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Pressure Effects on the Aggregation of Poly(*N*-isopropylacrylamide) and Poly(*N*-isopropylacrylamide-*co*-acrylic acid) in Aqueous Solutions

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ABSTRACT: Pressure effects on the phase separation of aqueous solutions of poly(*N*-isopropylacrylamide) (pNIPA) and poly(*N*-isopropylacrylamide-*co*-acrylic acid) (pNIPA/AAc) were examined at pressures up to 250 MPa. The cloud point temperature first increased and then decreased as the pressure was raised. The copolymer solutions had higher maximum temperature and pressure than the pNIPA solutions. Though the cloud point curve of pNIPA solutions had no concentration dependence, the cloud point curve of pNIPA/AAc solutions shifted toward higher temperature and pressure with decreasing concentration. The behavior of the specific volume of a 3.0 wt % pNIPA solution during the phase separation suggested that the hydrophobic interaction is not the main factor in the pressure dependence of the cloud point of pNIPA solution at high pressure.

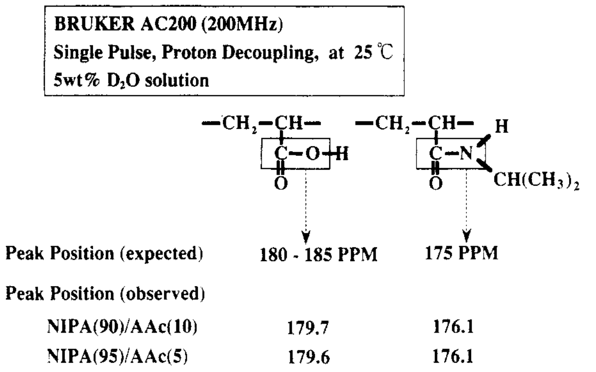
I. Introduction

Poly(*N*-isopropylacrylamide) (pNIPA) hydrogels are well known to undergo a volume phase transition in water in response to an infinitesimal change in temperature.[1-4] Similarly, aqueous solutions of NIPA polymer and many other poly(*N*-alkylacrylamide) derivatives are also known to have a lower critical solution temperature (LCST) type phase diagram, and the temperature where the polymer aggregates depends on the hydrophobic nature of the alkyl groups.[5] For the volume phase transition of temperature-sensitive hydrogels and LCST characteristics of the polymer solutions, the importance of interactions between polymers and solvents, such as cononsolvency, hydrophilicity, and hydrophobicity, has been pointed out by many researchers.[4-15] If hydrophobic interaction is responsible for the aggregation of pNIPA, the aggregated polymer chains should dissolve at high pressure, because hydrophobic aggregation is accompanied by a positive volume change.[16-18] Recent reports on the volume phase transition of NIPA gels and polymers under hydrostatic pressure seem to support the above expectation.[19,20] Examination of the pressure effects on the thermal aggregation of pNIPA would provide useful knowledge about the machanism of the phase behavior of the hydrophobic polymers.

The investigation of pressure effects on the hydrophobic interaction is also important for technological applications. High pressure is applied for food processing because foods are sterilized by pressure without changing their taste and fragrance.[21] Also, in some biochemical processes, pressure increases reaction rates and decreases the activity of enzymes.[22] Pressure effects are mainly caused by the denaturation of proteins and other biocompounds.[23-26] However, as proteins contain many hydrophobic and hydrophilic groups, it is very difficult to explain the pressure effect on all functional groups. It is, therefore, appropriate to examine pressure effects on hydrophobic model polymers.

In this paper, we focus on the contribution of the hydrophobic interaction to the pressure-induced aggregation/dissolution of aqueous pNIPA solutions by following pressure effects on the cloud temperature of pNIPA solutions. Experiments were also conducted with copolymers of *N*-isopropylacrylamide and acrylic acid (pNIPA/AAc) to examine the effect of hydrophilic groups incorporated in the hydrophobic chain.

Scheme I
Analysis of Monomer Ratio by ^{13}C NMR



Since hydrophobic interaction would be accompanied by a change of volume, we examined such volume change of a pNIPA solution during phase separation.

