

Synthesis of Water-Soluble Thermosensitive Polymers Having Phosphonium Groups from Methacryloyloxyethyl Trialkyl Phosphonium Chlorides–*N*-Isopropylacrylamide Copolymers and Their Functions

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Received 24 August 2001; accepted 9 April 2002

ABSTRACT: Water-soluble thermosensitive polymers having phosphonium groups were synthesized by the copolymerization of *N*-isopropylacrylamide (NIPAAm) with methacryloyloxyethyl trialkyl phosphonium chlorides (METRs) having varying alkyl lengths. The relative viscosities of the copolymer solutions increased with increasing content of phosphonium groups in the copolymers and decreased with increasing chain length of alkyl chains in the phosphonium groups. However, the copolymers of METR with octyl groups in phosphonium groups (METO) and NIPAAm became water insoluble with increasing contents of METO moieties in the copolymers. The transmittance at 660 nm of the copolymer solutions above the lower critical solution temperature (LCST) decreased gradually with increasing temperature and decreased with increasing chain length of alkyl chains in the phosphonium groups. The transmittance at 660 nm of

the copolymer solutions above the LCST was greatly affected by the addition of neutral salts such as KCl. The copolymers of METR with ethyl groups in phosphonium groups and NIPAAm and those of METR with butyl groups in phosphonium groups and NIPAAm had high flocculating abilities against bacterial suspensions. The METO–NIPAAm copolymer was found to have a high antibacterial activity. The flocculating ability and the antibacterial activity of the copolymers were affected by not only the content of phosphonium groups but also the alkyl chain length in the phosphonium groups in the copolymers. © 2002 Wiley Periodicals, Inc. *J Appl Polym Sci* 87: 386–393, 2003

Key words: solution properties; stimuli-sensitive polymers; water-soluble polymers

INTRODUCTION

Poly(*N*-isopropylacrylamide) (polyNIPAAm) is a thermosensitive polymer having a lower critical solution temperature (LCST) in an aqueous solution of around 32°C. Copolymers containing polyNIPAAm have been widely studied from fundamental and practical points of view.^{1–5}

On the other hand, it has been reported that water-soluble polymers having phosphonium groups had high antibacterial activity against bacteria such as *Staphylococcus aureus* (*S. aureus*) or *Escherichia coli* (*E. coli*).^{6,7} We also reported on the preparation of several water-insoluble resins or hydrogels having phosphonium groups and reported that they had antibacterial activity against bacteria such as *S. aureus* or *E. coli*.^{8–11} With these insoluble bactericides, we could prevent the residual toxicity of bactericides in water.

This article deals with the synthesis of water-soluble thermosensitive copolymers having phosphonium

groups by the copolymerization of *N*-isopropylacrylamide (NIPAAm) with methacryloyloxyethyl trialkyl phosphonium chlorides (METRs), their solution properties, and their functions.

EXPERIMENTAL

Materials

METRs with ethyl, butyl, and octyl groups in phosphonium groups were used; these monomers are abbreviated as METE, METB, and METO, respectively. METR and NIPAAm were kindly supplied by Nippon Kagaku Kogyo Co. (Tokyo, Japan) and Kohjin Co. (Tokyo, Japan), respectively, and were used without further purification. 2-2'-Azobisisobutyronitrile (AIBN; Nacalai Tesque Co. (Kyoto, Japan)) was purified by recrystallization from ethanol (Wako Pure Chemical Industries, Ltd., Osaka, Japan). Other chemical compounds were reagent grade and were used as received.

Synthesis of water-soluble copolymers

The structure of the METR–NIPAAm copolymers is shown in Figure 1.

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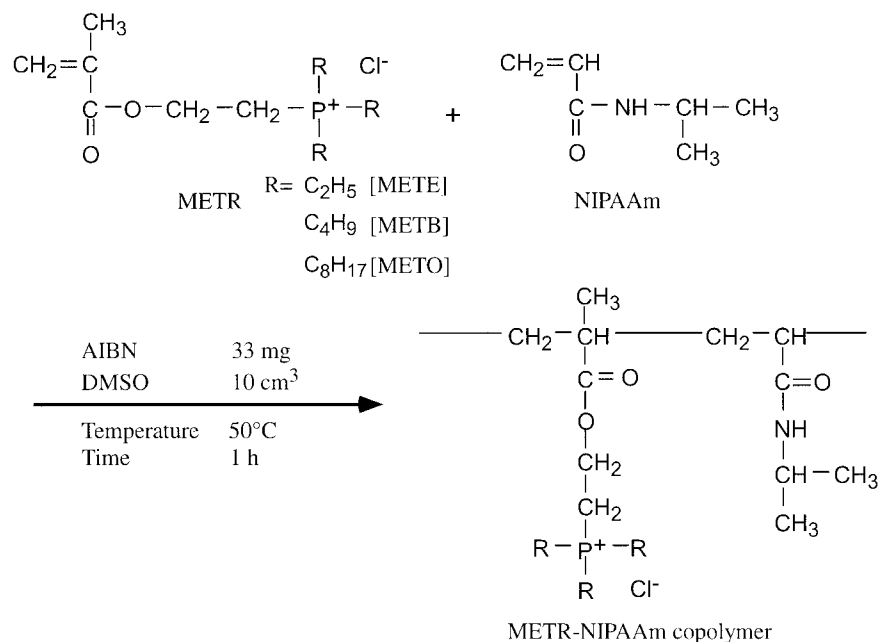


Figure 1 Synthesis of METR–NIPAAm copolymers.

