Temperature-Responsive Properties of Poly(acrylic acid-*co*-acrylamide)-*graft*-Oligo(ethylene glycol) Hydrogels

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ABSTRACT: A series of temperature- and pH-responsive hydrogels were prepared from acrylic acid (AAc), acrylamide (AAm), oligo(ethylene glycol)monoacrylate (OEGMA), and oligo(ethylene glycol)diacrylate by varying the AAc:AAm molar ratio and the OEGMA content. Phase-transition temperatures and swelling ratios of the obtained poly(AAc-*co*-AAm)-*graft*-OEG gels were measured as a function of temperature and pH. At pH < 5, the obvious transition temperatures ranging from 5 to 35°C were obtained as the AAc : AAm molar ratio was varied. The highest transition temperature was obtained at the AAc : AAm ratios of 5 : 5 and 6 : 4, and the sharp transition curves were observed at the AAc : AAm ratios from 5 : 5 to 8 : 2. The transition temperature further increased with increasing OEGMA content. It was suggested that OEG graft chains with a large mobility played an important role for the formation of hydrogen bonding in the hydrogels. The gels prepared here showed obvious reproducibility of the phase transition in response to temperature changes, which suggests the feasibility of their practical applications. © 2001 John Wiley & Sons, Inc. J Appl Polym Sci 80: 798–805, 2001

Key words: acrylic acid; acrylamide; oligo(ethylene glycol); graft copolymer; temperature-responsive hydrogel; hydrogen bonding

INTRODUCTION

Approaches such as stimuli-sensitive and swelling-controlled hydrogels have received much attention in current pharmaceutical research, because these gels can change the release rate of incorporated drugs according to the stimuli. It is well known that poly(*N*-isopropylacrylamide) (PNIPAAm) gel in water undergoes a volume phase transition in response to temperature changes.[1,2] This transition is dependent on the specific hydrophilic/hydrophobic balance effects and is sensitive to comonomers incorporated into the network.[3,4] There are many reports on self-regulated drug-delivery systems containing PNIPAAm.[4,5] In practice, however, such hydrogels are not suitable in the case of high body temperature, since the drug-release rate is essentially increased by the swelling of the gel at low temperature. An increase in body temperature induced by the presence of pathogens may be an important stimulus to the effective release of antihyperpyretic drugs. We have already reported temperature-responsive poly(acrylic acid) (PAAc)-*graft*-oligo(NIPAAm) hydrogels that release a drug at high temperature.[6]

On the other hand, hydrogels based on the interpolymer hydrogen bonds have been reported. Osada studied the interactions between a poly(methacrylic acid) (PMAA) membrane and poly-

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(ethylene glycol) (PEG) and revealed that addition of a PEG solution to the PMAA membrane caused a contraction of the membrane.[7] Klier et al. prepared self-associating PMAA-*graft*-PEG hydrogels.[8] Due to the hydrophobic stabilization, PMAA–PEG hydrogen-bonding complexes get stronger as temperature increases. Similarly, hydrogen-bonding complexes composed of PAAc and PEG result in the precipitation from solution[9]; the self-hydrogen-bonding in PAAc is solvated by water, whereas water does not displace the acid–ether hydrogen bonds in the complexes.[10] However, due to the lack of hydrophobic stabilization, PAAc–PEG hydrogen-bonding complexes dissociate as temperature increases,[11] contrary to the PMAA–PEG system. Moreover, PAAc and polyacrylamide (PAAm) are known to form strong hydrogen-bonding complexes below 25°C in an aqueous solution.[12] However, a random copolymer prepared from AAc and AAm restricts the complex formation and shows dull swelling/deswelling changes with temperature.[13] Katono et al. synthesized temperature-responsive interpenetrating polymer network (IPN) hydrogels composed of PAAc and PAAm,[13] and Lee et al. prepared semi-IPN hydrogels from PAAc and poly(vinyl alcohol).[14] These hydrogels are suitable for the drug-release devices at high body temperature.

