

Photoresponsive Permeability of Polyelectrolyte Complex Capsule Membrane Containing Triphenylmethane Leucohydroxide Residues

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SYNOPSIS

To develop a functional capsule, which can be controlled to release materials entrapped in the inner aqueous space responding to light, partly crosslinked poly(acrylic acid)-polyethylenimine complex capsules containing a copolymer of acrylic acid and bis[4-(dimethylamino)phenyl](4-vinylphenyl)methyl leucohydroxide as an additional membrane component with radii of ca. 3 μm have been prepared. The triphenylmethane derivative on the copolymer dissociated into an ion pair under ultraviolet light irradiation in the weak alkaline region. Permeation of *p*-toluenesulfonate through the capsule membrane was enhanced significantly by the photoirradiation after a several minutes time lag under a weak alkaline condition. The photoinduced increase in permeability of the capsule was pH-dependent. The maximum effect of light on the permeability was observed near pH 8, whereas less enhancement was observed above or below the pH value. When the irradiation of the light was stopped, permeation of the permeant decreased. The photoresponsive permeability change of the capsule membrane can be carried out reversibly. © 1995 John Wiley & Sons, Inc.

INTRODUCTION

In recent years, stimuli-responsive systems for ideal drug delivery have been actively investigated.^{1,2} Microcapsules are suitable as such drug-delivery systems, because they entrap a variety of drugs in their large inner spaces and release them at a suitable rate. Moreover, since release of drugs from microcapsules is controlled by their thin membranes, if structure and property of the membranes are changed by applying various stimuli, quick response of the release to the stimuli can be expected, compared with other systems such as polymer gel.

In spite of a number of advantages of microcapsules, a few approaches have been made to prepare stimuli-responsive microcapsules because the availability of materials for microcapsule membranes is restricted by the demands imposed by the

preparation method. Most of responsive microcapsules so far reported were prepared by modification of semipermeable microcapsule membranes with lipids³⁻⁵ and/or polymers.⁶⁻⁸

As another technique to prepare stimuli-responsive microcapsules, use of a polyelectrolyte complex has been attempted.⁹⁻¹¹ It has been shown that permeability of a polyelectrolyte complex capsule membrane consisting of poly(acrylic acid) or poly(methacrylic acid) and polyethylenimine changes responding to environmental pH and ionic strength.^{9,11} In addition, because introduction of molecules or groups with various functionalities such as enzymes into polyelectrolyte complex capsule membranes is possible by using a functional molecule-attached polyelectrolyte as a capsule membrane component, polyelectrolyte complex capsules with various functionalities can be made.

In this report, to obtain a functional capsule whose membrane permeability changes in response to light, poly(acrylic acid)-polyethylenimine complex capsules containing triphenylmethane leuco-

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hydroxide residues, which are well known to dissociate into ionic pairs under ultraviolet irradiation and thermally recombine with the counterion,¹² were prepared. The effect of ultraviolet irradiation on the release property of the capsules was also investigated.

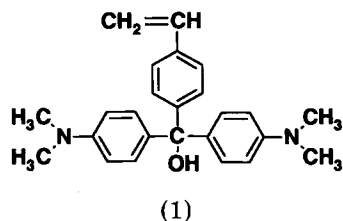
EXPERIMENTAL

Chemicals

Acrylic acid and poly(sodium acrylate) (molecular weight 44,000) was purchased from Kishida Chemical Co. (Japan). Polyethylenimine (branched type having primary, secondary, and tertiary nitrogens in the ratio of 1 : 2 : 1; molecular weight 40,000–50,000), phenylethylene glycol, and *p*-toluenesulfonic acid were supplied by Tokyo Kasei Kogyo Co. (Japan). 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC) was purchased from Nacalai Tesque Co. (Japan). Acrylic acid and phenylethylene glycol were purified by distillation and recrystallization from ligroin, respectively. Other chemicals were used without further purification.