First, desired amounts of METR and NIPAAm (2.263 g) were dissolved in 10 cm³ of dimethylsulfoxide (DMSO) in a glass vessel under a nitrogen atmosphere. The copolymers were obtained by radical copolymerization with AIBN (33 mg) as a radical initiator at 50°C for 1 h in sealed ampoules. After polymerization, the polymer mixture was dissolved into water. Then, the copolymers were purified by dialysis against deionized water. The copolymers were obtained by freeze drying. The phosphonium content in the copolymers was calculated from the phosphorus content of the dried copolymers. The phosphorus content was determined as follows: Dried copolymers (0.1 g) and 70% nitric acid (10 cm³) were placed in a Kjeldahl flask, and the mixture was heated until white steam came out; then, the mixture was cooled. Next, 60% perchloric acid (10 cm³) was added to this solution, and the mixture was heated until the solution became transparent. The concentration of phosphoric acid in the resulting solution was determined by the phosphovanadomolybdate method.¹²

Measurement of the molecular weight of the copolymers

The average molecular weight of the copolymers was determined by gel permeation chromatography (GPC) with polyethylene glycol and polyethylene oxide standards in 0.01 mol/dm³ LiBr dimethylformamide (DMF) solution. In this solution, the cationic charges of phosphonium groups in the copolymers were suppressed.

Measurement of the relative viscosity of the copolymer solutions

The relative viscosity of the copolymer aqueous solutions containing no salt was measured with an Ostwald viscometer (No. 1, Sansyo, Japan) at various temperatures.

Measurement of flocculating ability against bacterial suspensions

Flocculating ability was studied by observation of the optical density at 660 nm of the supernatant after the solutions were filtered with filter paper (No. 1, Advantec, Tokyo, Japan) after the copolymer solutions were mixed with about 10⁷ cells/cm³ bacterial suspensions for 1 h.

Measurement of antibacterial activity against *E. coli*

The bacterium used in this study was *E. coli* (IFO 3301), which was obtained commercially from the Institute for Fermentation (Osaka, Japan). A calculated cell suspension containing about 10⁷–10⁸ cells/cm³ was prepared and used for antibacterial tests. About 40 mg of copolymer was dissolved in 20 cm³ of sterile deionized water in a 50 cm³ Erlenmeyer flask. The copolymer solution was added into 20 cm³ of the cell suspension, and the flask was shaken at 30°C for a prescribed time. After the copolymer solutions were mixed with a bacterial suspension for a prescribed time, 1 cm³ of the bacterial suspension was pipetted from the flask, and 9 cm³ of sterile water were added

TABLE I
Content of Phosphorus in the METR–NIPAAm
Copolymers and Average Molecular Weight
of the Copolymers

Molar ratio in feed of METR:NIPAAm copolymer	P(wt%)		$M_w \times 10^{-5}$
	Calc.	Found	
METE:NIPAAm			
1:100	0.27	0.62	6.50
3:100	0.77	1.14	7.11
5:100	1.22	1.66	7.24
7:100	1.68	1.70	2.32
10:100	2.27	2.31	2.94
METB:NIPAAm			
1:100	0.27	0.64	7.19
3:100	0.75	0.83	6.68
5:100	1.19	1.23	5.80
7:100	1.60	1.77	2.89
10:100	2.14	2.18	2.70
METO:NIPAAm			
1:100	0.26	0.86	7.92
3:100	0.72	0.91	6.16
5:100	1.11	1.24	3.96

M_w = weight-average molecular weight. These values were determined by GPC with poly(ethyleneglycol) and poly(ethylene oxide) standards in 0.01 mol/dm³ LiBr DMF solution.

to the bacterial suspension. The suspension was diluted several times, and 0.1 cm³ of the diluted suspension was spread on an agar plate made of nutrient agar. The plate was kept at 30°C for 15–24 h, and the number of viable cells was calculated from those of the colonies formed on the plate.

RESULTS AND DISCUSSION

Synthesis of water-soluble copolymers

Table I shows the molar ratio of each monomer in the feed, the phosphorus content of the copolymers, and the average molecular weight of the METR–NIPAAm copolymers obtained. The phosphonium content in the copolymers was calculated from the phosphorus

content in the dried copolymers, which was determined by the phosphovanadomolybdate method. To get the thermosensitive METR–NIPAAm copolymers, we held constant the NIPAAm content (100 molar ratio), whereas the METR content was varied from 1 to 10 molar ratio to 100 molar NIPAAm.