In this article, as a part of our research on the thermo-sensitive hydrogel systems that contract at lower temperature and swell at higher temperature, we report the synthesis and swelling/deswelling properties of the poly(AAc-*co*-AAm)-*graft*-oligo(ethylene glycol) [P(AAc-*co*-AAm)-*graft*-OEG] gels, intending its application to controlled drug release and delivery. Copolymers of AAc and AAm were adopted as basic networks and OEG graft chains were introduced to control the phase-transition temperature.

EXPERIMENTAL

Materials

OEG (nominal MW = 600), acryloyl chloride, AAc, AAm, *N*,*N*-methylenebisacrylamide (MBAAm), and ammonium persulfate (APS) were purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan). Dichloromethane and triethylamine were distilled from CaH_2 just before use. All other chemicals were of reagent grade and used without further purification.

Preparation of OEG Monoacrylate (OEGMA) and OEG Diacrylate (OEGDA)

OEG (32.6 cm^3, 61.8 mmol) was dissolved in 100 cm^3 of dichloromethane, and a small amount of dichloromethane containing acryloyl chloride (5.0 cm^3, 61.8 mmol) and triethylamine (8.8 cm^3, 61.8 mmol) was added to the solution very slowly with stirring at 0°C. The reaction mixture was further stirred at room temperature for 4 h. After filtration of precipitated triethylammonium chloride, the solvent was evaporated. The product obtained as a viscous liquid was dissolved in a small amount of water, deionized with a mix-bed ion-exchange resin (Amberlite IRA-900 and Dowex HCR-W2) column, and then concentrated. The yield of OEGMA was 86.5%. In the same way, OEGDA was prepared from OEG (3.26 cm^3, 6.18 mmol), acryloyl chloride (1.0 cm^3, 12.4 mmol), and triethylamine (1.76 cm^3, 12.4 mmol). The yield of OEGDA was 92.3%. The obtained OEGMA and OEGDA were characterized by ^{1}H-NMR spectroscopy. The spectra were obtained in $CDCl_3$ with a Bruker ARX 300 spectrometer at 25°C.

Preparation of PAAc Gel

AAc (1.2 cm^3, 17.6 mmol), MBAAm (6.3 mg, 4.09 $\times 10^{-3}$ mmol), and APS (2.5 mg) were dissolved in deionized water (1.1 cm^3), and the solution was poured into a glass vial (i.d. 2 cm) and polymerized at 70°C for 6 h under a nitrogen atmosphere. After that, unreacted impurities were extracted with excess deionized water.

Preparation of P(AAc-*co*-AAm)-*graft*-OEG Gel

AAc (0.06–0.54 cm^3, 0.88–7.92 mmol), AAm (0.063–0.56 g, 0.88–7.92 mmol), OEGMA (0–280 mg, 0–0.44 mmol), and OEGDA (28.7 mg, 4.09 $\times 10^{-3}$ mmol) were dissolved in deionized water to obtain 10% solutions of monomers. Nitrogen was bubbled through the solution for 20 min to remove oxygen. Subsequently, 2.5 mg of APS was added to the solution, and the solution was poured into a glass vial. The radical polymerization was carried out under nitrogen at 70°C for 6 h. The obtained gels were extensively washed with deionized water. The feed compositions of the copolymers are listed in Table I, and the synthetic route is summarized in Figure 1. IR spectra of the gels were recorded on a Hitachi 270-50 IR spectrophotometer by a KBr pellet method after lyophilization.

Table I Feed Compositions for Preparation of P(AAc-*co*-AAm)-*graft*-OEG Gels

Sample Code	AAc (mmol)	AAm (mmol)	PEGDA (mmol)
10 : 90	0.88	7.92	0.0041
20 : 80	1.76	7.04	0.0041
30 : 70	2.64	6.16	0.0041
40 : 60	3.52	5.28	0.0041
50 : 50	4.40	4.40	0.0041
60 : 40	5.28	3.52	0.0041
70 : 30	6.16	2.64	0.0041
80 : 20	7.04	1.76	0.0041
90 : 10	7.92	0.88	0.0041

Amount of PEGMA was changed every 0.088 mmol.