Synthesis of Poly(acrylic acid) Having Triphenylmethane Leucohydroxide Residues

Bis[4-(dimethylamino)phenyl](4-vinylphenyl)methyl leucohydroxide (**1**) was synthesized according to the method reported by Manecke and Kosmehl^{13,14}:



A copolymer of the vinyl derivative of leucohydroxide and acrylic acid was prepared by radical copolymerization. Acrylic acid (1.73 g), the vinyl derivative of leucohydroxide (0.27 g), and 2,2'-azobisisobutyronitrile (0.10 g) were mixed and heated to 80°C in an atmosphere of argon for 4 days. The copolymer was dialyzed for 7 days against water. The content of the triphenylmethane leuco derivative in the copolymer was determined to be 2.83 unit mol % by elemental analysis. The molecular weight of the copolymer was estimated to be more than 300,000, taking poly(ethylene glycol) as the standard by high-

performance liquid chromatography analysis on an Asahipak GS-510 column (Asahi Chemical Industry, Japan) using water.

Preparation of Polyelectrolyte Complex Capsule

Polyelectrolyte complex capsules were prepared according to the method described in previous articles.^{9,11} An aqueous solution of a given pH containing poly(acrylic acid) and the copolymer was added dropwise from a pipette into an aqueous 0.5 wt % polyethylenimine solution of a given pH. The solution was stirred gently for 2 h at 30°C. The resultant capsules were washed several times with distilled water and then were partly crosslinked by incubating in 59 mM aqueous EDC solution containing 50 mM phosphate (pH 4.4) for 20 h at 30°C. The partly crosslinked capsules were kept in distilled water and washed repeatedly in order to remove the EDC remaining in the aqueous phase of the capsules.

Permeation Measurements

p-Toluenesulfonate and phenylethylene glycol were used as permeants. The permeants were loaded into the polyelectrolyte complex capsules by putting the capsules in an aqueous solution of each permeant (3.85 mM) containing 5 mM tris(hydroxymethyl)aminomethane at pH 7.5 for several days.

One permeant-loaded capsule was preincubated in the permeant solution adjusted at the pH of the measurement for more than 24 h. The capsule was taken out of the solution and then was put into 3 mL of a 5 mM tris(hydroxymethyl)aminomethane solution of a given pH at 30°C with stirring. Permeation of *p*-toluenesulfonate and phenylethylene glycol through the capsule membrane was detected by following the absorbance of the outer aqueous phase at 221.5 and 210 nm, respectively. The permeability constant P (cm s⁻¹) was determined using eq. (1) obtained from Fick's first law of diffusion⁹:

$$\ln[(C^f - C^i)/(C^t - C^i)] = -(V + V_c)APt/VV_c \quad (1)$$

where t represents the time since the start of the experiment. C^i , C^t , and C^f are the initial, intermediate, and final concentrations in volume V of the outer aqueous phase, respectively. V_c and A are the volume and surface area of one capsule, respectively.

Other Methods

Irradiation of the ultraviolet light (250 nm < λ < 380 nm) was carried out with a 500 W ultrahigh-pressure

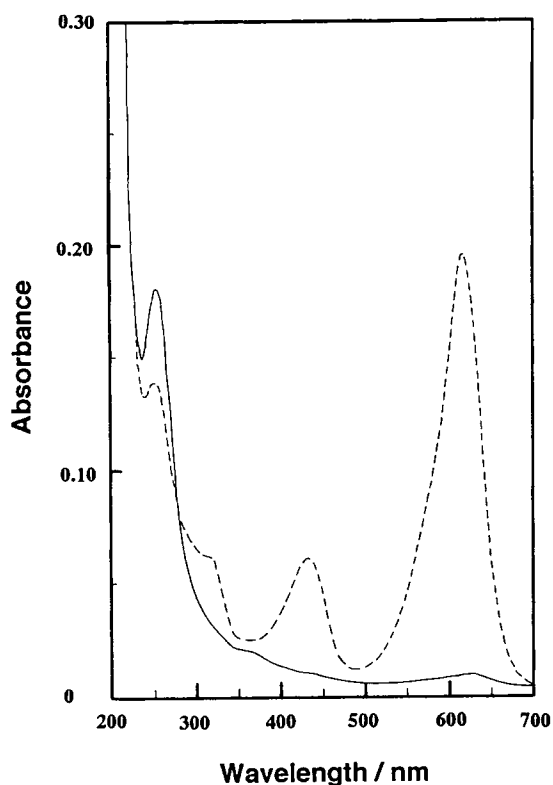


Figure 1 Absorption spectra of an aqueous solution of the copolymer at 30°C at pH 11.0 (—) in the dark and (---) after 10 min irradiation. The concentration of the polymer was 0.625 unit *M*.

mercury lamp (Ushio USH-500D), using a glass filter (Toshiba UV-D33S2). The diameter of the capsule was estimated from the weight of the capsule as reported elsewhere.⁹

