

Complex Formation between a Fluorescently-Labeled Polyelectrolyte and a Triblock Copolymer

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The association between a randomly pyrene labeled PAA polymer (PAAMePy55) and a PEO–PPO–PEO triblock copolymer (P123) in aqueous solutions of different NaCl concentrations and pHs has been studied by means of dynamic light scattering (DLS) and steady-state fluorescence spectroscopy at 40 °C. At acidic pH values, in the low P123/PAAMePy55 molar ratio regime (i.e., at low P123 concentrations), the relaxation time distributions retrieved from the DLS data analysis were monomodal and very similar to those obtained for the pure PAAMePy55 solution. The apparent hydrodynamic radius of PAAMePy55 at low pH is 18 nm. At higher molar ratios (i.e., at high P123 concentrations), still in the acidic pH regime, bimodal relaxation time distributions were obtained, where the fast relaxation mode is connected to the translational diffusion of free P123 micelles with a hydrodynamic radius obtained at infinite P123 dilution ($R_{H,P123=0}$) of 10–11 nm. This value coincides perfectly with the hydrodynamic radius of the pure P123 micelles at 40 °C, which was found to be ca. 10 nm at all pH values. The second mode corresponds to a complex consisting of one PAAMePy55 polymer chain and about 42 P123 micelles and with a $R_{H,P123=0}$ between 35 and 36 nm depending on pH. At pH 9, the mixed system also presented bimodal relaxation time distributions. At this high pH, the intermolecular association between PAAMePy55 and P123 is less strong than at acidic pH according to the steady-state fluorescence measurements. The fast mode is also in this case attributed to free P123 micelles whereas the second mode is related to the so-called “slow mode” commonly observed for polyelectrolyte solutions. In this system, it is related to the formation of multichain domains, that is, large domains formed by several PAAMePy55 chains that move in a common electrostatic field (i.e., a structure factor effect). The presence of P123 micelles does not lead to the total disruption of these domains. They may either contain entrapped P123 micelles or hydrophilic diblock impurities (originating from the P123 sample) that associate with the PAAMePy55 chains.

Introduction

The interaction between water-soluble polymers and surfactants is one of the most important and challenging field in the domain of colloidal science mostly because of its impact in industrial applications.^{1,2} Two of the main driving forces for the interaction are associated with the electrostatic and hydrophobic characteristics of the interacting polymer and the surfactant molecules. The polymer–surfactant interaction usually results in the formation of pearl-necklace structures (i.e., polymer chains decorated by surfactant micelles) that occur at lower surfactant concentrations than the critical micelle concentration (cmc) of the surfactant alone.^{1–5} If the water-soluble polymer carries charges along its chain, it is called a polyelectrolyte. Polyelectrolytes combine unique properties of polymers and electrolytes; in solution, they are dissociated into polyvalent macroions (or polyions) and a large number of small ions of opposite charge (i.e., counterions).⁶ The high charge of the polyion produces an electrical field, which attracts the counterions producing the physical characteristics of an electrolyte.⁶ The conformation of an anionic polyelectrolyte in solution is

strongly dependent on its degree of ionization (α) and interaction with the counterions. In the limit, it changes from a compact random coil ($\alpha = 0$) to a significantly expanded conformation at $\alpha = 1$. The degree of ionization has also been shown to have a great influence on the interaction between polyelectrolytes and surfactants and a large number of studies have thus been performed, where the interaction with both ionic^{3,4} and nonionic surfactants^{5,7–9} has been investigated. In both cases, the hydrophobic and electrostatic interactions showed to be of major importance.

The interaction between negatively charged polyelectrolytes, e.g. poly(acrylic acids), and oppositely charged surfactants is driven by electrostatic attractive interactions. On the other hand, the polyelectrolyte–nonionic surfactant association is driven by hydrophobic interactions at all pH values.^{5,7,8,10} In the nonionic form (low pH), the compact coil is a preferential site of adsorption and a strong interaction between the polymer and the nonionic surfactants is observed. When the pH is increased, the polyelectrolyte chain expands due to the intrapolymer electrostatic repulsions, its hydrophobicity decreases and for this reason the interaction with the surfactant is strongly decreased.^{5,7,8} However, even at low pH values, where the interaction is likely to occur, the interaction has shown to be largely dependent on the molecular weight of both the surfactant and the polyelectrolyte.

