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Preparation of Amphoteric Microgels of Poly(acrylamide/methacrylic acid/dimethylamino ethylene methacrylate) with a Novel pH−Volume Transition

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ABSTRACT: Amphoteric microgels consisting of poly(AAm/MAc/DMAEMA/MB) were prepared by semicontinuous polymerization in ethanol. Scanning electron microscopy (SEM) and dynamic light scattering (DLS) were employed for evaluating the microspheres and their pH−volume transitions in the aqueous solution, respectively. Monodispersed amphoteric microgels with a U-type of pH−volume transition, i.e., volume contracted in the pH range 6.5−7.5 and expanded beyond this, were prepared by adding MAc dropwise at a certain rate (20 g/80 min, standard recipes). It was observed that increasing [NaCl] or [$CaCl_2$] served to decrease the hydrodynamic diameter of amphoteric microgels, irrespective of pH. Moreover, in the region of pH 2−3, the pH of amphoteric microgels increased with the increase of ion strength, whereas inversely, for pH 9−10, the pH decreased with an increase of ion strength, regardless of the types of salts. The U-type curve of diameter vs pH could be replaced along with the pH axis by adjusting the charged amount of DMAEMA. Control-releasing behavior of amphoteric microgels was also investigated. It was observed that the dye was released to the same extent when the pH was changed from 6.8 to either acidic or basic conditions.

Introduction

Microcapsules have been widely applied in various fields[1−4] for the controlled-releasing of medicines, bioactive substances, catalysts, cosmetics, etc, and even more, as the microchambers for controlled reactions. Recently, with the development of chemotherapeutic and DNA repairing techniques, microcapsules that enable one to fine control the releasing of medicines were developed in a big way; namely, the microcapsules released the medicines only under specific conditions such as the ionic strength, including pH and temperature. That is to say, a small stimulus triggering a drastic release was pursued. A hydrogel microsphere (microgel) consisting of materials having a characteristic deswollen−swollen volume change in response to pH[1,2] or temperature[3,4] change is a typical example. The transition of hydrophobicity−hydrophilicity is an effective factor to capture and expel a molecule from a microsphere without any wall hindrance. Hence, a sharp controlled-releasing behavior is expected.

Amphoteric microgels contain both polyanionic and polycationic polymers; thus, they are considered as potential pH-sensitive microcapsules that release drugs only by responding to a specific pH. However, due to the instability of microgels in the aqueous solution, several special approaches have been tried so far, namely the conversion of the functional group on the surface of microspheres[5−7] and the preparation of microgels by using an amphoteric surfactant[8] or initiator.[9] Obviously, by applying these approaches, the amphoteric groups just exist on the surface of microspheres. Moreover, the number or ratio of two counter-ionic groups is not controllable, especially the approaches using amphoteric surfactant or initiator. For example, there are the amphoteric microgels of poly(acrylamide (AAm)/methacrylic acid (MAc) and the cross-linking agent, methylene bis(acrylamide) (MBAAm) and *p*-nitrophenyl acrylate (NPA). After the anionic microgels were prepared, they transferred the nitrophenyl group to the amino group by use of the diamine. However, due to the instability of NPA and the complicated preparative process, directly preparing the amphoteric microgels by using the amino monomer, *N*-(dimethylamino)ethylene methacrylate (DMAEMA), was essential.

A new understanding of microsphere formation gives new approaches to the preparation of microspheres. Therefore, in this paper, we will introduce a new method to prepare the amphoteric microgels with sharp pH−volume transition on the basis of our previous researches.[10−14]

Experimental Section

Materials. All the reagents and solvents used in this paper were purchased from Wako Pure Chemical Industries, Ltd., Japan. The monomers methacrylic acid (MAc) and (dimethylamino)ethylene methacrylate (DMAEMA) were purified by distillation under the reduced pressure. The monomer acrylamide (AAm), the cross-linker *N*,*N*′-methylene bis (acrylamide) (MB), the initiator, dimethyl 2,2′-azobis (isobutyrate) (DMAIB), dye Gentian VB, Vitamin E, and the solvent ethanol (EtOH, dehydrated) were used without further purification.

Polymerization. The operation of batch polymerization has been introduced in a previous paper.[13] The semicontinuous polymerization was carried out in the same reactor, similar to the batch polymerization described above. The remaining monomer, commonly MAc, dissolved in 20 g of EtOH was automatically added by using a syringe pump (Harvard PHD, HA2000I, Harvard Apparatus Co. Ltd, Japan) after the initiator solution was charged.