RESULTS AND DISCUSSION

Photoinduced Ionization of Copolymer of Bis[4-(dimethylamino)phenyl](4-vinylphenyl)methyl Leucohydroxide and Acrylic Acid

Figure 1 shows the effect of the ultraviolet light irradiation on the absorption spectra of the copolymer in an aqueous solution at pH 11.0. The decrease in absorbance at 266 nm and increase in absorbance at 310, 432, and 620 nm after irradiation indicate the generation of triphenylmethyl cations in the copolymer. Absorbance at 310, 432, and 620 nm increased with decreasing pH in the pH region between 6 and 11 in the dark. Triphenylmethane leucohydroxide usually dissociates in the weak acidic region under the dark condition,¹⁵ whereas dissociation of this group in the copolymer occurs in the weak al-

kaline region, presumably because carboxylate anions on the polymer facilitate dissociation of triphenylmethane leucohydroxide. This is supported by the result shown in Figure 2, which represents the effect of NaCl concentration on the absorption spectra of the copolymer in the dark. As is seen in Figure 2, absorbance of the copolymer at 310, 432, and 620 nm in the dark decreases and absorbance at 266 nm increases with increasing concentrations of NaCl. Because of the shielding effect of NaCl, the ability of carboxylate anions to promote ionization of triphenylmethane leucohydroxide residues decreases, resulting in a decrease of triphenylmethyl cations on the copolymer.

Since the amount of undissociating triphenylmethane leucohydroxide residue on the copolymer altered depending on pH, photoinduced generation of the triphenylmethyl cation was investigated at various pH's. Figure 3 represents the pH dependence of the increase in the absorbance of the copolymer at 620 nm after the light irradiation. Efficiency of the light irradiation-induced dissociation of triphenylmethane leucohydroxide residue shows a maximum at pH 11.0 in the absence of polyethylenimine. Because most of the triphenylmethane leucohydroxide

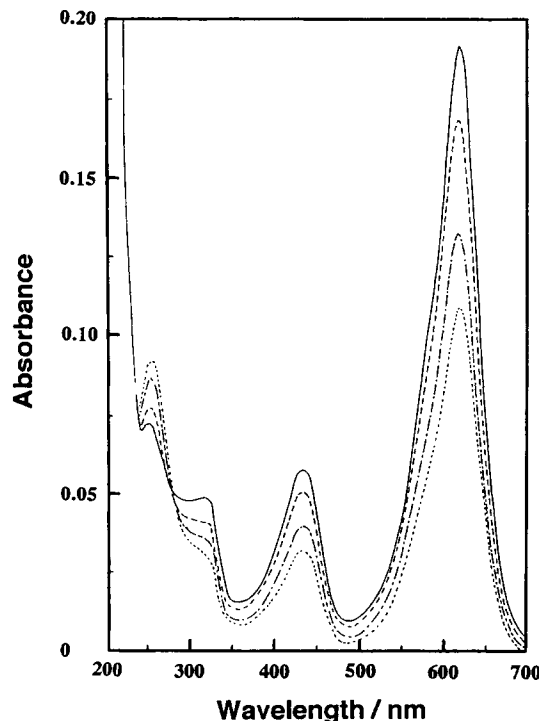


Figure 2 Absorption spectra of an aqueous solution of the copolymer in the dark at various concentrations of NaCl: (—) 0*M*; (---) 10 *mM*; (-·-) 0.1*M*; (····) 1*M*. The concentration of the copolymer was 0.625 unit *M*. The measurement was carried out at 30°C at pH 7.5.

