Rapid Redispersion of pH-Sensitive Microgel Flocculation with Poly(lysine)

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ABSTRACT: A concentrated aqueous flocculation of an ultrafine microgel of methyl methacrylate-acrylic acid (MMA-AA) copolymer mixed with poly(L-lysine) (PLL) exhibited rapid and reversible redispersion when the solution was shifted to pH 10-11. In lower pH where PLL was ionized, the microgel underwent bridging flocculation. A pH increase induced PLL to transform from an ionized coil to an α-helix conformation, which resulted in desorption of the bridging PLL to bulk solution from the microgel surfaces. The desorption was assisted by electrostatic repulsion between the microgels, whose charge was converted to be highly negative by the neutralization of the PLL. Removal of the PLL chain from microgels (spatial shift) due to its conformational change in concurrence with pH-dependent shrinkage of microgels instantaneously separated an individual microgel, which resulted in remarkably rapid and reversible redispersion. The large magnitude of the change in absorbance at the transition may be applied for electrooptical devices.

Introduction

Tanaka et al. went through with a generalized, theoretical approach to environmental-sensitive polymer gels and showed that swelling of a submicron-sized gel could respond within microseconds.[1] Since then, microgels have obtained strong interest from the viewpoint of functional materials for optoelectronic, chemomechanic, and biomedical devices. However, environmentally sensitive gels have not yet been in practical use in these applied fields, because response time and the driving method have been major problems. Environmental factors to affect microgels are temperature, electric field, pH, and solvent composition. Among them, microgel driving with pH is the most promising in terms of the lifetime of working gels.

Our interest lies in the application of microgels to electrooptical devices such as display and optical modulators. We have recently devised an electrically controlled pH modulator, which was composed of conductive polymer-coated electrodes.[2] pH-sensitive ultrafine microgels have also been prepared by our research group.[3] The volume of pH-sensitive microgel was electrically controlled in a pH-modulator cell.[4] However, in this system the magnitude of change in optical transmittance was too small to apply in practical uses, because of the reduction in the refractive index by the absorbance of water at the swollen state. In consequence, our study has focused on another strategy to obtain a higher optical effect by controlling pH-sensitive microgels.

In this paper, we present a new strategy for a pH-dependent reversible flocculation system consisting of a concentrated aqueous mixture of pH-sensitive ultrafine microgels and poly(L-lysine). Usually, rapid flocculation is obtained at a higher concentration of colloidal suspension by adding conventional flocculants. The reversibility of flocculation by polymers is already known;[5] however, rapid redispersion has hardly been expected, especially in a highly concentrated suspension. We have found that ionized PLL, which works as an effective flocculant, turns incompetent when its amino groups are deprotonated and rapidly completes redispersion of the pH-sensitive microgels. This property seems to have originated from the spatial shift of PLL upon its conformational transition and the simultaneous volume change of an ultrafine microgel in response to the solution pH. This system worked in a suspension of microgels whose concentration was high enough to cause a remarkable magnitude of change in absorbance, which is promising in its application to electrooptical devices.

Experimental Section

Materials. Poly(L-lysine) hydrobromide (PLL·HBr) with a molecular weight distribution of 5000-15 000 was purchased from Wako Pure Chemical Co. (Osaka, Japan) and used without further purification.

Mirogels, which were designated by microgel A and microgel N, respectively, were prepared by emulsion polymerization according to the same method as reported previously.[3] The weight ratios were MMA/AA/EGD = 20/2/0.6 for microgel A and MMA/EGD = 20/0.6 for microgel N, where MMA, AA, and EGD stand for methyl methacrylate, acrylic acid, and ethylene glycol dimethacrylate, respectively. Emal 2F (sodium dodecylsulfate; Kao-Atlas Co.) was employed as a surfactant, and an initiator was the $K_2S_2O_8/Na_2S_2O_3$ redox system. The polymerized samples were dialyzed against deionized water for 1 week. The mean diameters of microgel A and microgel N in relation to the solution pH are shown in Figure 1.