Transmittance Measurement

Volume-phase transition of the gel was traced by monitoring the optical transmittance at 600 nm on a Shimadzu UV-120-02 spectrophotometer. The P(AAc-*co*-AAm)-*graft*-OEG gel equilibrated in a 1/15 mol/dm^3 phosphate buffer solution (PBS) of pH 3.0 overnight was cut into pieces and placed in a cuvette. A cuvette holder in the spectrophotometer was thermally controlled by using a heating circulator. A thermocouple was inserted into the gel in the cuvette to obtain an accurate temperature reading. The temperature of the sample in the cuvette was changed from 2 to 50°C, and the transmittance value was read every 1° increment. The time duration between each temperature increment was 10 min. To examine the reversibility of the phase transition of the P(AAc-*co*-AAm)-*graft*-OEG gel, the temperature of the cuvette was alternately changed between 10 and 40°C over several cycles every 20 min using the same gel.

Determination of Swelling Ratio

Each gel was equilibrated in a predetermined PBS of pH 3.0–7.0 at 10°C in a water bath. The swollen gels were taken out of the PBS, tapped with filter paper to remove excess solution on the surface, and then weighed. Finally, the gels were dried to constant weight *in vacuo* at 50°C. The swelling ratio was calculated by dividing the swollen gel weight, W_s, by the dried polymer weight, W_d.

RESULTS AND DISCUSSION

Volume-phase Transition of PAAc Gel in OEG Solution

Hydrogen bonding is the primary determinant to change a swollen state of hydrogels that include proton-donating and proton-accepting groups. Microphase separation between polymer-rich and polymer-poor domains occurs before the system undergoes the volume-phase transition,[15] that is, the cloud point indicates microscopic inhomogeneity before the volume-phase transition when temperature decreases. We preliminarily measured the optical transmittance at 600 nm to estimate the phase-transition temperature of the crosslinked PAAc gel in an aqueous solution of

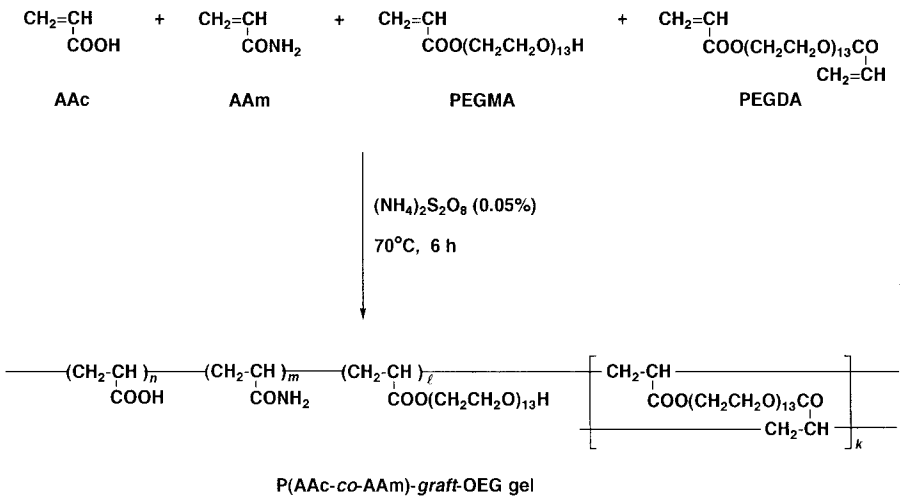

Figure 1 Synthetic route for P(AAc-*co*-AAm)-*graft*-OEG gels.