II. Experimental Section

1. Preparation of Samples. The polymer of NIPA (pNIPA) was synthesized by a free radical polymerization using AIBN initiator in benzene/acetone at 70 °C in a sealed tube. The copolymers of NIPA and AAc similarly synthesized. Mixtures of 95 mol % NIPA and 5 mol % AAc (pNIPA(95)/AAc(5)) and 90 mol % NIPA and 10 mol % AAC (pNIPA(90)/AAc(10)) were polymerized at 60 °C. Polymer molecular weight and its polydispersity were measured by GPC. The NIPA/AAc ratios of the copolymers were analyzed by ^{13}C NMR (Scheme I). Results of GPC and NMR analysis are given in Table I.

The synthesized polymers were dissolved in acetone, precipitated by *n*-hexane, dried in vacuo for 24 h at 50 °C, and stored in a sealed bottle. Before the experiments, the polymers were dissolved in degassed and deionized water and stirred for 24 h.

2. Cloud Point Measurements. Figure 1 shows the experimental apparatus (Figure 1a), high-pressure vessel (Figure 1b, Hikari Koatsu Co., Ltd.), and Pyrex glass cell (Figure 1c) used

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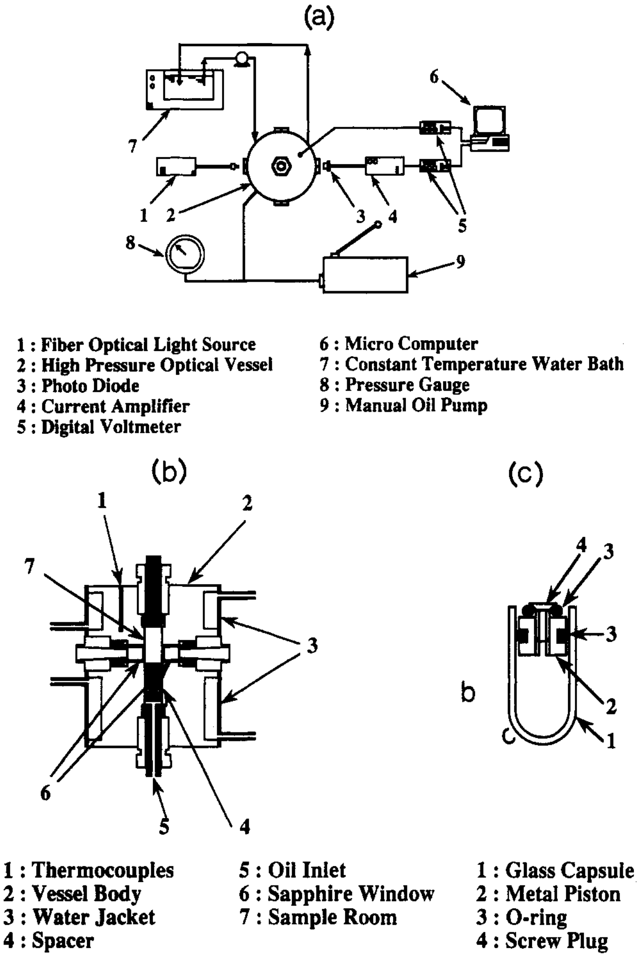
Table I
Properties of Samples

sample	M_w	M_w/M_n	% AAc
pNIPA	2.4×10^5	3.0	
pNIPA(95)/AAc(5)	2.6×10^6	1.8	9.5
pNIPA(90)/AAc(10)	2.9×10^6	1.7	4.6



1 : Fiber Optical Light Source
2 : High Pressure Optical Vessel
3 : Photo Diode
4 : Current Amplifier
5 : Digital Voltmeter
6 : Micro Computer
7 : Constant Temperature Water Bath
8 : Pressure Gauge
9 : Manual Oil Pump

1 : Thermocouples
2 : Vessel Body
3 : Water Jacket
4 : Spacer
5 : Oil Inlet
6 : Sapphire Window
7 : Sample Room

1 : Glass Capsule
2 : Metal Piston
3 : O-ring
4 : Screw Plug

Figure 1. Experimental apparatus for cloud point measurements: (a) schematic figure of the experimental apparatus; (b) cross-sectional figure of the high-pressure vessel; (c) schematic figure of the sample cell.

in the experiments. The vessel has four sapphire windows (12 mm diameter × 12 mm thick), and the maximum pressure for this apparatus is about 1000 MPa. The cloud points of the polymer solutions were determined by using a photodiode to measure the intensity decrease of light transmitted through the glass cell and a pair of sapphire windows of the high-pressure vessel. The uncertainty in the measurements of the temperature and pressure were within ±0.08 K and ±0.4 MPa, respectively.