The phosphonium content of the copolymers increased with increasing molar ratio of METR in the feed, but the observed values were larger than those calculated. The obtained METR–NIPAAm copolymers had average molecular weights of several tens of thousands. The molecular weights of the copolymers decreased with increasing METR content in feed, although the decrease in the molecular weight was not observed obviously in the METR–NIPAAm copolymers.

Temperature dependence of the viscosity of the copolymer solutions

Figure 2 shows the relative viscosity of various METR–NIPAAm copolymer aqueous solutions containing no salt as measured with an Ostwald viscometer at various temperatures.

In general, the viscosity of the polymer solutions increased with increasing molecular weight of the polymers; however, the relative viscosity of the aqueous solutions of 0.2 g/dm³ of the METE–NIPAAm and METB–NIPAAm copolymers increased with increasing phosphonium content in the copolymers irrespective of the order of their molecular weights, which were determined in a DMF solution of 0.01 mol/dm³ LiBr. This increase in the relative viscosity of the copolymers with increasing phosphonium content was brought about by the expansion of the polymer chains in water because of the repulsion of cationic charges of phosphonium groups introduced into the copolymers. It is known that the relative viscosity of the aqueous polyNIPAAm solutions decreases abruptly around 32°C of its LCST. However, the relative viscosity of the aqueous solutions of the METE–NIPAAm and METB–

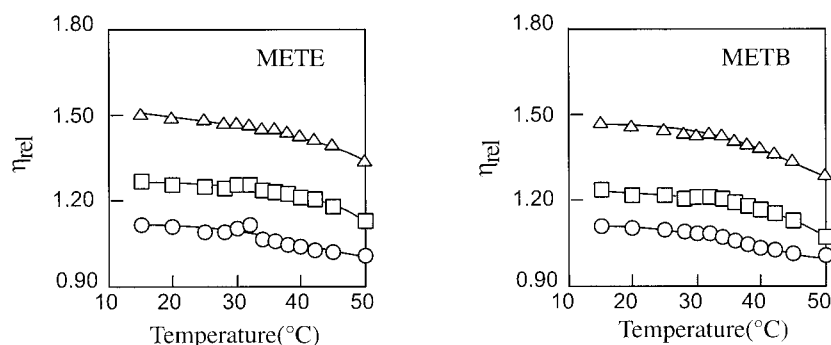


Figure 2 Temperature dependence of the relative viscosity of 0.2 g/dm³ METR–NIPAAm copolymer solutions. METR:NIPAAm (X:Y): (○) 1:100, (□) 3:100, and (△) 5:100. $\eta_{rel} = t/t_0$, where t and t_0 represent the flow time of the solution and the solvent (water), respectively.

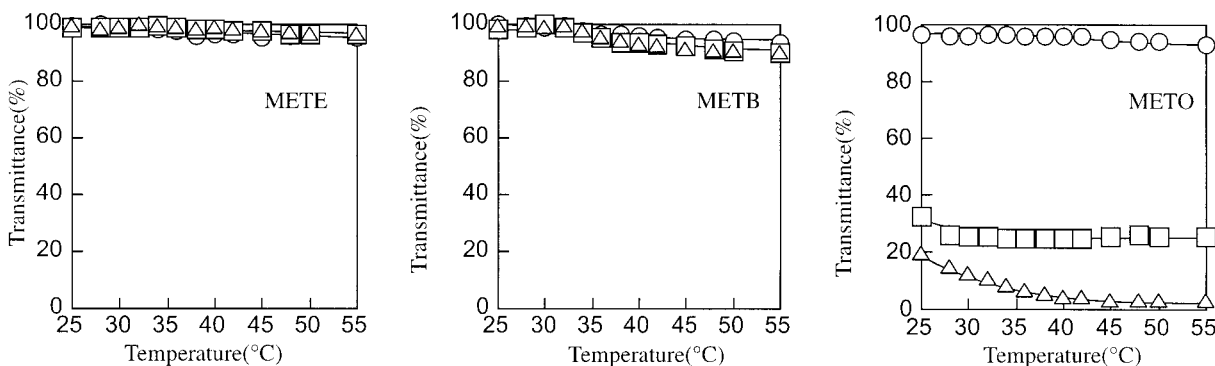


Figure 3 Changes in the transmittance at 660 nm of 2.0 g/dm³ METR-NIPAAm (X:Y) copolymer solutions at various temperatures. METR:NIPAAm (X:Y): (○) 1:100, (□) 3:100, and (△) 5:100.