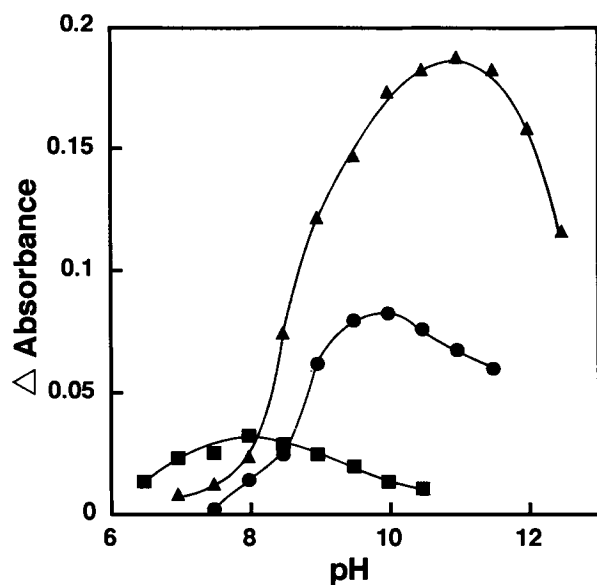


Figure 3 pH-dependence of increase in absorbance of an aqueous solution of the copolymer at 620 nm at various concentrations of polyethylenimine: (▲) 0 unit *M*; (●) 0.605 unit *M*; (■) 1.210 unit *M*. The concentration of the copolymer was 0.625 unit *M*. The measurement was carried out at 30°C. pH of the solution was adjusted by addition of small amount of NaOH solution.

residues already dissociate in the dark under the weak alkaline condition, further dissociation of these residues hardly occurs. On the contrary, since large amounts of hydroxide ions are present above pH 11, dissociation of triphenylmethane leucohydroxide residues is depressed with increasing pH.

On the other hand, the pH region where photoinduced generation of the cation occurs decreased in the presence of polyethylenimine. It is likely that in the presence of the polycation carboxylate ions on the copolymer bind to the polycation and, hence, triphenylmethyl cations recombine with hydroxide ions in the dark. Thus, efficient production of the triphenylmethyl cation occurred in the lower pH region, where fewer hydroxide ions are present. It is also seen in Figure 3 that the light irradiation-induced increase in the absorbance of the copolymer at 620 nm at the optimum pH decreases in the presence of polyethylenimine. In the presence of polyethylenimine, the solution of the copolymer is turbid and, hence, a heterogeneous system due to the complex formation between the copolymer and polyethylenimine. Therefore, the apparent change in absorbance might decrease in the presence of the polycation.

The time course of the absorbance of the copolymer at 620 nm under light irradiation and in the

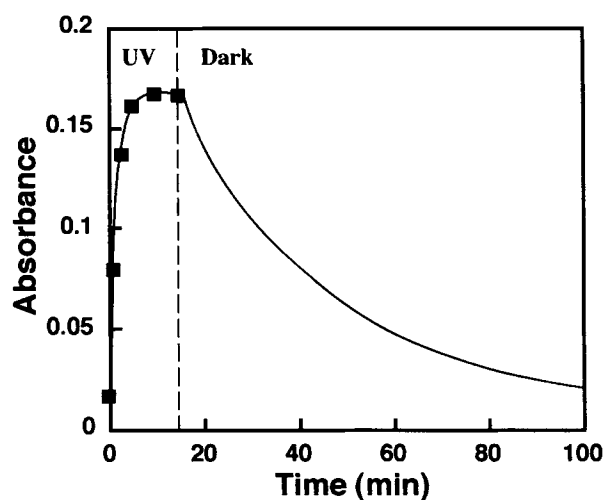


Figure 4 Absorbance of the copolymer in 5 mM carbonate-buffered solution at 620 nm under irradiation and in the dark as a function of time. The measurement was carried out at pH 10.3 at 30°C.

dark is illustrated in Figure 4. Absorbance of the copolymer increased immediately upon light irradiation and reached to a constant value within 4 min. When the light irradiation was stopped, the absorbance of the copolymer decreased gradually. These results indicate that the generation and disappearance of the triphenylmethyl cations is a time-consuming process.

Photoresponsive Permeability of the Capsule

Three kinds of polyelectrolyte complex capsules containing photosensitive groups, CP-1, CP-2, and CP-3, were prepared. The preparation conditions of these capsules are listed in Table I. The radii of CP-1, CP-2, and CP-3 were 3.40 ± 0.35 , 3.18 ± 0.45 , and 3.38 ± 0.38 nm, respectively. Residual polyanions which did not participate in the membrane formation in the inner aqueous phase of the capsules were

Table I Conditions of Capsule Preparation

Capsule	Polyanion Solution			Polycation Solution	
	Copolymer (wt %)	PAA ^a (wt %)	pH	PEI ^b (wt %)	pH
CP-1	0.3	2.7	11	0.5	5
CP-2	0.6	2.4	11	0.5	5
CP-3	0.6	2.4	11	0.5	7

^a PAA represents poly(acrylic acid).

^b PEI represents polyethylenimine.