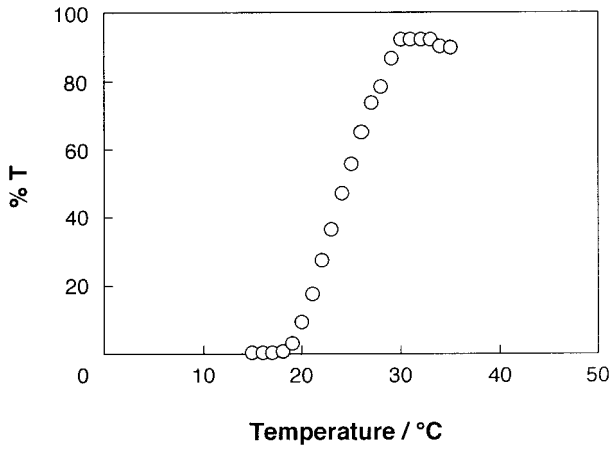

Figure 2 Temperature dependence of optical transmittance of PAAc gel in 40% aqueous solution of OEG.

OEG as shown in Figure 2. Figure 2 reveals that the PAAc gel becomes heterogeneous at the cloud point of 30°C when temperature decreases; the cloud point was determined from the intersection of the baseline at the highest transmittance and the tangent to the decreasing transmittance. Proton-accepting and proton-donating polymers may be involved in a negative heat of mixing (exothermic). This implies that the attractive interactions between the two polymers increase and those between polymer and solvent decrease with decreasing temperature. When the temperature of the system is increased, on the contrary, each polymer expands due to the increased compatibility with water and results in separate networks.

These phenomena allow us to expect that the hydrogel consisting of PAAc and OEG will contract at lower temperature and swell at higher temperature, and the practical problem is how we can introduce OEG chains with a large mobility into the network. If the polymers are covalently attached to each other, the probability of polymer–polymer interactions increases and complexation becomes much easier, since the local concentrations of the complexing species increase.[8] This means that covalent attachment of the complexing species is favorable for complexation. We introduced OEG graft chains to improve the phase-transition properties of the poly(AAc-*co*-AAm) gel adopted as a basic network.

Preparation of P(AAc-*co*-AAm)-*graft*-OEG Gel

We used OEG with the molecular weight of 600 in this study, considering that complexes were formed at a lower molecular weight in graft copolymers than observed in solution or in PMAA membranes exposed to the PEG solution[8] and that the molecular weight of OEG to form a complex with PAAc was 600 or more.[16] A polymerizable end group was introduced into OEG by reacting with acryloyl chloride to prepare the OEGMA macromonomer and the OEGDA crosslinker. No AAc peaks were detected in the ^{1}H-NMR spectra of OEGMA and OEGDA. OEGMA and OEGDA were further characterized by comparing the area of the signals assigned to the ethylene protons of OEG units (δ 3.65 and 4.31 ppm) with that of the peaks assigned to the vinyl protons (δ 5.8–6.5 ppm). Table II lists apparent chain molecular weights of OEGMA and OEGDA determined by ^{1}H-NMR. It shows that they include OEG without a polymerizable end group, to some extent.

Next, the OEGMA macromonomer was copolymerized with AAc and AAm in the presence of the OEGDA crosslinker (Fig. 1). All samples were clear and homogeneous in appearance during polymerization. Figure 3 illustrates the IR spectra of AAc, AAm, and the obtained P(AAc-*co*-AAm)-*graft*-OEG gel. The absorption peaks at 1620, 1410, and 820 cm^{-1} assigned to the C═C of unreacted monomers are absent. From these results, polymerization seems to proceed completely in water.

Phase Transition of P(AAc-*co*-AAm)-*graft*-OEG Gel

Although several types of hydrogen bonding are possible in this system, hydrogen bonding between carboxyl groups in AAc residues and amide groups in AAm residues may specifically be formed.[17] Figure 4 shows the temperature dependence of the transmittance of the gels with various AAc : AAm molar ratios. It is apparent that the AAc : AAm ratio affects the phase transition

Table II Chain Molecular Weights of OEGMA and OEGDA Determined by ^{1}H-NMR

Sample	Chain Molecular Weight	Substitution Degree[a]
OEGMA	640	0.91
OEGDA	603	1.93

[a] Calculated from $4I_1\mathrm{DP}_n/3I_2$, where I_1, I_2, and DP_n are the intensities of vinyl and methylene protons and number-average degree of polymerization of OEG, respectively.