NIPAAm copolymers decreased gradually with increasing temperature, and an abrupt decrease in the relative viscosity around the LCST (about 32°C) of polyNIPAAm was not observed. That is, the thermosensitivity of the METE-NIPAAm and METB-NIPAAm copolymers was considerably reduced by the introduction of phosphonium groups into the copolymers. This indicated that the association of polyNIPAAm moieties above the LCST became difficult because of the presence of phosphonium groups in the METE-NIPAAm and METB-NIPAAm copolymers.

The relative viscosity of the aqueous solutions of the METO-NIPAAm (3:100 and 5:100) copolymers could not be measured because they were insoluble in water. This was because of the higher hydrophobicity of the octyl groups in METO compared to the ethyl or butyl groups in the METE-NIPAAm and METB-NIPAAm copolymers, respectively.

Temperature dependence of the transmittance at 660 nm of the copolymer solutions

Figure 3 shows the temperature dependence of the transmittance at 660 nm of the aqueous solutions of the METR-NIPAAm (1–5) copolymers having different compositions. The transmittance of the METE-NIPAAm copolymer solutions hardly decreased with

increasing temperature, and the transmittance of the METB-NIPAAm copolymer solutions decreased slightly with increasing temperature above 30°C. These results indicate that the METE-NIPAAm and METB-NIPAAm copolymers were soluble in water in the temperature range from 25 to 55°C. The transmittance of the METO-NIPAAm (3:100 and 5:100) copolymer solutions was very low, although the transmittance of the METO-NIPAAm (1:100) copolymer solution was fairly high. This indicates that the METO-NIPAAm (3:100 and 5:100) copolymers were water insoluble even below the LCST of polyNIPAAm and that the METO-NIPAAm (1:100) copolymer was water soluble in the temperature range from 25 to 55°C. This was due to the higher hydrophobicity of octyl groups in METO compared to the ethyl or butyl groups in METE or METB, respectively. Furthermore, the transmittance of the METO-NIPAAm (5:100) copolymer solution decreased with increasing temperature, although the values were very low in the temperature range studied.

Effect of KCl addition on the transmittance of the METR-NIPAAm copolymer solutions

Figure 4 shows the transmittance at 660 nm of the METE-NIPAAm (1–5:100) copolymer solutions in the

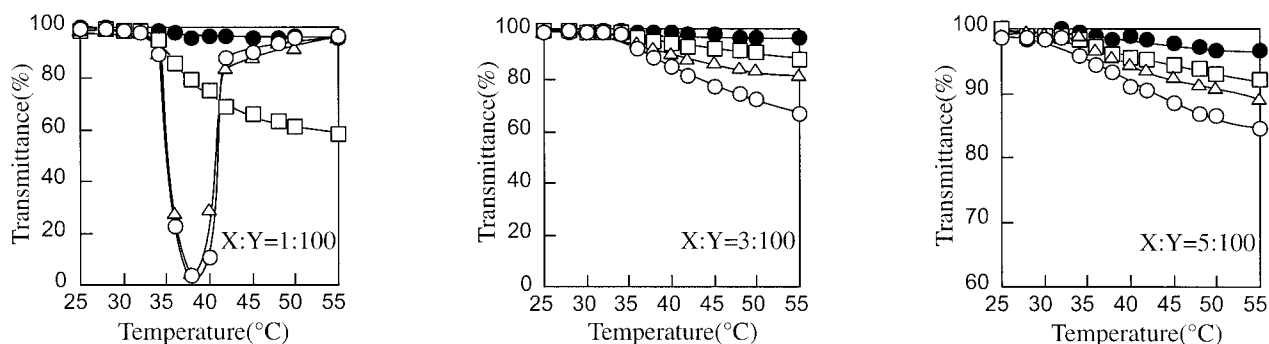


Figure 4 Effect of KCl addition on the temperature dependence of the transmittance at 660 nm of 2.0 g/dm³ METE-NIPAAm (X:Y) copolymer solutions. Concentration of KCl (mol/dm³): (●) 0, (□) 0.01, (△) 0.03, and (○) 0.05.