Instrumentation and the Measuring Method. The mean diameter and distribution of the diameters of the microgels were obtained by a dynamic light scattering system, DLS7000 (Ootsuka Electronics Co., Osaka, Japan), employing its standard analyzing software. Solution pH was measured with an HM-30S pH meter (TOA Electronics Co., Tokyo, Japan). Absorbance was measured by a UV spectroanalyzer Ubest-30 (Jasco Co., Tokyo, Japan). The measurement was based on its time scan mode at a wavelength of 600 nm, using a standard cell with a 5-mm light path. The circular dichroism (CD) spectra were obtained by a J600 spectropolarimeter (Jasco Co., Tokyo, Japan), using a quartz cell with a 1-mm light path. The pH of every sample solution was adjusted by adding HCl or NaOH solution. Every sample in the measuring cell was stirred by a magnetic stirrer before and during absorbance measurement.

The rate of absorbance (A) change was obtained in the following manner: 1 mL of sample was placed in the measuring cell, and

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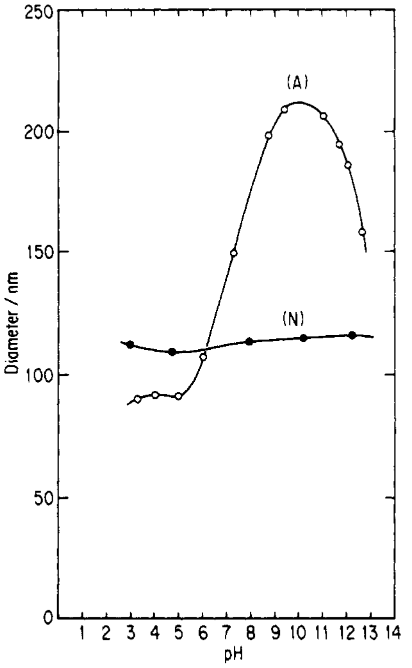

Figure 1. Mean diameters of the microgels in solution of various pH. Every solution pH was changed by adding HCl or NaOH: (-○-) microgel A; (-●-) microgel N.

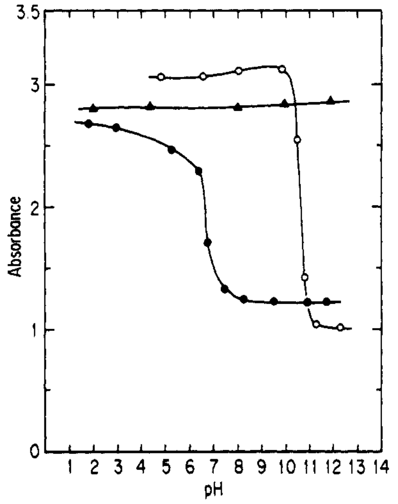

Figure 2. Relation of the absorbance of the microgel mixed with PLL to solution pH: (1; -▲-) mixture of 4.7 wt % microgel N and 0.46 wt % PLL·HBr; (2; -○-) mixture of 4.7 wt % microgel A and 0.46 wt % PLL·HBr; (3; -●-) mixture of 3.5 wt % microgel A and 0.12 wt % PLL·HBr.

0.3 mL of HCl or NaOH solution was rapidly injected. A hydrodynamically homogeneous sample solution was prepared within 0.2 s instantly after the injection. The rate of absorbance change at time $t \to 0$, $(\mathrm{d}A/\mathrm{d}t)_{t\to 0}$, was determined by extrapolating a slope of the recorded readings between 0.2 s and a few tenths of a second after the injection. ζ-Potential was measured with an electrophoretic light scattering spectrophotometer (ELS-800, Otsuka Electronics Co., Osaka, Japan) in an aqueous solution of 5 mM NaCl.

All the measurements were performed at room temperature.