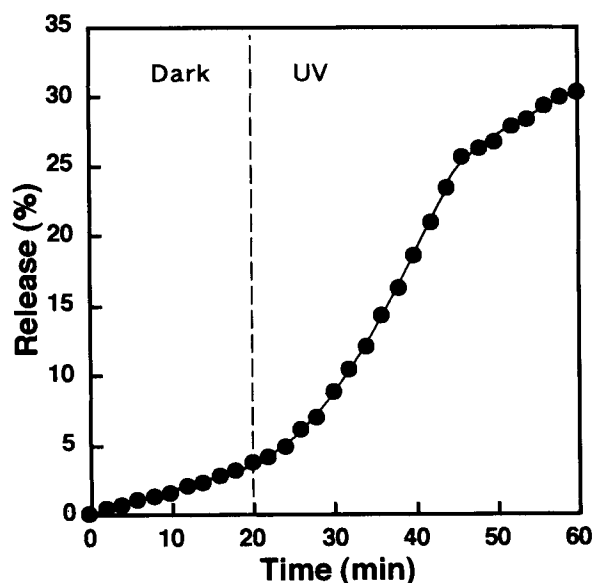


Figure 5 Release profile of *p*-toluenesulfonate from CP-1 in the dark and under irradiation at pH 8.0 at 30°C.

not detected by the HPLC analysis, indicating that the concentration of the polymers inside the capsule was less than 3×10^{-4} wt %.

Figure 5 depicts permeation profiles of *p*-toluenesulfonate through the CP-1 membrane at pH 8.0. The permeation of the permeant across the capsule membrane was enhanced upon light irradiation. The permeation is very slightly increased immediately after initiation of the light irradiation. However, the enhancement of permeation becomes more remarkable with the lapse of time. The permeation rate reached the maximum value after ca. 10 min. This time-dependent increase in the permeation rate is probably due to the time for the generation of triphenylmethyl cations as shown in Figure 4. It is apparent in Figure 5 that, while the permeation at the maximum rate is kept for about 15 min, the subsequent decrease in the permeation rate is observed. Since absorbance of the copolymer at 620 nm diminished gradually upon light irradiation when the irradiation time exceeded 30 min (results not shown), the long-time irradiation might induce decomposition of the photosensitive groups on the copolymer. Thus, it is likely that this decrease of the permeation rate after long-time irradiation is due to a decrease of photoinduced cations in the capsule membrane, resulting from decomposition of the photosensitive groups. Permeation through the capsule membrane not having photosensitive groups was also investigated and was not changed by the photoirradiation (result not shown).

Because photoinduced dissociation of the triphenylmethane leucohydroxide residues is pH-dependent as shown in Figure 3, enhancement of the permeation of *p*-toluenesulfonate through the capsule membrane upon light irradiation was investigated at various pH's. Figure 6 represents the permeability constant of *p*-toluenesulfonate across the capsule membrane in the dark and under light irradiation. Since the permeability constant under light irradiation changed with the time of the irradiation, the maximum value in the stationary state, which was observed after several minutes from the initiation of irradiation, is shown in Figure 6. No or only weak enhancement of the permeation upon light irradiation is observed in the neutral pH region. However, the photoinduced acceleration of the permeation becomes more remarkable with increasing pH and the maximum effect of the irradiation is seen between pH 8 and 8.5. In this pH region, the permeability constants upon the irradiation are ca. seven times those in the dark. On the other hand, the photoinduced increase in permeability is less significant above the pH region. This pH dependence of the photoinduced enhancement of the permeation might be attributable to the pH-dependent generation of triphenylmethyl cations under light irradiation as shown in Figure 3. The permeability constant in the dark increases with increasing pH because of swelling of the capsule membrane due to dissociation of the polyelectrolyte complex.⁹

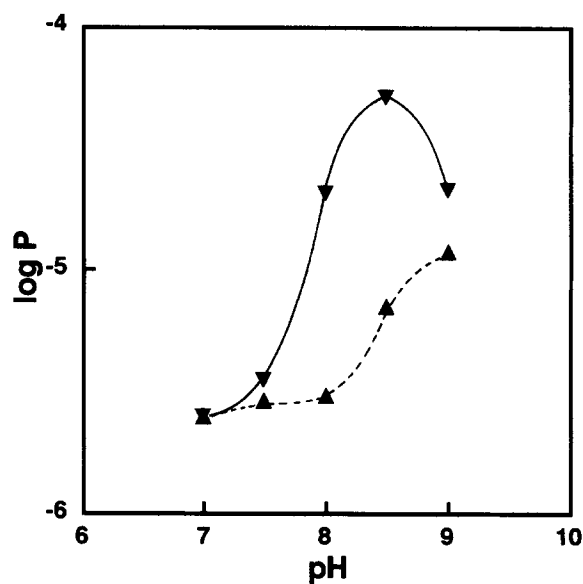


Figure 6 pH-dependence of permeability constant of *p*-toluenesulfonate through CP-1 membrane (▲) in the dark and (▼) under irradiation at 30°C.