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Although the interaction between polyelectrolytes and non-ionic surfactants has been subject of detailed studies, few studies have focused on the interaction between polyelectrolytes and triblock copolymers of poly(ethylene oxide) (PEO) and poly(propylene oxide) (PPO). The triblock copolymers composed of a middle block of PPO and two PEO end blocks (PEO–PPO–PEO) are available with varying chain length and block length ratio. Similarly to surfactants, the PEO–PPO–PEO block copolymers self-assemble in water, which is a thermodynamically good solvent for the PEO groups but a poor solvent for the PPO groups.^{11–15} In solution, they can exist as single copolymer chains (unimers), and upon increasing concentration (or temperature), micelle structures commence to be formed.^{11,12,16–18} The resultant micelle consists of dehydrated PPO blocks in the core surrounded by a water-swollen (hydrated) corona of PEO blocks as revealed by small-angle neutron and X-ray scattering measurements,^{19–25} see also, e.g., the review in ref 26. The micelle formation is mainly governed by the PPO block length (the dependence on the PEO block length is less pronounced), the total molecular weight of the copolymer (for a constant PPO/PEO ratio), and the copolymer concentration.^{11,13,27–29} The micellization temperature, as obtained from differential scanning calorimetry (DSC), decreases with the PPO block length.^{28–30} The transition temperature decreases also with increasing copolymer concentration^{11,13,29,30} or, expressed differently, cmc is very dependent on temperature.

Owing to the polydispersity of these copolymers (both chemical and molar mass heterogeneity), the unimer-to-micelle transition occurs over a broad temperature range, i.e. with a much less sharp critical micelle temperature (cmt) or cmc than ordinary surfactants. This has been demonstrated by dynamic light scattering (DLS),^{17,18,31} ultrasonic speed measurements,²⁰ DSC,^{20,30} NMR,³² and model calculations.^{33,34} In this region, micelles of a defined size coexist with unimers. Furthermore, Hvidt et al.^{30,35} have demonstrated that the PEO–PPO–PEO copolymer samples contain lower size contaminants (hydrophilic PEO–PPO diblock copolymers), which are not included into the micelles, and if these impurities are removed by fractionation using interaction chromatography, the unimer-to-micelle transition sharpens in temperature. The effect of these diblock impurities on the structure of the cubic liquid crystalline phases of the triblock copolymer F127/water system has recently been reported.³⁶ These may also play an important role in the polymer system studied in this work.

The intermolecular interaction and complex formation between poly(carboxylic acids), such as poly(acrylic acid), and their corresponding salts (polyacrylates) and PEO have been studied thoroughly in the literature. According to these investigations, which discuss the association as driven by hydrogen bonding, the stability and structure of the interpolymer complexes formed depends on the pH, the ionic strength, the molecular weight, and whether the polymers carry hydrophobic groups.^{10,37,38} As we have discussed previously, in aqueous solutions, hydrogen bonding is not a driving force for association.³⁹ On the other hand in a hydrophobic environment, it can play an important role on the association. Certainly, the interplay between electrostatic and hydrophobic interaction forces must also be present in these systems in addition to the hydrogen bonding.

The aim of the present study is to investigate the intermolecular interaction between a PAA polymer labeled with pyrene (PAAMePy55) and the EO₂₀PO₆₈EO₂₀ block copolymer (P123) by means of DLS. This follows the previous study where the same system was investigated by fluorescence techniques.³⁹ The

micelle formation of P123 and the interaction with both ionic (SDS and CTAC) and nonionic (C₁₂EO₆) surfactants were previously investigated by our group using differential scanning and titration calorimetry, static and dynamic light scattering, small-angle X-ray scattering, NMR-diffusiometry, and rheology.^{25,29,31,40–43} We will show that it is very fruitful to combine DLS and fluorescence spectroscopy in order to analyze in detail the intermolecular interactions in this polyelectrolyte–block copolymer system under various physical conditions such as NaCl concentration and pH.

Experimental Section

Polymers and Sample Preparation. Poly(acrylic acid) (PAA) with a nominal molecular weight of 150 000 g/mol was purchased from Wako Chemicals as a 25% water solution. The pyrene-labeled PAA sample was kindly provided by Dr. Dan Anghel, and the synthesis followed the same route as described elsewhere.⁴⁴ The Py content of labeled PAA corresponds to 55 monomers per chromophore per polymer chain. This sample is denoted PAAMePy55. The PEO–PPO–PEO triblock copolymer was a kind gift from BASF Corporation, Performance Chemicals, Mount Olive, NJ, and used without further treatment. The copolymer has the average composition EO₂₀PO₆₈EO₂₀ (denoted P123, where P stands for “paste” and 3 for 30 wt % of PEO) and a nominal molar mass of 5750 g/mol. The P123 sample is polydisperse both in mass and composition. The GPC characterization performed in ref 31 showed that 10% of the material corresponded to low-molar mass components. In a recent investigation of the heterogeneity of these copolymers using size exclusion chromatography, it was found that the P123 copolymer sample may contain up to 26 wt % of smaller-size diblock impurities.³⁵ Water purified by a Milli-Q system (Millipore Corporation, Bedford, MA) was used in all solutions used for the light scattering experiments.