Characterizations. The characterization methods are the same as reported in the previous papers,[13,14] if no specific comment is

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Table 1. Recipes for Preparation of Amphoteric Microspheres[a]

	molar ratio of monomers[a]					
run no.	AAm	MAc	DMEMA	MB[b] (mol %)	EtOH (wt %)	other
6027	6	1	1	3	95	
6028	6	1.8	0.2	3	95	
6029	6	1.8	0.2	10	95	
6031	6	1.8	0.2	5	76	19 wt % (1 N NaOH)[c]
6032	6	1.8	0.2	5	76	19 wt % (1 N HCl)[d]
6034	6	1.95	0.05	5	95	
6049−6056	6	1.5	1.5	3	95	
6054	3	1.5	1.5	3	95	
6057−6058	6	1	2	3	95	
6059	6	1.3	1.7			
6060	6	1.2	1.8			

[a] The total initial concentration of monomers was 5 wt %. [b] MB, cross-linking agent *N*,*N*′-methylene bis(acrylamide), molar percentage based on the total amount of monomers. [c] NaOH−EtOH solution. [d] HCl−EtOH solution. 6049: Batch polymerization. 6050: Dropwise addition of a mixture of MAc, DMAEMA, and MB (in 20 g of EtOH), Rate: 20 g/80 min. 6051: Dropwise addition of MAc. Rate: 20 g/80 min. 6053: Dropwise addition of DMAEMA (in 20 g of EtOH). Rate: 20 g/80 min. 6054: Dropwise addition of MAc (in 20 g of EtOH). Rate: 20 g/80 min. 6055: Dropwise addition of MAc (in 20 g of EtOH). Rate: 20 g/30 min. 6057: Dropwise addition of MAc (in 20 g of EtOH). Rate: 20 g/80 min. 6058: Dropwise addition of MAc, DMAEMA and MB (in 20 g of EtOH) into AAm solution. Rate: 20 g/80 min. 6059: Dropwise addition of MAc (in 20 g of EtOH). Rate: 20 g/80 min. 6060: Dropwise addition of MAc (in 20 g of EtOH). Rate: 20 g/80 min. [e] Note: initiator, DMAIB (dimethyl 2,2′-azobis(isobutyrate)) was 2 wt % based on the total amount of monomers.

made. Dynamic light scattering (DLS) (ELS-8000, Otsuka Electronics, Inc, Osaka, Japan) was employed to measure the hydrodynamic diameters of sample, by which the scattering light at 90° was collected, thereby the diameters were automatically calculated, and among them, scattering strength diameter (relative to the weight-average and number-average diameter) was selected to characterize the volume change of the samples. The solution of 0.5 wt % used for the DLS measurement was prepared as follows. The dry sample treated with the method described above was mixed with pH 2 HCl solution or pH 10 NaOH solution in a 30 mL screw bottle due to the coagulation of microgels in the neutral water, and the bottle was put in an ultrasonic automatic washer to disperse the sample for 1 h (Iuchi US-4, SMT Co Ltd, Fukuoka, Japan). Here, 1 M HCl (NaOH) was used to adjust the pH of dispersion and the pH was measured with a pH meter (MP220, Mettler-Toledo AG, CH8603, Schwertzenbach, Switzerland). All measurements of DLS were performed at 20 °C.

The sample for Cryo-TEM (JEM400SFX, JEOL, Japan) was prepared as following: the powder of amphoteric microgels was dispersed in pH 2 solution with a formulated concentration of ca. 1.5 wt %, and then the pH was adjusted with NaOH (1 N) solution to the pH value required. A copper grid was dipped into the aqueous dispersion to obtain a droplet on the grid, and a filter paper was used to drain off the excessive solution. Then the grid was put into liquid propane for a few seconds before being transferred into liquid nitrogen. The sample was transferred to a TEM holder in liquid nitrogen.

The control-release of Gentian VB was performed as following. The solution containing 5 wt % amphoteric microgels was formulated by dispersing amphoteric microgels in an aqueous solution of 10^{-4} mol/L Gentian VB, and then divided into three parts. The three parts of dispersion were pH-adjusted to pH 3, pH 7, and pH 10 respectively and then centrifuged at 3000 rpm/5 min. The serums of three parts were measured by vis−UV equipment (JASCO 8000, Nippon Benko Co. Ltd., Japan).

Results and Discussion

Preparation of Amphoteric Microgels. In the previous papers,[13,14] we reported that the microgels with the sharpest pH−volume transition were prepared by the recipe AAm/MAc = 3/1 and cross-linking degree MB 3 mol %. On the basis of this result, we attempted to prepare amphoteric microgels with a sharp pH−volume transition by modifying the recipe. The recipes are shown in Table 1, i.e. keeping AAm to a constant, AAm/(MAc + DMAEMA) = 3/1, but changing the ratios of MAc/EGDMA. The difficulty to prepare the amphoteric microgels was the instability of microgels during the polymerization.[4−9] Therefore, at first, the factors possibly contributing to the stability were tried, such as the ratios of anions/cations and charges of monomer. As shown in Table 1 (runs 6027−6034), batch polymerization was employed and the ratios of MAc/DMAEMA increased from 1/1 to 1.95/0.05 mol/mol in order to keep the net charges of microgels negative. The addition of NaOH or HCl (runs 6030−6033) to the polymerization system was also used for that purpose. As shown in Figure 1, the amphoteric microgels were prepared, but polydispersed. On the other hand, a method of increasing the cross-linking degree was tried (Table 1, runs 6029−6034), which we introduced in previous paper,[10,11,13] that showed it was valid to prepare monodispersed microgels, especially in the case of higher content of MAc. It was observed that the relative monodispersed microgels were only prepared with a higher cross-linking degree (run 6029 (Figure 1c)).