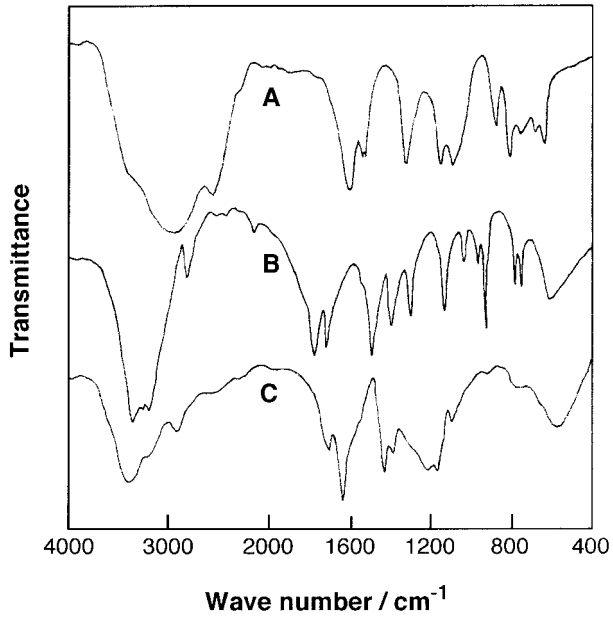

Figure 3 IR spectra of monomers and P(AAc-*co*-AAm)-*graft*-OEG gel: (A) AAc; (B) AAm; (C) P(AAc-*co*-AAm)-*graft*-OEG.

of the gels. The gels with AAc : AAm ratios of 1 : 9 and 2 : 8 are entirely transparent in the temperature range from 2 to 50°C, and for the gels with AAc : AAm ratios from 3 : 7 to 5 : 5, the phase-transition temperature increases as the AAc fraction increases; the transition temperature was taken as the midpoint of the temperature region where the transmittance change occurred. It can be seen that the transition temperatures for the gels with nearly equivalent AAc : AAm ratios (5 : 5 and 6 : 4) are the highest. Then, the transition temperature decreases again as the AAc fraction increases from 6 : 4 to 9 : 1. These results imply that strong attractive forces may exist at the 1 : 1 AAc : AAm molar ratio.

Additionally, in the synthesis of these graft gels, "template polymerization" is likely to lead to the formation of hydrogen-bonding complexes between OEG and excess AAc residues,[18] because the hydrogen bonding between PAAc and PEG is more favorable than is the self-hydrogen-bonding in PAAc.[10] Consequently, it seems that OEG chains also participate in the formation of hydrogen-bonding complexes as represented in Figure 5. This is supported by the fact that the transition temperature of the P(AAc-*co*-AAm) gel crosslinked with OEGDA is higher than that crosslinked with MBAAm as shown in Figure 6. It is not clear at this point why the latter exhibited a two-stage transition behavior.

Also, the phase transition is steeper in the cases of the gels with the AAc : AAm ratios from 5 : 5 to 8 : 2. It seems that the OEG chains grafted onto the main chain play an important role in the phase transition, since the comblike OEG chains can move rapidly due to the large mobility of terminal-grafted chains.[19] When the temperature is elevated beyond the phase-transition region, the grafted OEG chains may rapidly break the hydrogen bonds to form an extended network. In addition, dissociation of some initial complexes at higher temperature is considered to promote the dissociation of adjacent complexes,[9] because of the strong hydration forces of dissociated groups. Therefore, an abrupt dissociation of the complexes is induced by the cooperative dissociation of the complex unit at a characteristic temperature, indicative of a transition between hydrogen bonding and hydration force.