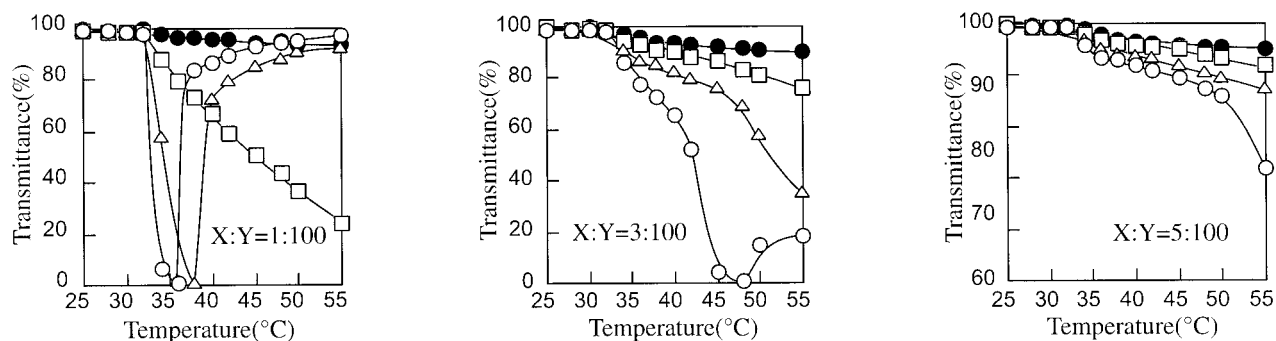


Figure 5 Effect of KCl addition on the temperature dependence of the transmittance at 660 nm of 2.0 g/dm³ METB-NIPAAm (X:Y) copolymer solutions. Concentration of KCl (mol/dm³): (●) 0, (□) 0.01, (△) 0.03, and (○) 0.05.

absence or the presence of KCl (0.01–0.05 mol/dm³). In the case of the METE-NIPAAm (1:100) copolymer, the transmittance of the copolymer solution in the absence of KCl hardly decreased with increasing temperature, but the transmittance of the copolymer solution in the presence of KCl (0.01 mol/dm³) decreased with increasing temperature above 32°C. The transmittance of the copolymer solution in the presence of KCl (>0.03 mol/dm³) decreased abruptly with increasing temperature from 32 to 36°C; then, it increased again abruptly with increasing temperature up to 46°C. In this case, precipitates of the copolymers were observed above 36°C. Therefore, the abrupt rapid decrease in the transmittance up to 36°C was due to the association of the copolymers by the hydrophobic interaction of polyNIPAAm moieties in the copolymers, and the abrupt increase in the transmittance above 36°C was due to the formation of precipitates of the copolymers. In the case of the METE-NIPAAm (3:100 and 5:100) copolymers, the transmittance of the copolymer solutions gradually decreased with increasing temperature above 35°C, and the degree of decrease in the transmittance increased with increasing KCl concentration in the solutions. The decrease in the transmittance with increasing KCl concentration in the solutions means that the solubility of the copolymers in water was reduced by the addition of KCl.

Figure 5 shows the transmittance at 660 nm of the METB-NIPAAm (1–5:100) copolymer solutions in the absence and in the presence of KCl (0.01–0.05 mol/dm³). The temperature dependence of the transmittance of the METB-NIPAAm (1:100) copolymer solutions was almost similar to that of the METE-NIPAAm (1:100) copolymer solutions, although the temperature at which the transmittance of the METB-NIPAAm (1:100) copolymer solution in the presence of KCl (>0.03 mol/dm³) decreased abruptly shifted slightly to a lower temperature. However, the temperature dependence of the transmittance of the METB-NIPAAm (3:100) copolymer solution was fairly different from that of the METE-NIPAAm (3:100) copoly-

mer solution. That is, the transmittance of METB-NIPAAm (3:100) copolymer solution in the presence of KCl (0.03 mol/dm³) decreased more greatly with increasing temperature from 32 to 55°C than that of the METE-NIPAAm (3:100) copolymer solution. Also, the transmittance of the METB-NIPAAm (3:100) copolymer solution in the presence of KCl (0.05 mol/dm³) decreased sharply with increasing temperature from 30 to 48°C and then increased with increasing temperature. These phenomena were due to the higher hydrophobicity of METB than of METE. The temperature dependence of the transmittance of the METB-NIPAAm (5:100) copolymer solutions was almost similar to that of the METE-NIPAAm (5:100) copolymer solutions.

Figure 6 shows the transmittance at 660 nm of the METO-NIPAAm (1–5:100) copolymer solutions in the absence or the presence of KCl (0.01–0.05 mol/dm³). The transmittance of the METO-NIPAAm (1:100) copolymer solutions in the absence of KCl was almost above 98%. This indicates that METO-NIPAAm (1:100) copolymer was soluble in water in the temperature range from 25 to 55°C. However, the transmittance in the presence of KCl (0.01–0.05 mol/dm³) decreased abruptly with increasing temperature from 25 to 33°C. Above 35°C, the transmittance increased again with increasing temperature. As mentioned before, this abrupt decrease in the transmittance up to 33°C was due to the association of the copolymers by the hydrophobic interaction of polyNIPAAm moieties in the copolymers, and the abrupt increase in the transmittance above 35°C was due to the formation of precipitates of copolymers. In the case of the METO-NIPAAm (3:100) copolymer, the transmittance of the copolymer solution in the absence of KCl was below 75% because this copolymer became difficult to dissolve in water. The transmittance of the METO-NIPAAm (3:100) copolymer solution in the presence of KCl (0.03 mol/dm³) became lower compared to that in the absence of KCl. This phenomenon indicates that the METO-NIPAAm (3:100) copolymer became more difficult to dissolve in water with the addition of KCl.