Aqueous polymer solutions (0.5, 1.0, 3.0, and 5.0 wt %) were sealed in the glass cell (Figure 1c). The cell was placed in the high-pressure vessel and pressurized to a scheduled value, and the temperature was raised slowly (0.1 °C/min). The cloud temperature at a given pressure was determined from the intersection of the baseline and the leading edge of the intensity–temperature curve. The temperature was then decreased well below the cloud temperature. After the solution became transparent again, the pressure was increased to another scheduled value, and the temperature was slowly increased again. This procedure was repeated until a whole cloud point curve was obtained. The cloud temperature at atmospheric pressure was determined by a differential scanning calorimeter.[5,8]

3. Specific Volume Measurements. The specific volume of the 3.0 wt % pNIPA solution was measured using a glass piezometer similar to that of Kumagai and Takahashi.[28]

III. Results

Experimental results of the pressure dependence of the cloud point are shown in Figures 2 and 3. For pNIPA solutions, the cloud points have a small concentration dependence, but for the copolymers, the concentration dependence is large. The lower the concentration, the higher the cloud point temperature and pressure at which the LCST is observed. The copolymer solution had a larger one-phase region compared with the pNIPA solutions.

For the 0.5 wt % solutions of the copolymers, it was very hard to determine the cloud points near the maximum. Especially in the case of the pNIPA(90)/AAc(10) solution, when the temperature increased faster than 0.5 °C/min, no cloud point was detected up to 80 °C, the highest temperature of which the experimental apparatus could be used. In addition, even when the cloud point was determined, clouding of the solution was not complete up to 80 °C.

Experimental results of the specific volume measurement are shown in Figures 4 and 5. The volumes were normalized to the specific volume of pure water ("specific volume ratio" (SVR)) under the same conditions. The data for water were obtained by linear interpolation of published values.[29]

Figure 4 shows the SVR at atmospheric pressure. The specific volume of the polymer solution abruptly increased at the cloud temperature. The volume change due to the

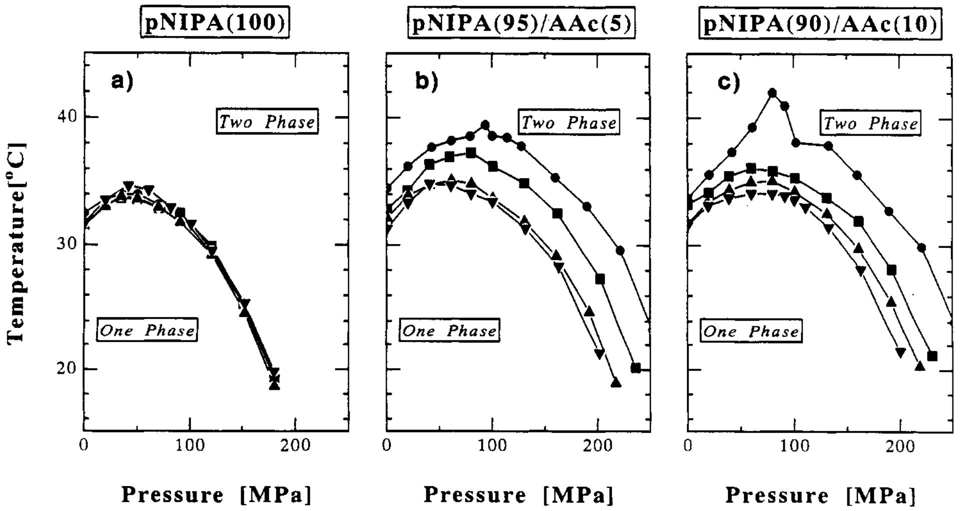


Figure 2. Pressure dependence of the cloud point of the pNIPA and pNIPA/AAc solutions as a function of the polymer concentration and composition: (●) 0.5 wt %; (■) 1.0 wt %; (▲) 3.0 wt %; (▼) 5.0 wt %.