The aqueous PAAMePy55 and P123 stock solutions were prepared and left to equilibrate overnight; the block copolymer solutions were prepared in a refrigerator at ca. 6 °C. The PAAMePy55 stock solutions were prepared at different pH values using water that was previously filtered through 0.2 μm filters from Whatman to exclude dust particles. The polyelectrolyte solutions were thereafter centrifuged at 3600 rpm for 20 min to further decrease the amount of dust. The P123 stock solutions prepared at different pH values were filtered cold through 0.2 μm filters from Whatman. The PAAMePy55–P123 solutions were mixed and left to equilibrate in the refrigerator a second night. All solutions were prepared by weighing. Prior to the measurement, the solution was equilibrated at room temperature for at least 20 min to avoid temperature jumps over the cmt of P123³¹ and, thereafter, it was equilibrated at the measuring temperature for at least 20 min. All measurements were performed at 40 °C, which was controlled to within ±0.01 °C by a F32 Julabo heating circulator.

Dynamic Light Scattering. The setup used for the DLS measurements is an ALV/DLS/SLS-5000F, CGF-8F based compact goniometer system from ALV-GmbH, Langen, Germany. A more detailed description about the instrumentation can be found in ref 31 with the difference being that *cis*-decahydronaphthalene is used as refractive-index-matching liquid. The laser is a CW frequency-doubled diode-pumped Nd:YAG solid-state Compass-DPSS laser from COHERENT, Inc., Santa Clara, CA, that operates at a wavelength (λ) of 532 nm with a fixed output power of 400 mW, which can be varied using an attenuator from Newport Inc. The DLS measurements were performed at different scattering angles (θ = 50°–130°)

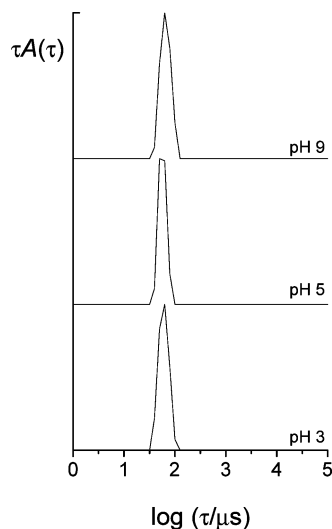


Figure 1. Relaxation time distributions obtained by regularized inverse Laplace transformation of the corresponding intensity correlation functions from DLS for the 1 wt % P123 block copolymer at three different pH values (pH 3, 5, and 9) at 40 °C and measured at $\theta = 90^\circ$.

This corresponds to the q range of 1.33×10^7 – $2.85 \times 10^7 \text{ m}^{-1}$, where q is the magnitude of the scattering vector, which in case of dilute systems is given by $q = 4\pi n_0 \sin(\theta/2)/\lambda$, in which n_0 is the refractive index of the solvent (in this case water, $n_0 = 1.332$). The output of the DLS experiment is the normalized time correlation function (pseudocross or auto) of the scattered intensity, $g^{(2)}(t)$ (or rather $g^{(2)}(t) - 1$). The normalized electric field correlation function $g^{(1)}(t)$, which is related to $g^{(2)}(t)$ by Siegert's relation, of a solution of polydisperse particles may be expressed as a Laplace transformation of the relaxation time distribution:^{45,46}

$$g^{(1)}(t) = \int_0^\infty \tau A(\tau) \exp(-t/\tau) d \ln \tau \quad (1)$$

where $\Gamma = \tau^{-1}$ is the relaxation frequency or rate, which is obtained from the first moment of the distribution.

A nonlinear regularized inverse Laplace transformation procedure, REPES,^{45,47} was used to directly analyze the experimental $g^{(2)}(t) - 1$ functions to obtain the distribution of relaxation times, $A(\tau)$, where τ is the relaxation time. In this study, the relaxation time distributions are presented as $\tau A(\tau) = \log(\tau/\mu\text{s})$ for equal-area representation.⁴⁸

Steady-State Fluorescence. Fluorescence spectra were recorded on a Jobin-Ivon SPEX Fluorog 3-22 spectrometer. The fluorescence spectra were corrected for the wavelength response of the system. The solutions for the fluorescence experiments were prepared in a similar way as for the DLS experiments with the exception of the filtering and centrifuging processes.

Results

Dynamic Light Scattering. P123 Block Copolymer in Aqueous Solution. Dynamic light scattering measurements were performed on 1 wt % P123 block copolymer solutions at three different pH values (3, 5, and 9) and at 40 °C. The relaxation time distributions [$\tau A(\tau)$ vs $\log(\tau/\mu\text{s})$], obtained from the regularized inverse Laplace transformation of the measured time correlation functions of the scattered intensity are presented in Figure 1. As can be observed from Figure 1, the relaxation time distributions consist of a narrow and well-defined peak. The

relaxation rate Γ of this single relaxation mode shows a linear dependence with the square of the magnitude of the scattering vector q^2 (data not shown). This indicates that this mode corresponds to the translational diffusion process of the P123 micelles. The P123 concentration used is far above the cmc values of the P123 at 40 °C, which are obtained from DSC^{29,31} and fluorescence³⁹ measurements are $0.2 \times 10^{-3} \text{ wt } \%$ ($3.48 \times 10^{-7} \text{ M}$) and $1.8 \times 10^{-6} \text{ M}$, respectively. The apparent mutual diffusion coefficient (D) of the P123 micelles can thus be obtained from the slope of Γ vs q^2 according to the following:⁴⁹