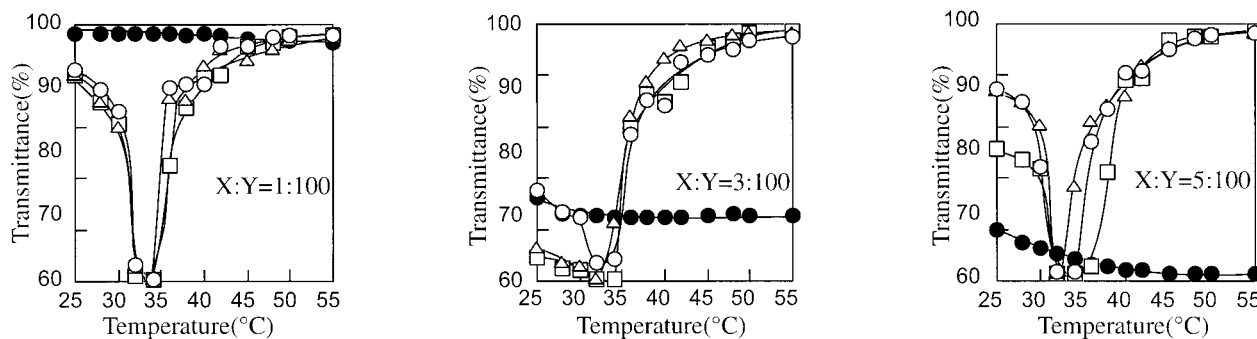


Figure 6 Effect of KCl addition on the temperature dependence of the transmittance at 660 nm of 2.0 g/dm³ METO-NIPAAm (X:Y) copolymer solutions. Concentration of KCl (mol/dm³): (●) 0, (□) 0.01, (△) 0.03, and (○) 0.05.

However, the transmittance of the METO-NIPAAm (3:100) copolymer solution in the presence of KCl (0.05 mol/dm³) increased again. This was due to the fact that the suspended copolymers were partially precipitated by the addition of KCl. Similar phenomena were also observed in the case of the METO-NIPAAm (5:100) copolymer in the presence of KCl. The METO-NIPAAm (5:100) copolymer was more difficult to dissolve in water than the METO-NIPAAm (3:100) copolymer because the former contained more METO moieties than the latter. The transmittance of the METO-NIPAAm (5:100) copolymer solution at 25°C increased as KCl was added up to 0.03 mol/dm³. We observed a small amount of the precipitates of the copolymer in the solutions after the addition of KCl (>0.03 mol/dm³). This resulted in an increase in the transmittance due to the decrease in the suspended copolymers in the supernatant. The transmittance of the METO-NIPAAm (5:100) copolymer solution in the presence of KCl decreased abruptly with increasing temperature up to 35°C and then increased with increasing temperature. The decrease in the transmittance up to 35°C means that the association of the suspended copolymers by the hydrophobic interaction of polyNIPAAm moieties increased with increasing temperature from 25 to 35°C. The increase in the transmittance above 35°C was due to the formation of the precipitates of the copolymers.

In Figure 7, the temperature dependence of the transmittance of various METR-NIPAAm (3:100) copolymer solutions in the presence of KCl (0.01 mol/dm³) is compared. The effect of the addition of KCl on the transmittance the METR-NIPAAm (3:100) copolymer solutions was in the following order:

METO-NIPAAm > METB-NIPAAm
> METE-NIPAAm copolymer

This means that longer alkyl chain in the phosphonium groups affected more strongly not only the solubility of the METR-NIPAAm copolymers in water but also the

hydrophobic interaction between polyNIPAAm moieties in the copolymers in the KCl solutions.

Effect of sodium benzenesulfonate (SBS) or sodium *n*-dodecylbenzenesulfonate (SDBS) addition on the transmittance of the METR-NIPAAm copolymer solutions

Figure 8 shows the transmittance at 660 nm of the METR-NIPAAm (3:100) copolymer solutions in the absence or the presence of SBS or SDBS (2 mmol/dm³). The temperature dependence of the transmittance in deionized water containing no SBS or SDBS is shown in Figure 8(a). The transmittance of the METE-NIPAAm and METB-NIPAAm copolymer solutions decreased slightly with increasing temperature above 30°C, and it was above 90% in the temperature range studied. This result indicates that these copolymers were water soluble. The transmittance of the METO-NIPAAm copolymer solution was fairly low because the copolymer was insoluble in water. The transmittance of the METO-NIPAAm copolymer solution did not decrease with increasing temperature above 30°C.