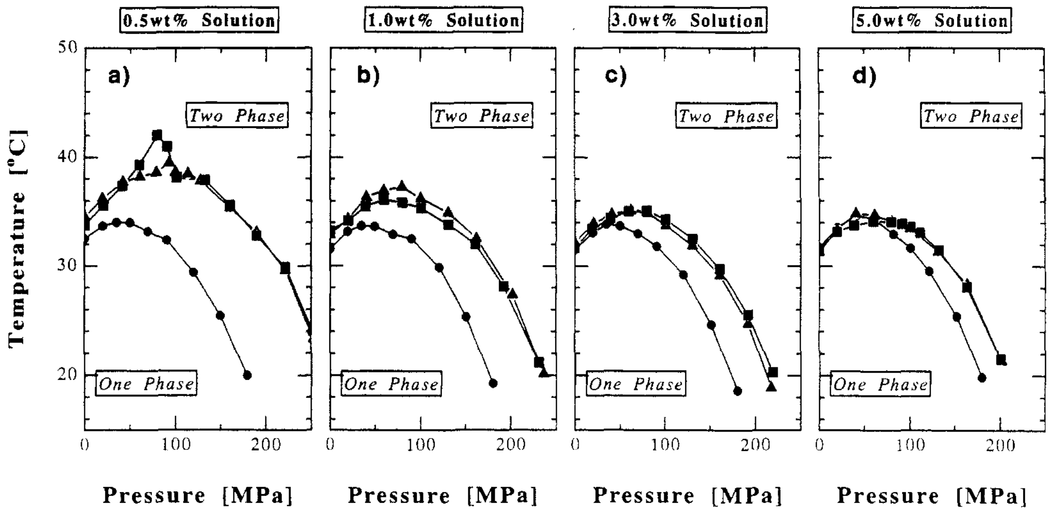


Figure 3. Pressure dependence of the cloud point of the pNIPA and pNIPA/AAc solutions as a function of the polymer concentration and composition: (●) pNIPA; (▲) pNIPA(95)/AAc(5); (■) pNIPA(90)/AAc(10).

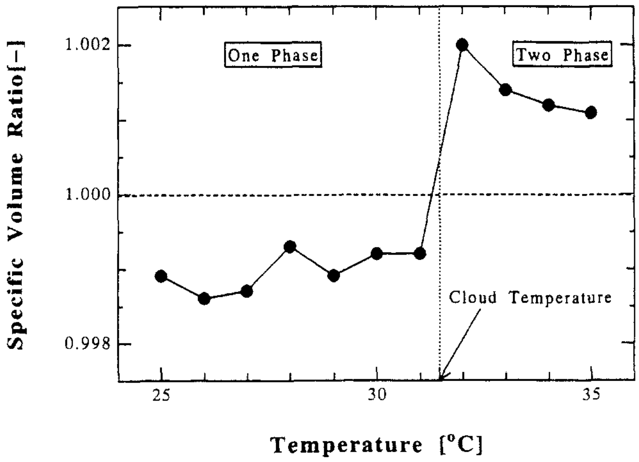


Figure 4. Specific volume ratio of the 3.0 wt % pNIPA solution at atmospheric pressure.

aggregation of polymer chains at atmospheric pressure was 10.6 cm^3 per mole of NIPA monomer unit. The value is consistent with the volume change accompanying the dehydration of a methylene or methyl group, estimated as 0.5–2 cm^3/mol at atmospheric pressure.[30]

Figure 5a shows the SVR at high pressure. Experimental conditions are shown in Figure 5b as horizontal lines. In the figure, open circles represent the experimental cloud point obtained in this work. It is seen that the SVR does not change on phase separation, and the temperature at which the large change in SVR occurs is independent of pressure.

IV. Discussion

The most interesting features of the cloud temperature behavior obtained in this work are the existence of the maximum as shown in Figures 2 and 3 and the pressure independence of the SVR of the pNIPA solution under high pressure shown in Figure 5. Previous papers suggested the importance of hydrophobic interaction for the thermally induced aggregation of pNIPA at atmospheric pressure, which would have implied that the cloud point should increase monotonously with an increase in pressure.

As we expected, at low pressures (up to ca. 50 MPa for pNIPA), the cloud temperature increased with an increase in pressure. The results of earlier reports by Lee et al. (0.1–10 MPa)[19] and Ohta et al. (0.1–40 MPa)[20] were obtained in this low-pressure region. In both reports, they concluded that pressure prevents the aggregation of polymer chains. The unexpected observation of a cloud point maximum suggests the existence of some factors other than hydrophobic interaction acting on the aggregation of polymer chains under high pressure.