$$D = \left(\frac{\Gamma}{q^2} \right)_{q \rightarrow 0} \quad (2)$$

D is concentration dependent both due to hydrodynamic and intermolecular interactions. The diffusion coefficient at infinite dilution at $c_{\text{P123}} = 0$, D_0 (with its associated error), can be estimated from a diagram of D as a function of the P123 concentration c_{P123} in each case (data not shown). The hydrodynamic radius of the P123 micelles at infinite dilution is thereafter directly obtained by the Stokes–Einstein equation:

$$R_H = \frac{k_B T}{6\pi\eta_0 D_0} \quad (3)$$

where k_B is Boltzmann's constant, T is the absolute temperature, and η_0 is the viscosity of the solvent ($\eta_0 = 0.6449 \times 10^{-3} \text{ Pa s}$ for water at 40 °C).

The obtained hydrodynamic radius of the P123 micelles, at 40 °C, was found to be about 10 nm at all pH values (i.e., $10.8 \pm 0.3 \text{ nm}$ at pH 3, $9.8 \pm 0.1 \text{ nm}$ at pH 5 and $9.9 \pm 0.1 \text{ nm}$ at pH 9, where the error is given as one standard deviation (σ) estimated from diffusion-coefficient determinations at different angles). With intensive light scattering studies of the P123 block copolymer, performed at different concentrations and temperatures, similar values of the hydrodynamic radius were obtained at 40 °C (the discrepancies are due to the different batches used).^{31,42} The presence of inorganic salts, such as NaCl, are known to reduce the cmc and the cmt, as well as to induce the micelle-to-rod transition, i.e., to induce the micellar growth, due to the dehydration of the PEO blocks leading to a decrease in the spontaneous curvature of the block copolymer film. However, in our study the presence of salt (1.4 and 10 mM of NaCl, at acidic and basic pH values, respectively) does not affect the size of the P123 block copolymer micelles. Recent studies on the influence of NaCl concentration on the sphere-to-rod transition, of the P123 block copolymer (1 wt %) with 10% ethanol, revealed that the apparent translational diffusion coefficient decreases from $\sim 2.7 \times 10^{-11}$ to $\sim 2.5 \times 10^{-11} \text{ m}^2/\text{s}$ when the NaCl concentration increases from 0 to 0.5 M.⁵⁰ This corresponds to an increase of $\sim 1 \text{ nm}$ in the hydrodynamic radius of the micelle. More recently, Denkova et al.⁵¹ studied the effect of several inorganic salt (KF, KCl, KI, LiCl, and CsCl) on the P123 micelle structure in solution. LiCl and KI showed to have little influence on the micellar structure when compared with the other salts where sphere-to-rod transition was shown to be promoted by their presence in solution. However, the concentrations used in both studies are well above the concentrations used in our study [2 M vs 0.001 (at pH 3–5) or 0.01 M (at pH 9)]. Therefore, we can infer that for the low salt concentrations used in this study no significant increase would be expected in the R_H .

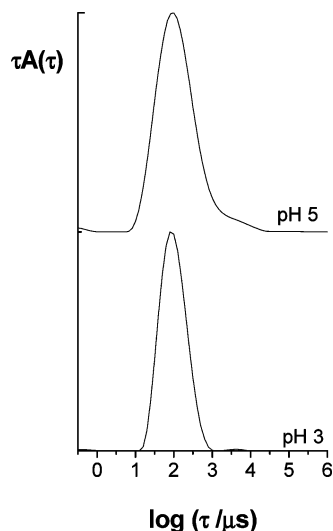


Figure 2. Relaxation time distributions for 0.20 wt % of PAAMePy55 at pH 3 (bottom curve) and 5 (top curve) in the presence of 1.4 mM of NaCl. Measurements at 40 °C and at $\theta = 110^\circ$.

PAAMePy55 Polymer in Aqueous Solution. The aqueous PAAMePy55 polymer solutions were also studied at three different pH values (3, 5, and 9) and at 40 °C by using DLS. The intensity correlation functions obtained for the salt-free PAAMePy55 solutions were multiexponential, and the relaxation time distributions obtained from the Laplace inversion analysis were therefore multimodal. The analyses were carried out with carefulness considering the difficulties of this type of calculations. However, the REPES algorithm used here is more robust than that of CONTIN.⁴⁵ At all pH values, although more pronounced at intermediate and high pH, two main modes were obtained (data not shown): a fast mode in addition to a slow mode (with a large amplitude) positioned at higher relaxation times. On the basis of the q^2 dependences of Γ (see eq 2), the fast and slow modes can be attributed to two different translational diffusive processes. These modes are characteristic for DLS results on polyelectrolyte solutions.⁵² In previous studies, the fast mode has been attributed to the coupled diffusion of a polyion and counterions in a common electrostatic field where the polyion experiences an electric force due to the concentration fluctuations of the surrounding counterions.^{53–55} The translational diffusive motion as measured by DLS will therefore become faster due to the polyion–counterion interactions.⁵⁶ Furthermore, the apparent hydrodynamic radius $R_{H,app}$ obtained from the slow diffusive mode (using eqs 2 and 3) confirms that this translational mode cannot be due to the diffusion of a single PAAMePy55 chain, but instead to the diffusion of several PAAMePy55 chains that move together forming large domains in the solution (i.e., a static structure factor effect) as the $R_{H,app}$ value obtained is much larger than that of the single chain. This multimodal behavior is very common in highly charged polyelectrolyte systems and has been extensively studied by Sedláč, ^{53–55,57–66} who also showed that the presence of low molecular weight electrolytes has a strong effect on the obtained relaxation time distributions.

