_4$) decreases the radius of gyration of PNIPA compared to its value in water, whereas the salting-in salt (NaSCN) increases it. Moreover, the second virial coefficients measured by light scattering indicate that the NaSCN solution is a better solvent for PNIPA compared to water, whereas Na$_2$SO$_4$ solution is a worse solvent.

The SANS patterns of semidilute solutions have been interpreted as representing three stages: semidilute polymer coils, crumpled coils, and collapsed polymer globules. The microstructure of PNIPA in water and in aqueous NaSCN at 8 and 180 °C and in aqueous Na$_2$SO$_4$ at 8 °C is readily described by the model of dynamic thermal concentration fluctuations characterized by a single correlation length $\xi$. Furthermore, the relationship of $\xi$ to polymer concentration followed the mean-field elaboration by Shibayama and co-workers\textsuperscript{19} of the structure factor presented by Panyukov and Rabin\textsuperscript{41,42}, rather than the prediction of scaling theory. At higher temperatures, yet below the cloud point, the SANS patterns may be interpreted as indicating small-scale heterogeneities (ca. 6–8 nm), following the crumpled coil model. This kind of behavior is observed in PNIPA solutions containing Na$_2$SO$_4$, even at 18 °C. Studies are currently underway to further evaluate the crumpled coil state.

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**REFERENCES AND NOTES**