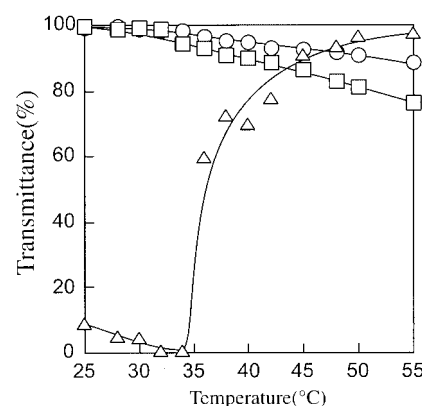


Figure 7 Changes in the transmittance at 660 nm of 2.0 g/dm³ various METR-NIPAAm (3:100) copolymer KCl (0.01 mol/dm³) solutions. Copolymers: (○) METE-NIPAAm, (□) METB-NIPAAm, and (△) METO-NIPAAm.

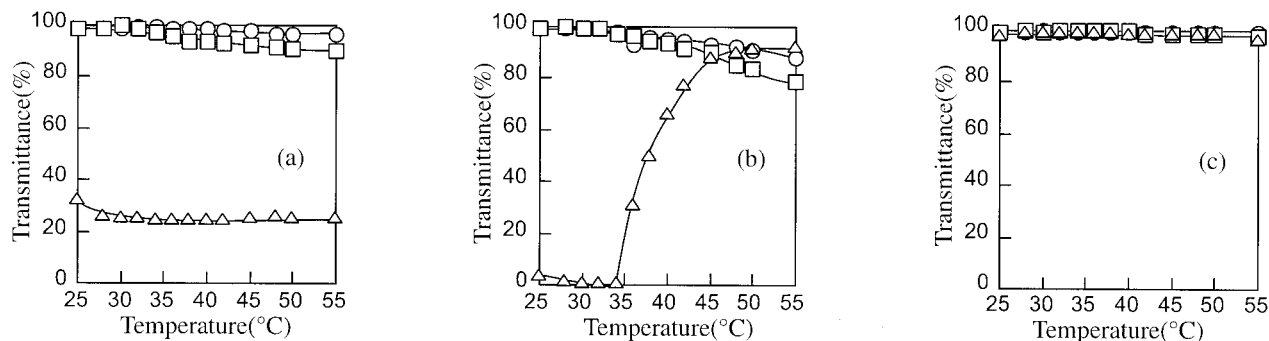


Figure 8 Effect of SBS or SDBS (2 mmol/dm^3) on the transmittance at 660 nm of 2.0 g/dm^3 METR-NIPAAm (3:100) copolymer solutions at various temperatures: (a) in the absence of SBS or SDBS, (b) in the presence of SBS, and (c) in the presence of SDBS. Copolymers: (○) METE-NIPAAm, (□) METB-NIPAAm, and (△) METO-NIPAAm.

Figure 8(b) shows the temperature dependence in the presence of SBS (2 mmol/dm^3). In the presence of SBS, the temperature dependence of the transmittance of the METE-NIPAAm and METB-NIPAAm copolymer solutions became a little larger than that in the absence of SBS or SDBS. However, the transmittance of the METO-NIPAAm copolymer solution in the presence of SBS decreased to almost zero below 35°C , and it increased abruptly above 35°C . This means that the addition of SBS accelerated the association of the METO-NIPAAm copolymers due to the hydrophobic interaction of the polyNIPAAm moieties in water below 35°C and the formation of the precipitates of the copolymers above 35°C .

In the presence of SDBS (2 mmol/dm^3), the transmittance of all the METR-NIPAAm copolymer solutions became almost 100% in the temperature range studied [Fig. 8(c)]. That is, the temperature dependence of the transmittance of the METR-NIPAAm copolymer solutions disappeared by the addition of SDBS. This indicates that SDBS made all the METR-NIPAAm copolymers more water soluble. This was due to the adsorption of SDBS on the METR-NIPAAm copolymers by the hydrophobic interaction between dodecyl groups of SDBS and the hydrophobic moieties in polyNIPAAm.

Flocculation of bacteria with the METR-NIPAAm copolymers

The flocculating ability of the METR-NIPAAm (1–5:100) copolymers against bacteria (*E. coli*) suspensions was evaluated by measurement of the optical density at 660 nm of the supernatant after the copolymers were mixed with a bacterial suspension (Fig. 9). In the case of the METE-NIPAAm and METB-NIPAAm copolymers, the optical density of the bacterial suspension decreased with increasing amounts of copolymers added and then increased. The concentration of copolymers added at which the lowest optical density was obtained was defined as the optimum dosage of the copolymer for the flocculation of the bacterial suspensions. This means that the flocculating ability of the copolymers against the *E. coli* suspension increased with increasing amount of copolymers and then decreased. It is known that most bacteria such as *E. coli* or *S. aureus* have negative charges on the surfaces of the bacteria in water. Therefore, this result means that the flocculation of bacteria was mainly brought about by the neutralization of anionic charges on the surface of the bacteria with cationic copolymers, followed by the bridging of the bacteria. Furthermore, the optical density at the optimum dosage