In the PAA system at low pH values, the polyelectrolyte is less charged (α is low) and the slow mode effect is therefore less pronounced than at pH 9 (as will be shown below in Figure 3). Figure 2 shows the relaxation time distributions of PAAMePy55 polymer in the presence of 1.4 mM NaCl at pH 3 and 5. As can be seen, the distributions are dominated by one main peak and the amplitude of the slow mode has strongly decreased

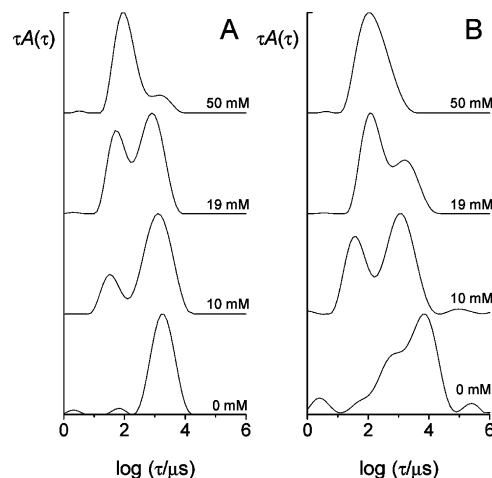


Figure 3. Relaxation time distributions for (A) 0.20 wt % PAA and (B) 0.20 wt % PAAMePy55 at pH = 9 with different NaCl concentrations (ranging from 0 to 50 mM). Measurements at 40 °C and at $\theta = 110^\circ$.

as compared to the situation in the absence of NaCl (see Figure 3 below). This is the evidence that the domains have been almost totally vanished. The relaxation rate of the main mode shows a linear q^2 dependence and corresponds to the translational diffusion of the single PAAMePy55 polymer chain with an $R_{H,app}$ value of approximately 18 nm [$R_{H,app} = 18 \pm 2$ nm ($\pm\sigma$), at acidic pH values]. A similar value ($R_{H,app} \sim 20$ nm) was found by Fundin et al.⁴ for a polyacrylate chain, with $M_w = 90$ kg/mol, in the presence of 20 mM of NaBr.

In Figure 3, we compare the relaxation time distributions of the unlabeled PAA in water at pH 9 and at various ionic strengths with those of the pyrene-labeled PAA. Under salt-free conditions and low ionic strengths, both systems display complex multimodal relaxation time distributions. The fast and the slow modes are, as described above, the result from the coupled diffusion of the polyion and the counterions and from the formation of multichain domains, respectively. These latter structures present $R_{H,app}$ values in the range of 80–200 nm (depending on the NaCl concentration). An estimation of the hydrodynamic radius of a fully ionized PAA chain of 150 000 g/mol at pH 9 and at low ionic strength, gives 42–50 nm depending on the geometrical shape used (semitorus or torus), which suggests that these domains are composed of several chains.⁶⁷ Increasing the salt concentration from 10 to 20 mM causes a shift in the relaxation time of the two modes: the slow diffusion process becomes faster and the fast process slower. As the salt concentration increases up to 50 mM, the charges of the PAA polymer chain are almost totally screened, leading to a more compact polymer conformation. Although the slow mode is still present in the relaxation time distribution, it loses weight and the “uncharged” PAA chain is the main scattering object in solution. It is worth noting that in the absence of NaCl, a third middle mode was also obtained, which could be related to the “interaction mode” that Sedláč has described to be present in a two-polymer system (a polydisperse system as is the case of that presently studied).^{55,58,59} However, we do not wish to further pursue this issue since the regularized inverse Laplace transformation procedure REPES could still produce small peaks that are artifacts.