Results and Discussion

Static Properties of Flocculation. In the case of the microgel mixed with PLL, three types of microgel behavior were observed in the relationship between the absorbance and the solution pH as shown in Figure 2. Microgel N, which was a stable colloidal suspension, flocculated in every pH when mixed with PLL. The high absorbance value was kept constant. On the other hand, a mixture of microgel A with PLL demonstrated a reversible flocculation that was accompanied by a remarkable absorbance change depending on the solution pH. The absorbance change can be explained by the scattering property of the particle. As the diameter of the dispersed microgel is far smaller than the wavelength of the visible light, the scattered light is very weak. According to the Rayleigh theory, the scattering cross section is proportional to the sixth power of the particle diameter. Therefore, a sharp increase in absorbance was observed upon flocculation of the microgel A, since the size of the resulting particle increased remarkably. There were two types of dispersions in the case of mixtures of microgel A and PLL. At a higher ratio of microgel A/PLL·HBr, a sharp transition in absorbance occurred at pH 10–11, while in the lower ratio a gradual transition occurred at around the neutral pH (Figure 2). The terms higher pH transition and neutral pH transition refer to the two types of transitions that occur at the respective higher and neutral pH in the following description. The effect of the microgel A/PLL·HBr ratio on the transition pH is shown in Figure 3. Here, the microgel concentration was kept constant with the changing ratio of microgel A/PLL·HBr. The transition pH changed markedly, depending on the ratio, especially from pH 7 to 10 when the ratio is 15 and 10, respectively. Variation of the gel concentration in the suspension provided little change in the transition as long as the weight ratio was kept constant. In Figure 4, a few pH transitions at higher pH were shown at various gel concentrations. Steep transitions were observed in all cases; however, the transition pH was shifted by one unit at the lowest concentration.

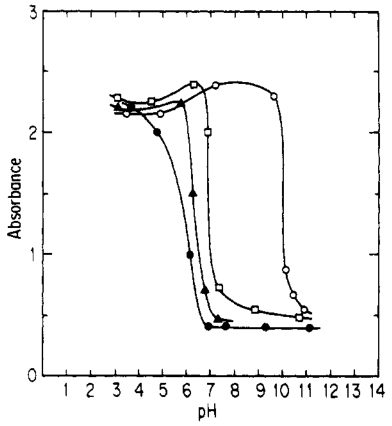

Figure 3. Effect of the microgel A/PLL·HBr ratio on absorbance at various pH. The concentration of microgel A was 1.2 wt %. The weight ratios are (1; -○-) 10.2, (2; -□-) 15.3, (3; -▲-) 30.6, and (4; -●-) 40.8.

Microgel N was negatively charged because of the employed initiator and the anionic surfactant, although no ionizable groups were contained in its component monomers. Therefore, microgel N flocculated in the presence of cationic PLL as a flocculant. The flocculated gel exhibited no redispersion, even by the neutralization of PLL at a higher pH value.

On the other hand, in the cases of microgel A, the negative charge increased due to the dissociation of the carboxyl groups in the copolymer, and the resulting increase in electrostatic repulsion among the microgels seemed to cause dispersion. However, the flocculation was maintained in the presence of a large amount of PLL until its amino groups were deprotonated at pH 10–11, since the dissociated carboxyl groups had been neutralized by cationic PLL. This flocculation is the third type, which

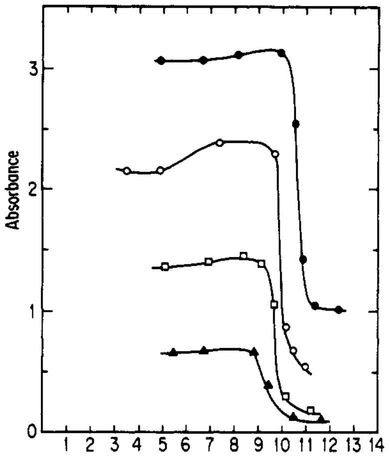

Figure 4. Effect of the amount of microgel on the absorbance change at various pH. The weight ratio of microgel A/PLL·HBr was 10.2. Concentrations of microgel A were (1; -●-) 4.7 wt %, (2; -○-) 4.7/4 wt %, (3; -□-) 4.7/12 wt %, and (4; -▲-) 4.7/24 wt %.