However, as shown in Figure 2, the typical characteristics of pH−volume transition prepared by above methods was similar to those of poly(AAm/MAc) microgels,[13,14] except that the pH−volume transition moved to higher pH. The pH−volume transition of cationic polymer, PDMAEMA, i.e., where the volume expanded at lower pH and contracted at higher pH, was not observed. This result indicated that most of the DMAEMA monomer was not combined into the polymers of microgels. Therefore, it was concluded that the amphoteric microgels could not be prepared along with the conventional mechanisms, which said that AAm first polymerized in the continuous phase and precipitated to form nuclei, and then the remaining monomers swelled the nuclei and polymerized in the nuclei.

Coexistence of MAc and DMAEMA in the growing microgels was the basic premise used to prepare the amphoteric microgels. The new mechanism suggested that the Coulombic interaction of MAc−DMAEMA likely was more active than Mac−AAm, but poly-MAc,[10−12] poly-DMAEMA, and their copolymer were soluble in ethanol. It destroyed the minimonomer droplets of AAm−MAc complexes, and resisted the precipitated poly-AAm segments to take DMAEMA into the growing microgels. This was the reason that for batch polymerization, polydispersed microgels were prepared and the sharp pH−volume transition was not obtained. The key premise was that AAm polymerized and precipitated in the minimonomer droplets. It played the role to take the soluble molecules of MAc or segments of poly-MAc into the microgels by intensive interaction. Such a consideration gave us a hint that it was

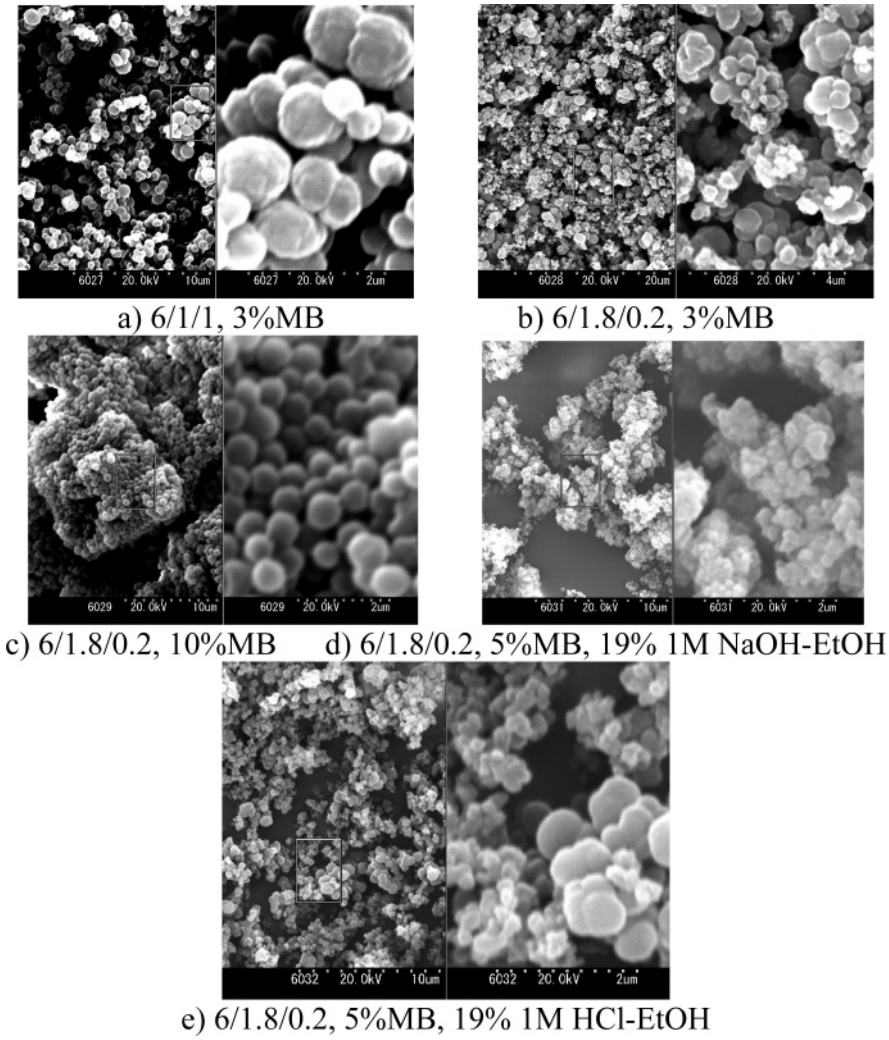

Figure 1. SEM photos of microspheres by batch polymerization.