The same behavior as the unlabeled PAA system at pH 9 is observed for the PAAMePy55 system at pH 9 (Figure 3B). At 10 mM concentration of NaCl, the relaxation time distribution shows two main modes (both with a q^2 dependence) corre-

sponding to the $R_{H,app}$ values of $7 (\pm 0.9)$ nm and $135 (\pm 20)$ nm, respectively. These modes are most likely the characteristic fast and slow modes of the PAAMePy55 system at pH 9 (similar to the unlabeled PAA system). This appearance of a bimodal distribution can thus be considered the “fingerprint” of the PAAMePy55 system and will be used in the forthcoming discussion of the mixed PAAMePy55–P123 system. Furthermore, we believe that the minor difference in relative amplitude observed in the relaxation time distributions of the PAAMePy55 and the PAA polymers is clearly connected to the hydrophobic labeling of the former polymer. Indeed, it seems clear from the above results that the hydrophobic interactions between the pyrene polymer groups, together with the screening of the electrostatic interactions due to the presence of NaCl, results in an even more compact conformation in the case of PAAMePy55 than in the case of the untagged PAA. At 50 mM NaCl, the two modes merge into a single one, and monomodal relaxation time distributions are obtained. This single mode corresponds to the translation diffusion of a single PAAMePy55 chain with a $R_{H,app}$ of 21.0 ± 0.9 nm ($\pm \sigma$).

This value is very similar to that obtained for the corresponding PAAMePy55 polymer at acidic pH values, i.e. 18 nm (see Figure 2). Therefore, we may conclude that the screening of the electrostatic interactions by increasing the salt concentration leads to the disruption of the PAAMePy55 multichain domains and hence a less pronounced structure factor.

PAAMePy55–P123 Mixed System. After the investigation of the pH dependence and the response to simple salt addition of the P123 block copolymer and PAAMePy55 polyelectrolyte neat systems, DLS experiments were carried out on the PAAMePy55–P123 mixed system. A small amount of NaCl was used to avoid further complications related to the electrostatic interactions as described above, but still not too much to screen the interactions between the charges within the polyelectrolyte chains. The NaCl concentration used in all experiments was 1.4 mM at pH 3 and 5 and 10 mM at pH 9.

In the low P123 concentration regime (low P123/PAAMePy55 molar ratios, $n_{P123}/n_{PAAMePy55} = MR$) the relaxation time distributions were, at all investigated pH values, very similar to those obtained for the pure PAAMePy55 solution (with similar $R_{H,app}$ values) (data not shown). However, due to the limited sensitivity of the DLS method, an association between the P123 unimers and the PAAMePy55 polymer chain cannot be excluded in this region. In the high molar ratio regime, on the other hand, where low I_E/I_M ratio values are obtained from fluorescence measurements (ref 39; see also Figure 8 below), the intermolecular association is clearly seen in the DLS results. The measurements in this regime were performed using four different sets of samples, each with a fixed PAAMePy55 concentration and with varying P123 concentration from ~ 1.5 to ~ 4 wt %. The PAAMePy55 concentrations used were 0.005, 0.010, 0.015, and 0.02 wt %. The relaxation time distributions obtained for the 0.015 wt % PAAMePy55 solution with varying P123 concentration at pH 3, 5, and 9 are presented in Figure 4. Also included in the figures are the distributions of the pure P123 and PAAMePy55 solutions at each pH, respectively.

At low pH values, the relaxation time distributions show two diffusive modes; the q^2 dependences of the fast and slow modes were analyzed in the angular range from 70° to 130° (see Figure 5 for the pH 3 case). The corresponding translational diffusion coefficients were obtained from the slopes (i.e., by application of eq 2) and the $R_{H,app}$ values were calculated from eq 3.

Comparing the relaxation time distributions of the PAAMePy55–P123 system with those of the pure P123 micellar solutions

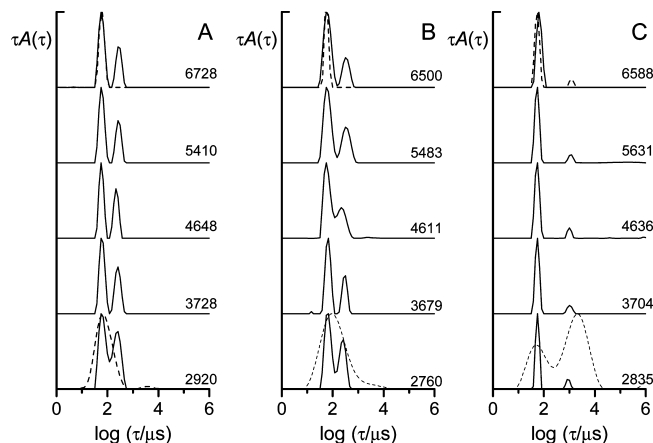


Figure 4. High P123/PAAMePy55 molar ratio regime at 40 °C. Relaxation time distributions for molar ratios ranging from ~ 2700 to ~ 6728 : (A) at pH 3 (and 1.4 mM NaCl), (B) pH 5 (and 1.4 mM NaCl), and (C) pH = 9 (and 10 mM NaCl). In terms of concentration, the presented relaxation time distributions correspond to a constant PAAMePy55 concentration (0.015 wt %) with varying P123 concentration (from 1.5 to 3.5 wt %). The dashed lines correspond to the distributions of P123 in NaCl solution (P123 micelles, 1.74 mM, top) and of PAAMePy55 in NaCl solution (0.20 wt%, bottom) at pH 3, 5, and 9, respectively. Measurements are at 40 °C and at $\theta = 90^\circ$.