Moreover, Figure 7 depicts that the phase-transition temperature of the P(AAc-*co*-AAm)-

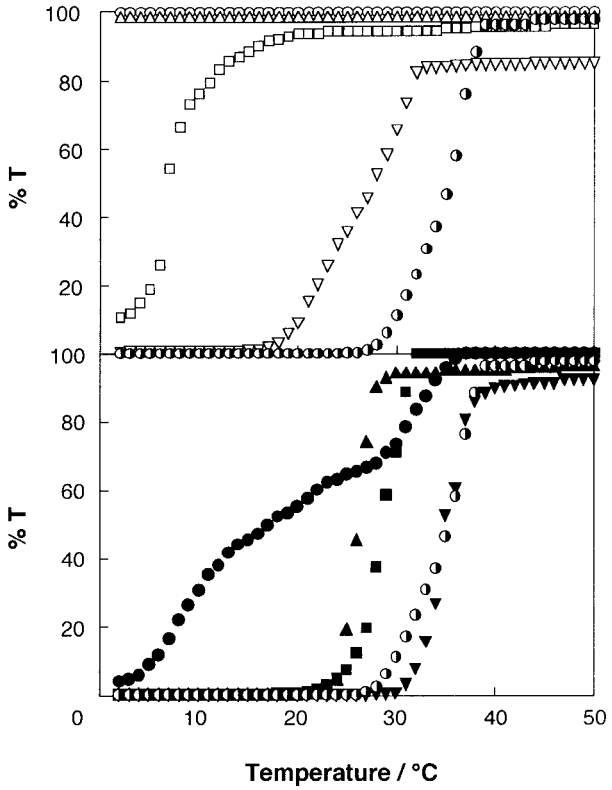

Figure 4 Dependence of optical transmittance of P(AAc-*co*-AAm)-*graft*-OEG gels on AAc : AAm ratio: (○) AAc : AAm = 1 : 9; (△) AAc : AAm = 2 : 8; (□) AAc : AAm = 3 : 7; (▽) AAc : AAm = 4 : 6; (◑) AAc : AAm = 5 : 5; (▼) AAc : AAm = 6 : 4; (■) AAc : AAm = 7 : 3; (▲) AAc : AAm = 8 : 2; (●) AAc : AAm = 9 : 1.

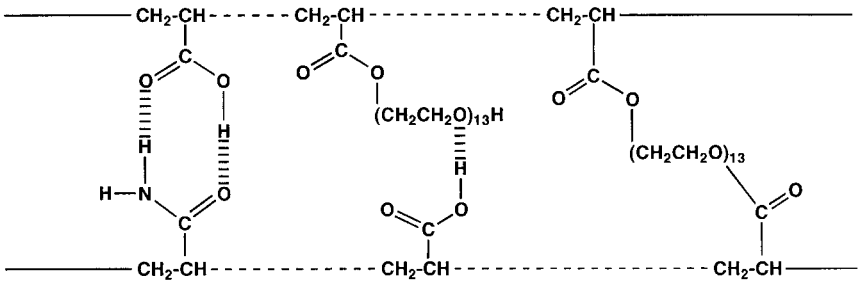

Figure 5 Schematic representation of hydrogen bonding in P(AAc-*co*-AAm)-*graft*-OEG gels.

graft-OEG gel shifts to higher temperature with increasing OEGMA content, although complexation and dissociation of the P(AAc-*co*-AAm) occurred at 35°C.[13] The gels containing 3 and 4% OEGMA were opaque over the temperature range examined. This result also indicates that the complexes are stabilized by an increase of OEGMA content in the graft copolymer. The mechanical strength of the gels, however, decreased with further increase in the OEG content; the gels containing OEGMA over 5% were too fragile to handle.