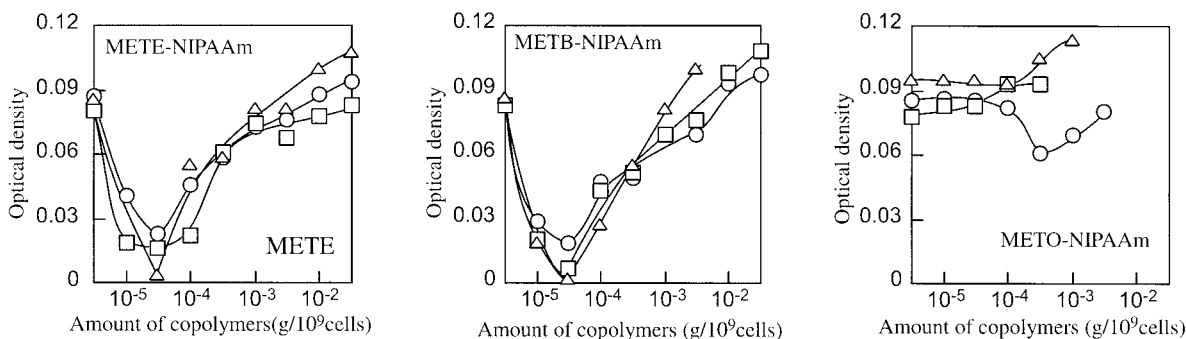


Figure 9 Changes in the optical density of an *E. coli* suspension (10 cm^3 in deionized water) at 660 nm after contact with various copolymer solutions at 30°C (shaking rate = 220 min^{-1} , shaking time = 1 h). METR:NIPAAm (X:Y): (○) 1:100, (□) 3:100, and (△) 5:100.

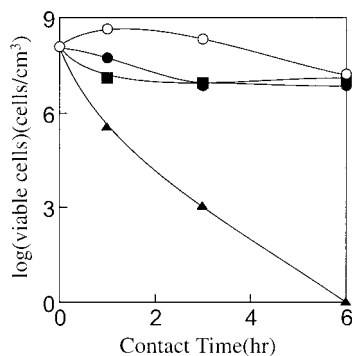


Figure 10 Changes in viable cell numbers (40 cm^3 of *E. coli* suspended in deionized water) after contact with 0.04 g of METR:NIPAAm (3:100) copolymers at 30°C . Copolymers: (○) blank, (●) METE-NIPAAm, (■) METB-NIPAAm, and (▲) METO-NIPAAm.

decreased with increasing content of phosphonium groups in the METE-NIPAAm and METB-NIPAAm copolymers. The remarkable increase in the optical density above the optimum dosage of the METE-NIPAAm and METB-NIPAAm (1–5) copolymers was due to the dispersion of the flocculated bacteria because of an excess adsorption of cationic polymers on the surface of bacteria. The METO-NIPAAm (3:100 and 5:100) copolymers hardly exhibited the flocculating ability against *E. coli* suspension, and only the METO-NIPAAm (1:100) copolymer exhibited very low flocculating ability at a higher optimum dosage than that in the case of the METE-NIPAAm and METB-NIPAAm copolymers. This was due to the fact that the METO-NIPAAm (3:100 and 5:100) copolymers were water insoluble, but the METO-NIPAAm (1:100) copolymer was water soluble, as mentioned before.

Antibacterial activity of the copolymers against *E. coli*

The antibacterial activity of the METR-NIPAAm (3:100) copolymers against *E. coli* was investigated. We evaluated the antibacterial activity by counting the numbers of viable cells after contact with the copolymers (Fig. 10). The METE-NIPAAm and METB-NIPAAm copolymers exhibited low antibacterial activity against *E. coli*. However, the METO-NIPAAm copolymer exhibited high antibacterial activity, although the METO-NIPAAm copolymer was not completely water soluble. Many researchers have reported that antibacterial activity was not affected only by electrostatic interaction but by hydrophobic interaction between bactericides and bacteria.^{6–8} We also

found that not only phosphonium groups but also octyl groups in the METR-NIPAAm copolymers greatly affected the antibacterial activity against *E. coli*.

CONCLUSIONS

1. Water-soluble thermosensitive polymers having phosphonium groups were prepared by the copolymerization of METR with NIPAAm
2. The relative viscosity of the METR-NIPAAm (1–5:100) copolymer solutions increased with increasing METR content and decreased gradually with increasing temperature.
3. The thermosensitivity of the METR-NIPAAm copolymers was greatly affected not only by the addition of neutral salts but also by the hydrophobicity of alkyl groups in of METR introduced into the copolymers.
4. The METE-NIPAAm and METB-NIPAAm copolymers had high flocculating ability against the *E. coli* suspension.
5. The METO-NIPAAm (3:100) copolymer had high antibacterial activity against *E. coli*, but the METE-NIPAAm and METB-NIPAAm copolymers had very low antibacterial activity.

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