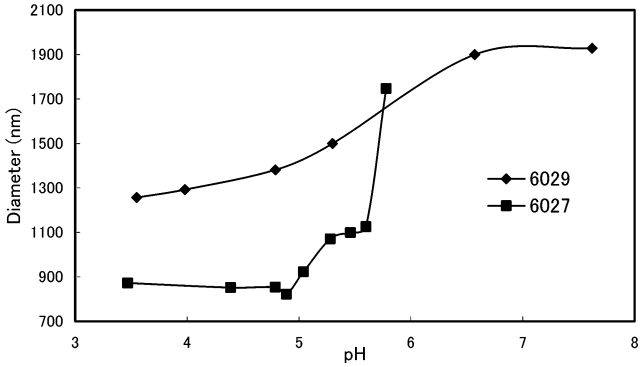

Figure 2. pH−volume transition of microspheres by batch polymerization.

possible to prepare monodispersed amphoteric microgels, if we localized the interaction of MAc−DMAEMA within the microgels and eliminated the free Mac and DMAEMA in the continuous phase as much as possible. Accordingly, two strategies were applicable. The first one (strategy I) was to let AAm first polymerize with DMAEMA, thereby localizing the interactions within the microgels, or briefly, fixing DMAEMA first. On the other hand, the next one (strategy II) was to fix Mac first. Both of these strategies implied that semicontinuous polymerization was essential. Therefore, as shown in Table 1 (runs 6050−6056), various dropwise addition methods were applied with a fixed molar ratio of MAc/DMAEMA = 1/1 and a molar ratio of AAm/(MAc + DMAEMA) = 2/1. A molar ratio of AAm/(MAc + DMAEMA) = 1/1 was also tried (run 6054), but unfortunately microgels were not prepared probably due to the high lyophilicity of polymer complexes. Instead, a macrogel was obtained.

For comparison, the other polymerization methods were preliminarily applied. The SEM photos of these microspheres are shown in Figure 3 and the curves of their pH−volume transitions are shown in Figure 4. Run 6049 was prepared by batch polymerization. The size of the microgels was large and the distribution was polydisperse as shown in Figure 3. As for their pH-sensitivity, as shown in Figure 4, a common characteristic was observed, i.e., agglomeration of the microgels took place at neutral pH (dotted line in Figure 4). This phenomenon indicated that both amino groups and carboxylic groups were incorporated into the microgels. Compared with other samples, run 6049, e.g., the sample prepared by batch polymerization, gave a small pH−volume change at lower pH. This result indicated that less of poly-DMAEMA was incorporated into the microgels. Run 6050 was semicontinuous polymerization, by which the mixture of MAc, DMAEMA, and MB was added dropwise into the polymerization system of AAm. In this case, the poly-AAm first precipitated to form many polydispersed entities due to randomly physical aggregation.

Therefore, as shown in Figure 3b, irregular and polydispersed microgels were prepared. The post-added MAc and DMAEMA interacted with the precipitated entities of poly-AAm, thereby