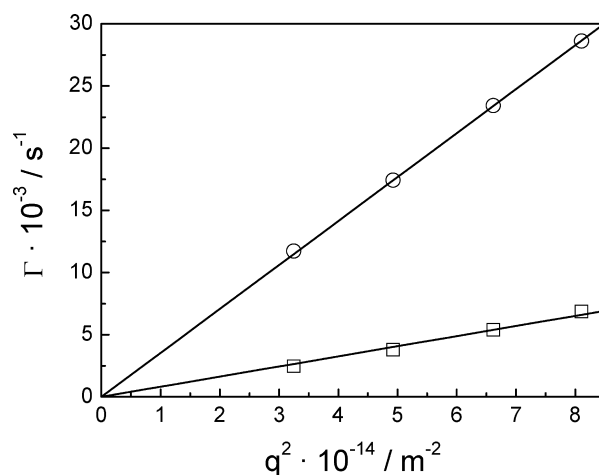


Figure 5. Dependence of the relaxation rates on the magnitude of the scattering vector squared for 0.015 wt % PAAMePy55 in 1.4 mM NaCl with 3.0 wt % P123 (MR = 5410) at pH = 3: (O) fast mode, attributed to the P123 micelles; (□) slow mode, attributed to the PAAMePy–P123 complexes.

in Figure 4, we observe that the fast mode of the mixed system completely overlaps the relaxation time distribution of the pure P123. Hence, this mode corresponds to the translational diffusion of the free P123 micelles in the mixed solution. The data presented in Figure 5 give $R_{H,app} = 10.0 \pm 0.1$ nm for the fast mode, where the error is given as one standard deviation. Another observation is that the second (slower) mode of the mixed system does not completely overlap with the relaxation mode of the pure PAAMePy55 polymer at low pH, i.e., the former is positioned at longer times than the latter (compare Figure 2 with Figure 4A and B). This decrease in the diffusion coefficient of the PAAMePy55 polymer chain in the mixed case at low pH means that there is an increase of the PAAMePy55 hydrodynamic radius. Hence, we interpret this second mode to be associated to a complex formed between PAAMePy55 and P123. For a fixed PAAMePy55 concentration (0.015 wt %), at pH 3, the corresponding $R_{H,app}$ varies between 37.6 ± 0.4 and 45 ± 3 nm depending on the P123 concentration. These values

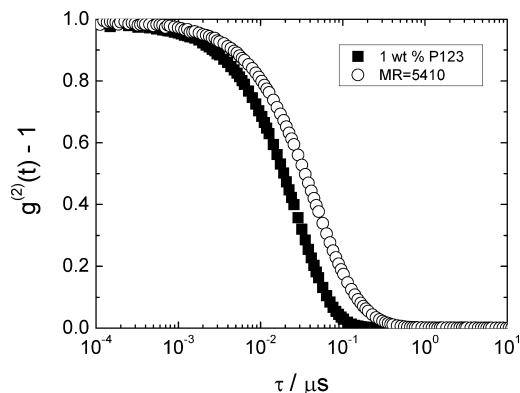


Figure 6. Intensity correlation functions of the pure P123 solution and of the mixed system (MR = 5410) in the presence of 1.4 mM of NaCl and at pH 3, $\theta = 90^\circ$ and $T = 40^\circ\text{C}$.

are significantly higher than those obtained for the pure PAAMePy55 polymer chain ($R_{H,\text{app}} \sim 18\text{ nm}$ at low pH). This expansion of the polymer chain, which is in agreement with the fluorescence results, where a decrease of the I_E/I_M ratio was observed³⁹ (see Figure 8 below), clearly indicates that P123 micelles associate with PAAMePy55.

In Figure 6, two representative intensity correlation functions are displayed, one for a mixed solution and one for a pure P123 solution in the presence of 1.4 mM of NaCl (both at pH 3). It is clearly observed that the correlation function of the mixed system is shifted toward longer times and therefore contains an additional slow relaxation process, which shows the complex formation between the two species in the solution.

At pH 9, the relaxation time distributions are very different from those obtained at pH 3 and 5 (Figure 4). Two modes are still present; the faster is due to the diffusion of free P123 micelles in the mixed solution, as was observed at acidic pH values. However, the slow mode is not due to the diffusion of individual PAAMePy55–P123 complexes, since it is much slower than that observed at acidic pH values. This mode belongs to the “fingerprint” relaxation pattern of the pure PAAMePy55 polymer system mentioned above. This becomes evident when comparing this distribution to that of the pure PAAMePy55 system at pH 9 (see Figure 3B). The slow mode, which is present in the mixed system, may thus be related to the (slow) mode caused by the multichain domains due to electrostatic interactions that are present in the pure PAAMePy55 system. The fluorescence results (I_1/I_3 and decay times)³⁹ obtained at pH 9 are clearly indicative of interaction between the PAAMePy55 and P123 block copolymer, although weaker than at acidic pH values. The DLS data are more difficult to analyze and which will be further discussed below.