Similar maxima in the dependence of solubilities on pressure were reported by Sawamura et al.[30] for aromatic hydrocarbons in water. They calculated the volume change accompanying the solute hydration and concluded that at

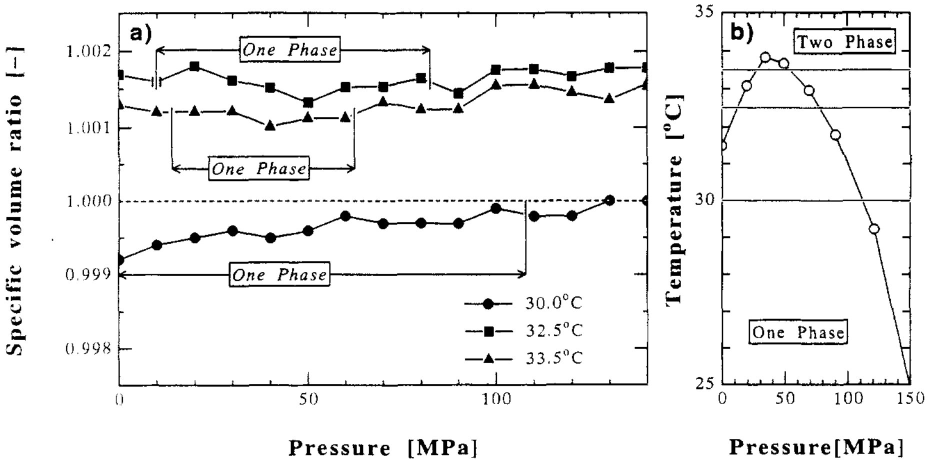


Figure 5. Specific volume ratio of the 3.0 wt % pNIPA solution at high pressures. (a) Pressure dependence of the SVR of the 3.0 wt % pNIPA solution at various temperatures. (b) Experimental conditions for the SVR measurements.

the maximum pressure the volume change turns from negative to positive. They interpreted the maximum as the destruction of the hydration shell around the solute which stabilizes the solute molecule in the solvent. This may also be an explanation for the cloud point curves of polymer solutions. However, the results of SVR measurements also have to be considered.

As shown in Figure 4, at atmospheric pressure, the cloud point coincides with the point where the SVR increases. This suggests that the aggregation of polymer chains at atmospheric pressure could be explained by hydrophobic interaction. However, the results in Figure 5 show that the temperature where the volume change occurs is independent of pressure and exists between 30 and 32.5 °C. If the dissolution of aggregated chains reflected hydrophobic interaction, the SVR should change at the cloud point and be larger in the two-phase regions than in the one-phase region. Experimental results suggest that some factors stabilize the polymer chains in the solution in spite of the occurrence of the hydrophobic interaction. At the present stage of the study, we cannot suggest what factors are effective for the aggregation of polymers under high pressure. Clearly, it is not the hydrophobic interaction. A spectroscopic technique may be useful for the investigation of this phenomenon.[31]

As shown in Figures 3 and 4, copolymerization with AAc shifts the cloud point curves toward higher temperature and pressure. Solutions of copolymers which had a similar molecular weight had almost the same cloud point curves for the same concentrations in spite of the difference in the content of AAc. It is well known that under high pressure, the dissociation constant of weak acids increases with an increase in pressure.[32] For example, the dissociation constant of acetic acid at 2000 atm became 2.24 times larger than that at atmospheric pressure. However, in this case, it seems hard to explain all effects of AAc by the Donnan effect only.

The largest differences between the pNIPA and the copolymers except for the copolymerized AAC are the molecular weight and its polydispersity. The cloud point of aqueous solutions should be independent of molecular weight at atmospheric pressure because the controlling factor for the aggregation is the hydrophobic interaction.[5,8] However, at high pressure, the hydrophobic interaction is no longer an important factor. The cloud point may then depend on the molecular weight, but, as the polymer samples used were not fractionated, more experimental data are needed to establish this point.

The concentration dependence of the cloud point curve could be explained by the shape of the coexistence curve of polymer solutions. In general, the cloud point increases with the dilution of polymer solutions which exhibit a LCST type phase diagram. The difference in the concentration dependence between pNIPA and copolymers may reflect a difference in molecular weight and polydispersity.

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