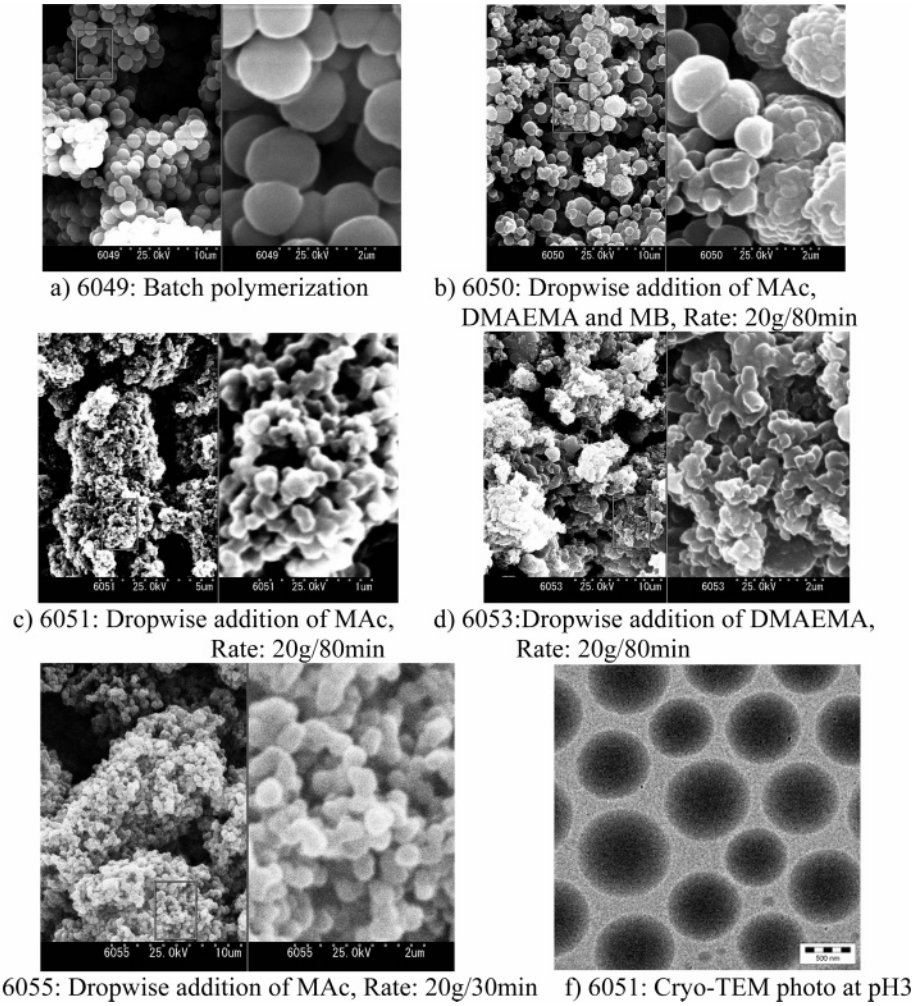

Figure 3. SEM photos of microspheres prepared by semicontinuous polymerization.

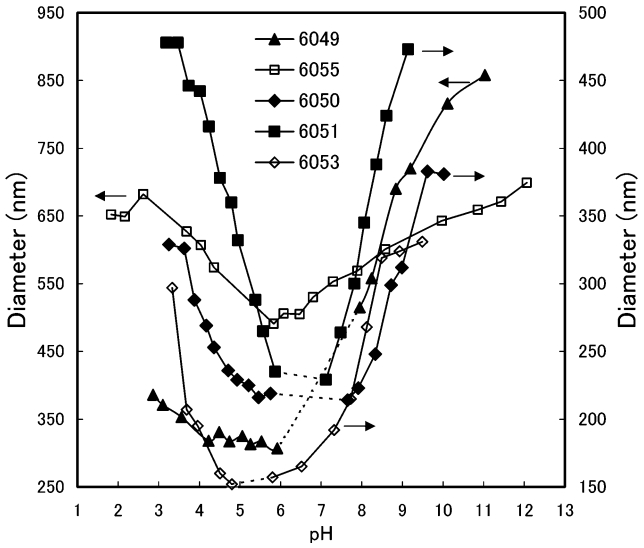

Figure 4. pH−volume transition of microspheres by semicontinuous polymerization.

combining into the entities. As a result, the entities showed the typical pH−volume transition of an amphoteric polyelectrolyte. The hydrodynamic diameter of microgels, as shown in Figure 4, decreased with increasing pH when the pH was lower than pH 6.0, whereas when the pH was higher than pH 7.5, the diameter increased with increasing pH.

Run 6051 was carried out based on strategy I, namely fixing DMAEMA first. MAc was added dropwise into the polymerization system of AAm, DMAEMA, and MB. It should be remarked that the phenomenon of precipitation stemming from poly-AAm was not observed before MAc was added. This result implies that the copolymerization of AAm-DMAEMA may take place in the polymerization system, but the copolymer did not precipitate, probably due to the high solubility of poly-DMAEMA segments in ethanol. It was the key premise used to prepare the monodispersed microgels, because if precipitation happened, the same result as run 6050 would be obtained. Thereafter, the post-added MAc interacted with poly-AAm segments or free AAm in the system to form mini monomer droplets. The SEM photo of the products is shown in Figure 3c. The microspheres seem small, but the shape of the microspheres was not distinct. The reason may be attributed to the extremely low glass transition temperature of poly-DMAEMA (−35 °C[15]). Therefore, in order to confirm the formation of amphoteric microgels, the cryo-TEM photo of run 6051 was taken at pH 3. As shown in Figure 3f, the microgels were clearly observed. The comparatively smaller size indicated that, as we expected, the fixed poly-DMAEMA segments played stabilizing role in term of steric hindrance. Consequentially, as shown in Figure 4, a U-type of the sharp pH−volume transition of typical amphoteric microgels was observed. The curve of pH−volume transition was nearly symmetric in the two pH regions. The symmetrical curve implied that an amount of

DMAEMA almost equal to MAc was taken into the polymer of the microgels.

Run 6053 was performed in accordance with strategy II, e.g., fixing MAc first. DMAEMA was added dropwise into the polymerization system of MAc, AAm, and MB. It sounded similar to strategy I, but quite different in truth. As we know, the polymerization first takes place within the minimonomer droplets of AAm (MB)-MAc[10,12,16,17] to form network nuclei. Both the precipitation of poly-AAm in the minimonomer droplets and the cross-linking structure of the initial polymer cause the fixed poly-MAc segments to be buried in the solid loci of microgels. Therefore, the capture efficiency of DMAEMA was possibly low. The pH−volume transition of run 6053 was shown in Figure 4. Obviously, compared with that of run 6051, the curve of run 6053 was unsymmetrical and moved to a lower pH region. For example, it moved from pH 5.0 to pH 6.0 (dotted line in Figure 4) wherein the agglomeration of microspheres occurred. Another trial was to investigate the effect of the dropwise addition rate. The dropwise addition of MAc into the polymerization system of AAm, DMAEMA and MB was carried out at a rate of 20 g/30 min (run 6055), faster than those reported above (20 g/80 min). Microgels with a distinct shape were observed in Figure 3e; meanwhile, as we commented above, such a distinct shape may also imply that less DMAEMA was taken into the microspheres. Actually, as shown in Figure 4, a V-type and gradual curve of the pH−volume transition was observed. Moreover, the pH region in which the amphoteric microgels agglomerated was very narrow due to the neutralized charges. These results indicated that the higher the addition rate, the smaller the amount of MAc and DMAEMA that was taken into the polymer of the microgels.