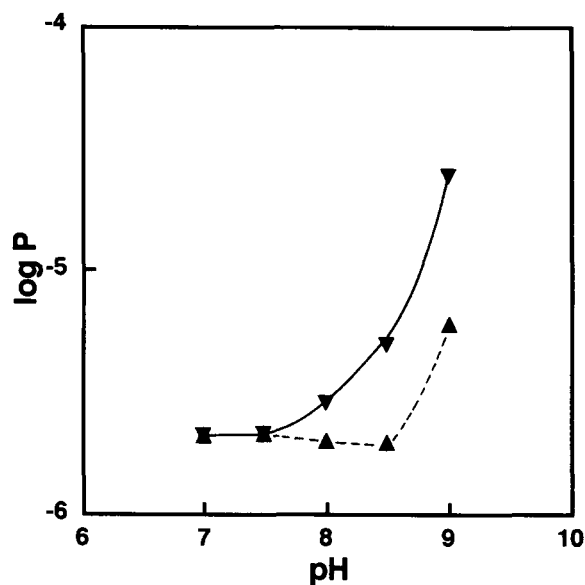


Figure 7 pH-dependence of permeability constant of *p*-toluenesulfonate through CP-2 membrane (▲) in the dark and (▼) under irradiation at 30°C.

It is expected that the capsule membrane containing larger amounts of photosensitive groups reveals a more significant photoinduced change in permeability. Thus, CP-2, in which the content of photosensitive copolymer is twice that in CP-1, was prepared and the photosensitivity of the permeability of the capsule membrane was investigated. Figure 7 shows the pH dependence of the permeability constant of *p*-toluenesulfonate through the CP-2 membrane in the dark and under the irradiation. The maximum value of the permeability constant under light irradiation appears in Figure 7. Contrary to expectation, CP-2 reveals a lesser effect of photoirradiation on the permeability than does CP-1. Because triphenylmethane leucohydroxide residues are highly hydrophobic, the capsule membrane containing too much of this group should become dense,¹⁵ resulting in low permeability even when triphenylmethyl cations are generated. This agrees with the lower permeability of CP-2 than that of CP-1 in the dark.

As is apparent in Figure 3, the amount of the triphenylmethyl cation on the copolymer increases with decreasing pH in the dark under experimental conditions. If the capsule is prepared at higher pH, a larger amount of the unionized triphenylmethane residue in the dark should exist in the capsule membrane, resulting in a higher response to the light. Therefore, the influence of the pH of the polymer solutions in the capsule preparation on the photoresponsive release property of the resulting capsule

was investigated. Figure 8 depicts the permeability constant of *p*-toluenesulfonate through the membrane of CP-3, which was prepared at a higher pH value than was CP-2, under light radiation as well as in the dark. The photoresponse of CP-3 is apparently more significant than that of CP-2 above pH 8. Therefore, this result suggests that the CP-3 membrane possesses a greater amount of triphenylmethane leucohydroxide residues which are not ionized in the dark and, hence, are light-sensitive compared to the CP-2 membrane.

To obtain information about the mechanism of the photoresponsive permeation through the capsule membrane, the permeation of phenylethylene glycol, which is a nonionic molecule, through the membrane was investigated. However, photoinduced enhancement of the permeation was not observed (result not shown). This result suggests that the electrostatic interaction between permeants and photosensitive groups in the capsule membrane plays an essential role in the photosensitive permeation property of the capsule membrane. Because, under the weak alkaline condition, where the photoinduced permeation enhancement of *p*-toluenesulfonate occurs, the capsule membrane is negatively charged due to the deprotonation of polyethylenimine constituting the capsule membrane, permeation of the negatively charged molecule, *p*-toluenesulfonate, through the capsule membrane is depressed.⁹ However, when triphenylmethyl cations are generated in the capsule membrane by photoirradiation, charge