Oscillatory Phase Transition of P(AAc-*co*-AAm)-*graft*-OEG Gel

To confirm the reversibility and reproducibility of the phase transition of the P(AAc-*co*-AAm)-*graft*-OEG gel, the transmittance change of the gel with the 6 : 4 AAc : AAm molar ratio and 1 mol % OEGMA content was examined between 10 and 40°C. This temperature change was designed to cross the transition region in temperature. Transmittance changes in response to alternate temperature changes are shown in Figure 8. The lowest transmittance observed at 10°C increases when the temperature is changed to 40°C and the highest transmittance observed at 40°C decreases when the temperature is changed to 10°C, and it shows a very sharp transition shape. This reversibility, together with the mechanical strength, of our gels seems to be useful for a controlled-release device that releases incorporated drugs only at a high body temperature. Many researchers found the increased release rate of drugs through the swollen gels at lower temperature and the lower release rate of drugs through the collapsed gels at higher temperature using PNIPAAm. It should be emphasized that our system will show an increased release rate of drugs at higher temperature and a lower release rate of drugs at lower temperature.

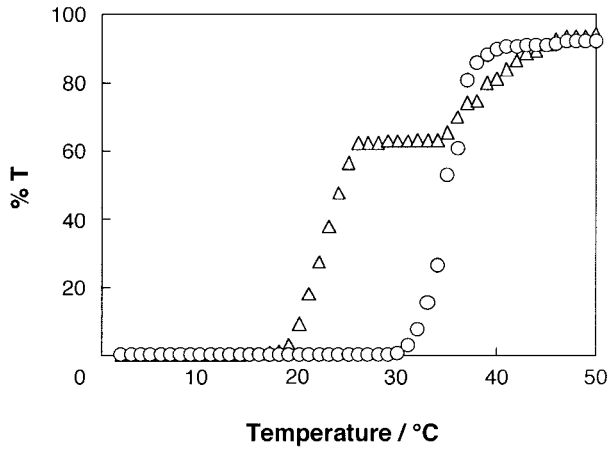

Figure 6 Dependence of optical transmittance of P(AAc-*co*-AAm)-*graft*-OEG gels on crosslinking agent: (○) OEGMA; (△) MBAAm.

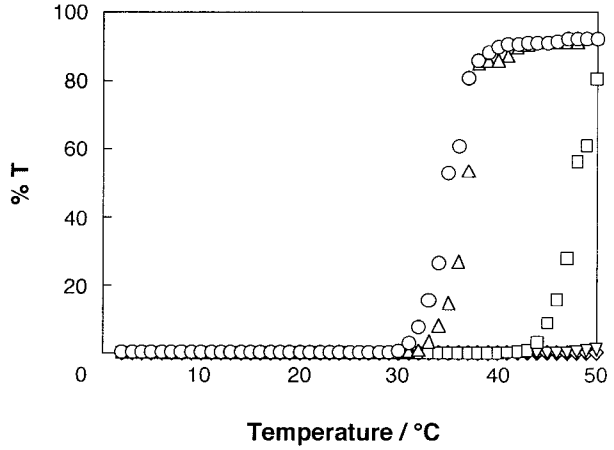

Figure 7 Dependence of optical transmittance of P(AAc-*co*-AAm)-*graft*-OEG gels on OEGMA content: (○) 0% OEGMA; (△) 1% OEGMA; (□) 2% OEGMA; (▽) 3% OEGMA; (◇) 4% OEGMA. Preparation composition was AAc : AAm = 6 : 4.