Readers might question us about the concepts of pK_a of poly-MAc and pK_b of poly-DMAEMA. We have commented on pK_a of MAc in the previous paper[13,14] that for the constrained poly-MAc segments in a network, thus confined by thermodynamic factors, there was no independent and appropriate pK_a to describe the dissociating behavior of the MAc unit in a hydrogel microsphere. It is the same situation as for the pK_b of DMAEMA. In any case, the amino group of DMAEMA and the carboxylic group of MAc may consist as a pair of ions as in a buffer solution. It implies the possibility that the hydrodynamic diameter of amphoteric microgels may change due to the ratio variation of anions/cations, but the pH remains relatively constant. This possibility was not reflected in the curves of the pH−volume transition, because the investigation was solely designed to understand the volume change with the variation of pH.

Design of the pH—Volume Transition. In the above section, we discussed the preparation of amphoteric microgels with a U-type pH−volume transition. In this section, we addressed the possibility to design the pH−volume transition curve of amphoteric microgels, i.e., setting the pH−volume transition down into the pH range required.

Adding more MAc may move the range of agglomeration to a lower pH region. Run 6053 was an example. Compared to run 6051 (pH 5.9 to pH 7.1 (Figure 4)), the range of run 6053 shifted in the range pH 4.8 to pH 5.8. Here, our interest was whether it was possible to shift the pH−volume transition to higher pH, which meant the addition of more soluble DMAEMA into the microgels. As shown in Table 1, several trials were carried out (runs 6057−6058) where the ratio of MAc/DMAEMA was equal to 1/2. The microgels were not prepared, but a polymer solution was obtained. It indicated that the amount of DMAEMA was too large to render the copolymer including

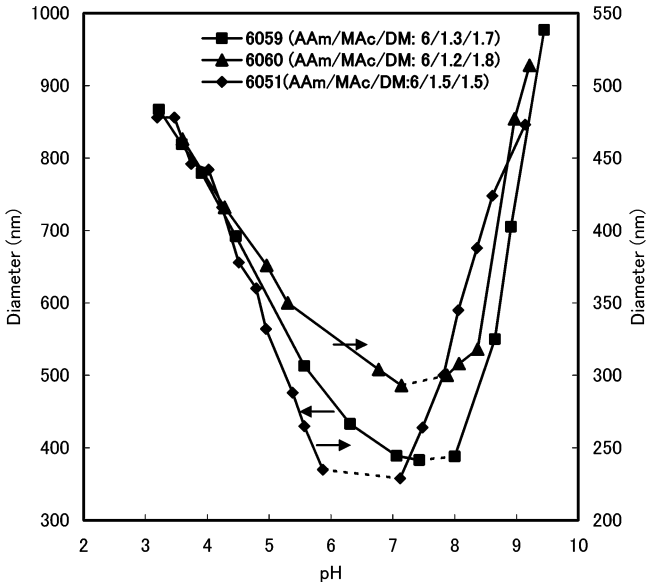

Figure 5. Effect of MAc/DMAEMA on the pH−volume transition of amphoteric microspheres.

poly-AAm segments to precipitate. Therefore, run 6059 and 6060 were performed with less DMAEMA in recipes (Table 1). As shown in Figure 5, compared with the pH−volume transition of run 6051, a tendency became obvious: the pH range of agglomeration moved to higher pH as the content of DMAEMA increased in the recipes, though the increment of DMAEMA was small. It implies that the design of a pH−volume transition was possible. Meanwhile, the size of the microgels changed a lot. It might result from the instability of microgels during the formation process.

Effects of Ion Strength on the Hydrodynamic Diameter of Amphoteric Microgels. Amphoteric microgels (run 6051) with a sharp pH−volume transition were selected as the sample to investigate the effects of ion strength. Two pH regions, i.e., pH 9−10 and pH 2−3 were stressed because, at neutral pH, the microgels agglomerated. In the previous paper,[14] we reported that [NaCl] and [$CaCl_2$] affected the hydrodynamic diameter of poly(AAm/MAc) in different modes. Addition of $CaCl_2$ rendered the hydrodynamic diameter to monotonically decrease as [$CaCl_2$] increased. However, at low [NaCl], an abnormal phenomenon where the diameter increased was observed with increasing [NaCl]. Osmotic equilibrium of ions and the effect of electric field shielding of counterions were the dominant reasons. Additionally, the pH decreased with the increase of salt concentration, irrespectively of the salt being NaCl or $CaCl_2$. It was concluded that the exchange of cations with H of carboxylic groups proceeded as the concentration of salt increased. In the case of amphoteric microgels, however, as shown in Figure 6 and Figure 7, the hydrodynamic diameter monotonically decreased as the concentration of salt increased, regardless of the properties of the salt. Moreover, the pH increased with the increase of ion strength at pH 2−3, while at pH 9−10, the pH decreased with the increase of ion strength, irrespective of the type of salts. Thermodynamic factors such as the free energy of polymer−solvent mixing and equilibrium of osmotic pressure were considered as the dominant factors for the behaviors of amphoteric microgels at different ion strengths. Tertiary amines of DMAEMA and the carboxylic acid of MAc composed the amphoteric microgels of run 6051. At lower pH such as pH 2, the protonated tertiary amine contributes to the low free energy of polymer−water mixing, while at a higher pH, such as pH 10, the dissociated carboxylic acid