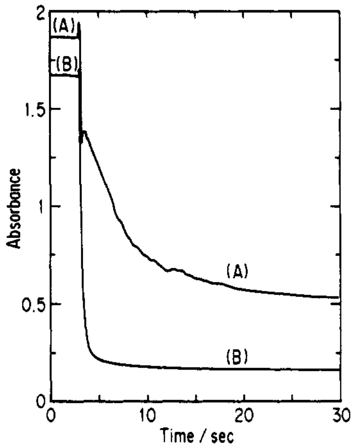

Figure 5. Time course of absorbance on jumped pH change. Two types of dispersion at different pH were observed. (A) The solution pH was changed from pH 5.2 to pH 8.0. The microgel A/PLL·HBr ratio was 20.6. (B) The solution pH was changed from pH 9.5 to pH 12.1. The microgel A/PLL·HBr ratio was 10.3.

accompanies a remarkable dynamic property as described in the following section.

Dynamic Properties of Flocculation. In the flocculation of microgel A, the rate of absorbance change, $\mathrm{d}A/\mathrm{d}t$, in higher pH transition was much larger than that in the neutral pH transition. An example of the difference between the two transitions is shown in Figure 5. It is clearly seen that the rate of absorbance change caused by pH jumping from pH 9.5 to pH 12.1 was more rapid than that from pH 5.2 to pH 8.0.

The rate of absorbance change in the dispersion and flocculation, $|\mathrm{d}A/\mathrm{d}t|_{t\to 0}$, was plotted against the microgel concentration (n), and the result is shown in Figure 6. In the flocculating process, $|\mathrm{d}A/\mathrm{d}t|_{t\to 0}$ increased with the microgel concentration, as observed in the general flocculation process. In dispersion, $|\mathrm{d}A/\mathrm{d}t|_{t\to 0}$ showed a maximum value at a certain microgel concentration. The maximum value, 6 absorbances/s, in the measuring cell with a 0.5-cm light path corresponds to 100 times the change of transmittance in the 50-μm light path happening in 10 ms. This dynamic property seems comparable to that of conventional liquid crystal devices, provided such abrupt pH change is carried out by an electrical method. Moreover, it is expected that a real extrapolated value of $|\mathrm{d}A/\mathrm{d}t|_{t\to 0}$ is much larger than that determined by the manual injection method as was mentioned above.

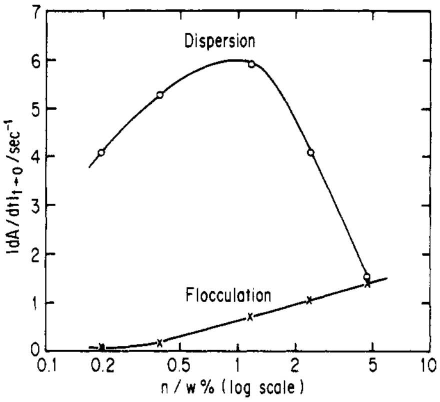

Figure 6. Rate of absorbance change $|\mathrm{d}A/\mathrm{d}t|_{t\to 0}$ vs the concentration (n) of microgel A. In dispersion, pH was jumped from 9.5 to 11.5–12.0. In flocculation, pH was jumped from 12.0 to 8.0–8.5. The weight ratio of microgel A/PLL·HBr was 10.3.

Reversibility of the dispersion at the higher pH transition was investigated by measuring the diameter of the particles. As shown in Figure 7, the diameter distribution in the histogram B reverted to that of the initial state unipeak distribution (A) after a cycle of flocculation and redispersion, leaving a small amount (3.7 wt %) of the flocculate. Most of the remaining flocculate was eventually dispersed by gently mixing the sample by inversing the measuring tube, as shown in the histogram C.