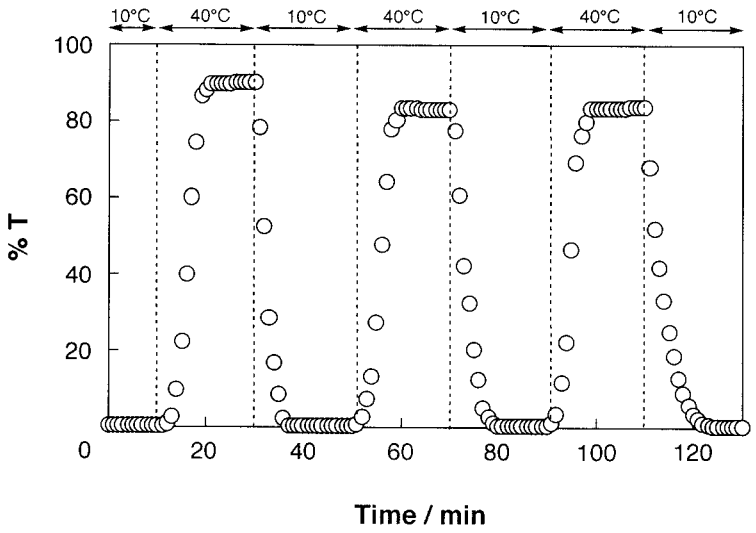

Figure 8 Oscillatory phase-transition behavior of P(AAc-*co*-AAm)-*graft*-OEG gel. Preparation composition was AAc : AAm = 6 : 4 and 1% OEGMA.

pH Dependence of Swelling of P(AAc-*co*-AAm)-*graft*-OEG Gel

Polymer systems that demonstrate a phase transition in response to more than one variable have recently been investigated. Most of them are thermosensitive and ionizable hydrogels that respond to both temperature and pH.[8,14] Figure 9 shows the effect of pH on the equilibrium swelling ratios of the P(AAc-*co*-AAm)-*graft*-OEG gel at 10°C. Because the pK_a of PAAc is 5.4, the cloud point was not observed and the gel was always clear at pH > 6. This result coincides with the fact that hydrogen-bonding complexes are stabilized by suppressing ionic dissociation of the carboxyl groups in AAc residues. At lower pH, there is sufficient protonation of the carboxylic acid groups, causing the formation of hydrogen bonding and resulting in a collapsed gel as described above. At higher pH, an increasing electrostatic repulsion between charged sites on the carboxylate groups disrupt the formation of hydrogen bonding. Hydrogen bonding acts as a driving force to shrink the gel, whereas hydrated free polymer chains increase the degree of swelling. Accordingly, changes of pH result in swelling ratios from 3 at pH 3 to 74 at pH 7.

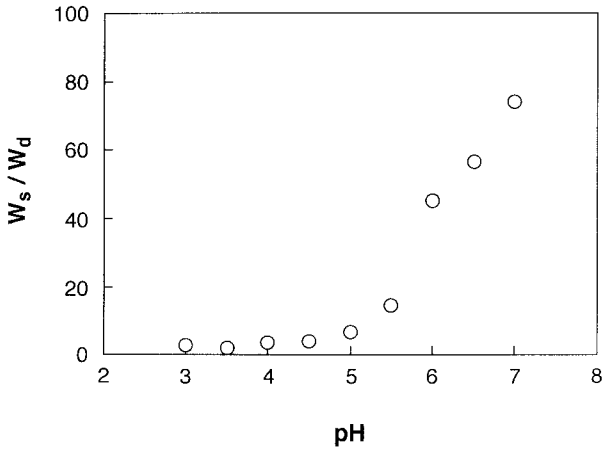

Figure 9 pH dependence of swelling ratio of P(AAc-*co*-AAm)-*graft*-OEG gel at 10°C. Preparation composition was AAc : AAm = 6 : 4 and 1% OEGMA.

In conclusion, the P(AAc-*co*-AAm)-*graft*-OEG hydrogels showed the obvious cloud points and the significant phase transition behavior at pH < 5. The phase-transition temperature was the highest at nearly equivalent molar ratios of AAc : AAm (5 : 5 and 6 : 4) and further increased with increasing OEGMA content. Therefore, the P(AAc-*co*-AAm)-*graft*-OEG hydrogels are thought to be the temperature-responsive gels that contract at lower temperature and swell at higher temperature. Such a system would be highly useful to increase the release rate of drugs at higher body temperature, not at lower body temperature.

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