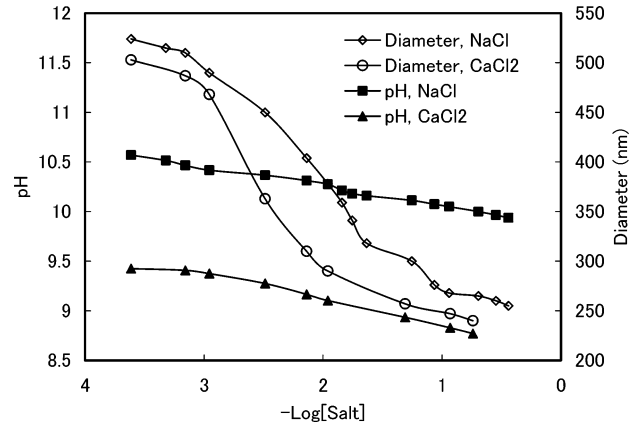

Figure 6. Effects of NaCl and $CaCl_2$ on the hydrodynamic diameters of microspheres at higher pH.

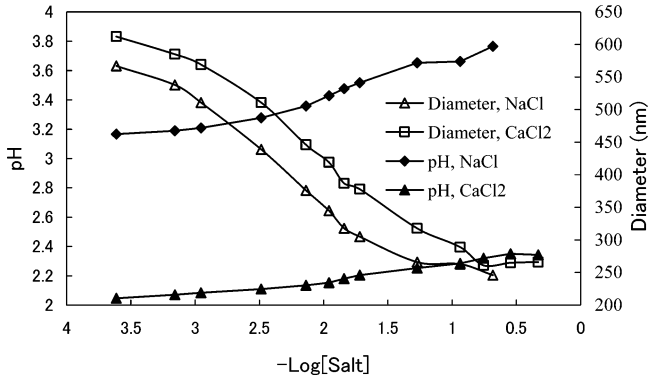

Figure 7. Effects of NaCl and $CaCl_2$ on the hydrodynamic diameters of microspheres at lower pH.

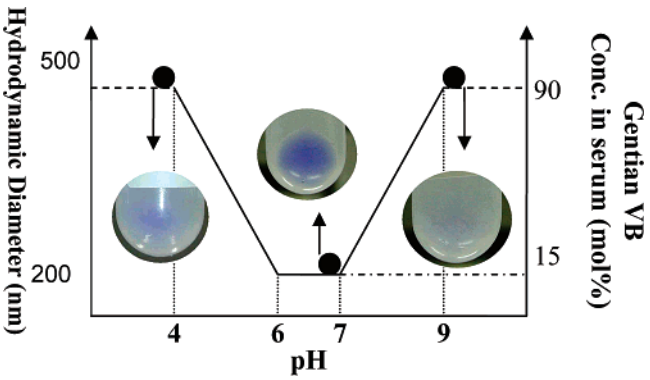

Figure 8. Controlled release of Dye Gentian VB by amphoteric microspheres (run 6051).

contributed to it. Therefore, at lower pH, the number of cations was dominant in amphoteric microgels. In such a situation, the equilibrium of osmotic pressure favored a large number of anions, Cl^-, to osmose into the microspheres. Numerous Cl^- anions in the microsphere shielded the electric field stemming from the protonated tertiary amines, and furthermore, they forced the carboxylic groups to associate. These two effects determined that the amphoteric microgels of run 6051 contracted and the pH of the solution increased when the salt was added. At higher pH, the cations, i.e., Na^+ and Ca^{2+}, played the same roles as Cl^- did at the lower pH. A remarkable difference was that at higher pH, cations made the protonated tertiary amine to release protons. That was why the pH of solution decreased with the increase of [salt].

Controlled-Release Behaviors of Amphoteric Microgels. An amphoteric dye, Dye Gentian VB, was selected for the evaluation of controlled-release behavior of amphoteric microgels, run 6051. A quantitative analysis of the controlled-release behavior of amphoteric microgels was performed, run 6051. The ionic interaction was considered as the main factor to controlled release of the dye. It had the same electric charges as the amphoteric polymer in the microspheres. Therefore, as shown in Figure 8, at pH 3.5, there was only 10% of dye absorbed by the microgels, whereas at pH 6.8, there about 85% of dye was in the microgels. At pH 9.3, there was 6% of dye absorbed. In other words, when the pH changed from pH 6.8 to pH 3.5 or pH 9.3, there was about 75−80% dye released from the amphoteric microgels to the aqueous solution.