Mechanism of Reversible Flocculation. Two mechanisms of flocculation, interparticle bridging and charge neutralization, are commonly accepted for the colloidal suspension with polymeric flocculants.[6] The properties of the present system differ from those of conventional ones in that pH-dependent changes in charge and volume of the microgel as well as pH-induced conformational change of the flocculant polymer play indispensable roles. The ζ-potential of microgel A was −17.2 and −39.5 mV at pH 3 and 7, respectively. The ζ-potential of a mixture of microgel A and PLL·HBr whose weight ratio was 10.2 was −0.8 mV at pH 7 and −39.5 mV at pH 11.5. The mixture exhibited the higher pH transition as shown in curve 1 of Figure 3. The cationic PLL chains adsorbed on the negatively charged surface of the microgel, and part of them simultaneously adsorbed on both surfaces of two microgels to form bridging. These data suggested that the increased charge of the microgel due to the dissociation of the carboxyl groups was neutralized by the adsorption of the cationized PLL, while at around pH 11.5 the dispersed microgel regained its fully dissociated negative charge because of neutralization (deprotonation of the amino groups) of the counterion PLL.

In the case of a sample of microgel A with PLL·HBr whose weight ratio was 15.3, the ζ-potential was −13.6 mV at pH 7. The sample exhibited a transition at around neutral pH (curve 2 in Figure 3). At the weight ratio the amount of PLL was insufficient to neutralize the microgel, so that the microgel was gradually dispersed by an electrostatic repulsion force depending on the extent of dissociation of its carboxyl groups. Thus, the dispersion was related to the charge of the microgel, which was varied by the dissociation of the carboxyl groups and the deprotonation of the amino groups of the adsorbed PLL.

The reversible flocculation was attributed to steric repulsion due to the overlapping of the PLL segment, which prevented particles having less charge from dropping into the minimum of the DLVO-type potential. The steric force commences at a surface separation of ca. 6 R_g, where

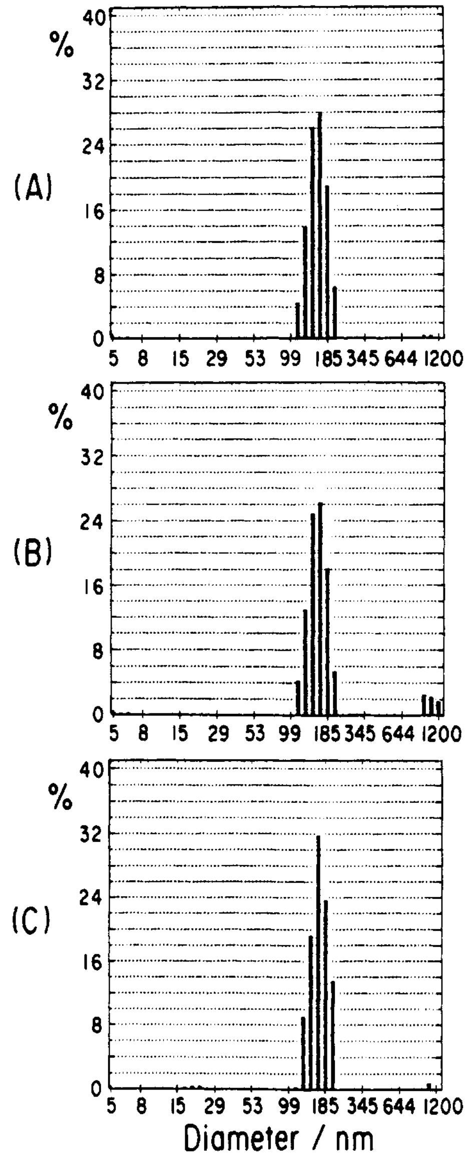

Figure 7. Reversibility of the particle diameter distribution. The weight ratio of microgel A/PLL·HBr was 10.3. (A) Initial sample at pH 12.0. (B) Measured immediately after a series of dispersion (pH 12), flocculation (pH 8), and redispersion (pH 12). (C) Measured after mixing of (B) by inversing the measuring tube.