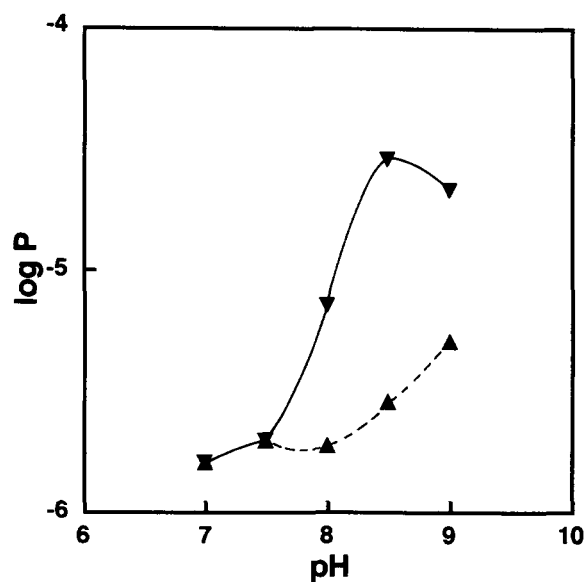


Figure 8 pH-dependence of permeability constant of *p*-toluenesulfonate through CP-3 membrane (▲) in the dark and (▼) under irradiation at 30°C.

neutralization in the capsule membrane should occur. Moreover, the triphenylmethane residues might exist as an aggregated form in the membrane because of their great hydrophobic nature. Aggregation of the triphenylmethane residues is assumed by the following observations: Although the molecular weight of the copolymer is much higher than that of poly(acrylic acid), the copolymer shows much less viscosity than does the poly(acrylic acid) used in this study, indicating compact conformation of the former due to hydrophobic interactions between the triphenylmethane residues. The copolymer having less of the triphenylmethane residues was also prepared by the same method (content of the triphenylmethane residue: 0.6 unit mol %, molecular weight: 80,000). The molecular weight of the copolymer with a larger amount of the triphenylmethane residues is much higher than that with less of the residues. This fact also suggests intermolecular association of the former induced by aggregation of the triphenylmethane residues. It is likely that *p*-toluenesulfonate permeates through the triphenylmethane residue-rich domains in the capsule membrane under photoirradiation. This may be why the capsule membrane reveals the photoresponse in spite of the relatively low content of the photosensitive groups in the capsule membrane.

Figure 9 shows the photoresponsive release of *p*-toluenesulfonate from the capsule. While the permeant is released at a low rate in the dark, upon light irradiation, the release rate of the permeant increases remarkably after a several minute time

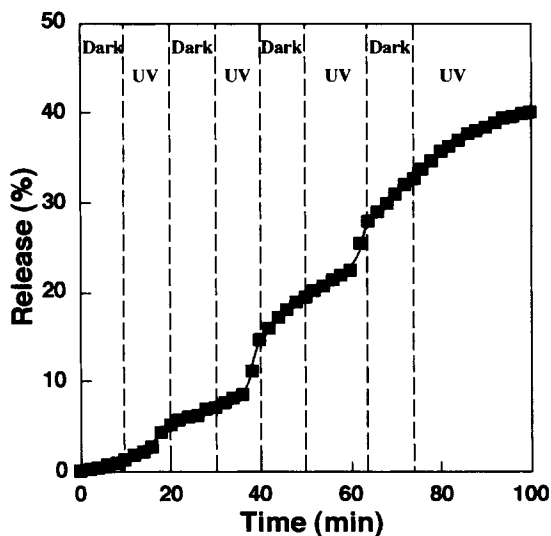


Figure 9 Photoresponsive release of *p*-toluenesulfonate from CP-1 at pH 8.0 at 30°C.

lag. When the capsule is again put in the dark, the release rate decreases immediately. This permeability control responding to the light can be achieved several times. However, the photoresponse is diminished after 75 min, possibly because of the degradation of the photosensitive groups in the capsule membrane, as mentioned above.

In conclusion, the partly crosslinked poly(acrylic acid)-polyethylenimine complex capsule membrane having the triphenylmethane leucohydroxide residues prepared in this study was found to control its permeability for an anionic molecule, *p*-toluenesulfonate, responding to ultraviolet light irradiation. Although the capsule leaves something to be desired, it might have potent usefulness for photosensitive drug-delivery devices.

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