Conclusions

Amphoteric microgels consisted of poly(AAm/MAc/DMAEMA/MB) were prepared by semicontinuous polymerization in ethanol. Batch polymerization produced bigger and polydispersed microspheres, and less MAc and DMAEMA were taken into the microgels. The monodispersed amphoteric microgels with novel pH−volume transitions were prepared by adding MAc dropwise at rate of 20 g/80 min into the polymerization system of AAm, DMAEMA, and cross-linking agent, MB. Higher addition rate or dropwise addition of other monomer or monomer mixture resulted in the microspheres with a smaller amount of DMAEMA. The unique pH−volume transition of amphoteric microgels could be designed by adjusting the charged amount of DMAEMA within a limited ratio of MAc/DMAEMA = 1/2 (mol/mol). Beyond the limited ratio, a soluble copolymer was produced due to the high content of soluble poly-DMAEMA segments. Differing from the polyanionic microgels, it was observed that increasing [NaCl] or [$CaCl_2$] in the dispersion of amphoteric microgels was to decrease the hydrodynamic diameter, irrespective of pH. Moreover, in the region pH 2−3, the pH of dispersion of amphoteric microgels increased with the increase of ion strength, whereas inversely, for pH 9−10, the pH decreased with the increase of ion strength, regardless of the types of salts. Controlled-release behavior of amphoteric microgels with novel pH−volume transition was also investigated. It was observed that, at pH 3.5, there was only 10% of Dye Gentian VB absorbed by the microspheres, whereas at pH 6.8, there was about 85% dye in the microgels. At pH 9.3, there was 6% of dye absorbed in the microspheres. In other words, when the pH changed from pH 6.8 to pH 3.5 or pH 9.3, there was about 75−80% dye released from the amphoteric microgels to the aqueous solution.

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References and Notes

(1) Lin, C-L.; Chiu, W-Y. *J. Polym. Sci., Part A: Polym. Chem.* **2006**, *44*, 1648.
(2) Marcilla, R.; Sanchez, P. M.; Lopez, R. B.; Lopez, C. E.; Ochoteco, E.; Grande, H.; Mecerreyes, D. *J. Polym. Sci., Part A: Polym. Chem.* **2006**, *44*, 3958.
(3) Lin, C-L.; Chiu, W-Y.; Lee, C-F. *J. Polym. Sci., Part A, Polym. Chem.* **2006**, *44*, 356.
(4) Hazot, P.; Chapel, J. P.; Pichot, C.; Elaissari, A.; Delair, T. *J. Polym. Sci., Part A: Polym. Chem.* **2002**, *40*, 1808.

(5) Kawaguchi, H.; Fujimoto, K.; Nakazawa, Y.; Sakagawa, M.; Ariyoshi, Y.; Shidara, M.; Okazaki, H.; Ebisawa, Y. *Colloids Surf. A: Phys. Chem. Eng. Asp.* **1996**, *109*, 147.
(6) Kashiwabara, M.; Fujimoto, K.; Kawaguchi, H. *Colloid Polym. Sci.* **1995**, *273*, 339.
(7) Musyanovych, A.; Adler, H. J. P. *Langmuir* **2005**, *21*, 2209.
(8) Fang, S.-J.; Fujimoto, K.; Kondo, S.; Shiraki, H.; Kawaguchi, H. *Colloid Polym. Sci.* **2000**, *278*, 864.
(9) Gu, S.-C.; Akama, H.; Nagao, D.; Kobayashi, Y.; Konno, M. *Langmuir* **2004**, *207*, 7948.
(10) Ni, H.-M.; Kawaguchi, H. *J. Polym. Sci., Part A: Polym. Chem.* **2004**, *42*, 2823.
(11) Ni, H.-M.; Kawaguchi, H. *J. Polym. Sci., Part, A: Polym. Chem.* **2004**, *42*, 2833.
(12) Ni, H.-M.; Du, Y.-Z.; Ma, G.-H.; Nagai, M.; Omi, S. *Macromolecules* **2001**, *34*, 6577.
(13) Ni, H.-M.; Kawagchi, H.; Endo, T. *Colloid Polym. Sci.* **2007**, *285*, 873.
(14) Ni, H.-M.; Kawagchi, H.; Endo, T. *Colloid Polym. Sci.* **2007**, *285*, 819.
(15) Brandrup, J.; Immergut, E. H. *Polymer Handbook*, 3rd ed.; Wiley: New York, 1989; Vol. II.
(16) Nakazawa, Y.; Kamijo, K.; Fujimoto, H.; Kawaguchi, Y.; Yuguchi, H.; Urakawa; Kajiwara, K. *Angew. Makromol. Chem.* **1996**, *240*, 187.
(17) Kamijo, Y.; Fujimoto, K.; Kawaguchi, H.; Yuguchi, Y.; Urakawa, H.; Kijiwara, K. *Polym. J.* **1996**, *28*, 309.

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