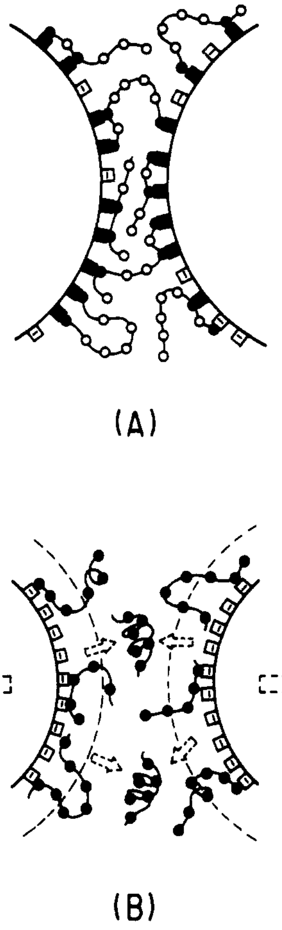

Figure 8. Schematic representation of two microgel surfaces (A) in flocculation and (B) in dispersion: (○) $-NH_3^+$ of PLL, (⊟) $-COO^-$ of MMA/AA copolymer, (■●) ion pair of $-COO^-$ and $-NH_3^+$, (●) $-NH_2$ of PLL (deprotonated amino group).

R_g is the radius of gyration of the concerned polymer.[7] Six R_g of PLL·HBr of molecular weight 15 000 is ca. 18 nm, while the length in the fully stretched form is ca. 26 nm.[8] A part of the polymer segments extended in solution may form bridges between the particles, which induce attraction, whereas the steric interaction may prevent particles from contacting more closely with each other. It is known that circular dichroism (CD) at 222 μm is a suitable index of the α-helix conformation, which is the case of PLL. The molecular rotation $[\theta]_{222}$ was -8.1×10^3 (deg·cm²/dmol) at pH 11.5 for a PLL·HBr solution whose concentration was the same as that of sample 1 in Figure 3. Meanwhile, $[\theta]_{222}$ was -6.6×10^3 at pH 11.5 for the sample with the microgel. In this case the mole concentration of PLL·HBr was corrected by taking the turbidity of the microgel at 222 μm into consideration. (The uncorrected value was -5.7×10^3.) The sample was flocculated by varying down to pH 8, and then the resulting supernatant solution was readjusted to pH 11.5. $[\theta]_{222}$ of this transparent solution was -2.1×10^3. Since molecular rotation $[\theta]_{222}$ is presumed to indicate the PLL concentration as long as the pH was above 11.5, the above values, -2.1×10^3 and -6.6×10^3, were compared to estimate the amount of the desorbed PLL. The difference between the rotation index for the flocculated state (-2.1×10^3) and that for the dispersed state (-6.6×10^3) can thus be regarded as the amount of the desorbed PLL (-4.5×10^3). This calculation was based on the assumption that no part of the microgel-adsorbed PLL takes the α-helix conformation. In the process of redispersion, the difference (-4.5×10^3) indicates that a considerable amount of the adsorbed PLL was released from the gel surface, while the rotation value for the supernatant solution corresponds to free PLL. It is hardly considered that rapid detachment of PLL from the charged microgel is due merely to the deprotonation of PLL. The detachment seems to be attributed to the cooperative actions of the following factors: the increase in the interparticle electrostatic repulsion force upon deprotonation of the adsorbed PLL, the enthalpy change upon conformational transition of PLL from random coil to α-helix, and the pH-dependent volume change of the microgel. Response time of these pH-dependent factors is reported to be less than a millisecond.[1,9] The instantaneous spatial shift, which is the concurrence of the conformational transition of the bridging polymer and the shrinkage of the microgels, seems to cause rapid and reversible redispersion. A postulated mechanism is schematically depicted in Figure 8.

For further detailed study on the dispersion, in addition to the discussion from the standpoint of the thermodynamics, a more strict method for measuring $|dA/dt|_{t\to 0}$, such as a pH-jump stopped-flow method, will be required. Electrical modulation of the absorbance with the use of a pH modulator is expected to bring the new flocculation system into practical use for electrooptical devices. The authors believe that the new system will be promising as an alternative to the present liquid crystal systems, in case other evaluation factors such as endurability are satisfactory.

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Registry No. PLL, 25104-18-1; (MMA)(AA)(EGD) (copolymer), 34439-19-5.