Complex Formation and Complex Size. In the high molar ratio regime and at acidic pH values, a clear association between the PAAMePy55 polymer and the P123 micelles is observed when examining the relaxation time distributions (Figure 4) and the intensity correlation functions (Figure 6). Due to the high P123 concentrations used, the presence of free micelles is also detected in addition to the slower mode associated with the PAAMePy55–P123 complex as previously mentioned. Four sets of experiments were carried out using four fixed PAAMePy55 polymer concentrations and varying the P123 concentration, at the three pH values studied. For each fixed PAAMePy55 concentration, the fast and slow diffusion coefficients (D_{fast} and D_{slow}) associated are plotted as a function of the P123 concentration and the resultant graph is presented in Figure 7.

We observe in Figure 7 that at all pH values (pH 5 data not shown) the two diffusion coefficients both display a linear

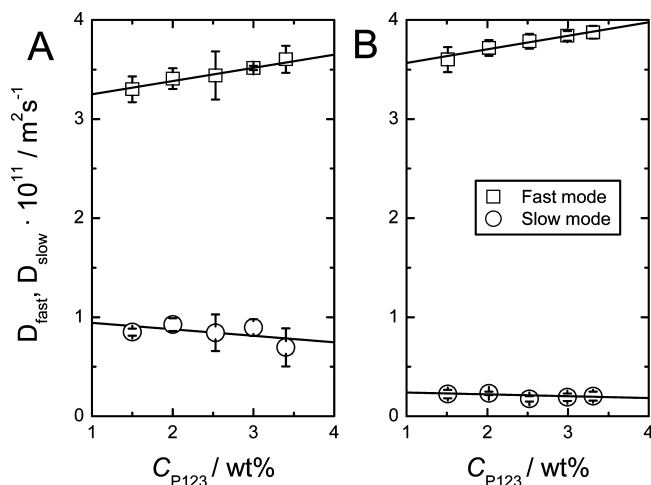


Figure 7. Diffusion coefficients of the PAAMePy55–P123 system as a function of P123 concentration, at pH 3 (and 1.4 mM NaCl) (A) and at pH 9 (and 10 mM NaCl) (B). The data points represent the averages of D values obtained at different PAAMePy55 concentrations: 0.005, 0.01, 0.015, and 0.02 wt % and the error bars represent \pm one standard deviation; $T = 40^\circ\text{C}$.

dependence on the P123 concentration and that they appear to be independent of the PAAMePy55 concentration in the range of dilute concentrations studied. The dependence of the diffusion coefficients on the P123 concentration can be expressed by the following equation:

$$D_{\text{fast,slow}} = D_{\text{P123}=0}^{\text{fast,slow}} (1 + k_{\text{slope}} C_{\text{P123}} + \dots) \quad (4)$$

where $D_{\text{P123}=0}^{\text{fast}}$ and $D_{\text{P123}=0}^{\text{slow}}$ are the apparent fast and slow diffusion coefficients at infinite dilution of P123 but at a finite concentration of PAAMePy55, and C_{P123} is the P123 block copolymer concentration. The term k_{slope} has a similarity to the dynamic virial coefficient,⁴⁹ but in this study it is merely interpreted as the slope of the linear function.

Since the diffusion coefficients are independent of the PAAMePy55 concentration, the values were therefore averaged and only one linear fit of D_{fast} and D_{slow} , respectively, to C_{P123} was performed (Figure 7). The extrapolation of the fast diffusion coefficient to $C_{\text{P123}} = 0$ thus gives the diffusion coefficient $D_{\text{P123}=0}^{\text{fast}}$ and, consequently, $R_{H,\text{P123}=0}^{\text{fast}}$ (from eq 3). At pH 3 and 5, $R_{H,\text{P123}=0}^{\text{fast}} = 11.4 \pm 0.3$, and at pH 9, it is $10.3 \pm 0.1\text{ nm}$, thus displaying an almost constant size with pH. These values are very similar to the different radii values of the pure P123 micelle in water obtained at 40°C in previous studies by using DLS (9.8 nm ³¹ and 10.1 nm ⁴²) and SAXS (10 nm ²⁵). We may thus conclude that free P123 micelles (i.e., micelles that do not associate with the PAAMePy55 polymer chains) must be present in mixed solution. This observation is not surprising since the P123 concentrations are well above the cmc of P123 at 40°C ($1.8 \times 10^{-6}\text{ M}$), and it is also above the concentration at which the PAAMePy55 and the P123 micelles start to associate (3.5×10^{-6} and $6.7 \times 10^{-6}\text{ M}$, at pH 3.6 and 5, respectively), determined from the fluorescence measurements (see Figure 8 below and ref 39).

Similarly, the slow diffusion coefficients at $C_{\text{P123}} = 0$, $D_{\text{P123}=0}^{\text{slow}}$, and the hydrodynamic radii $R_{H,\text{P123}=0}^{\text{slow}}$ associated with them, were estimated at all three pH values. Also in this case, $R_{H,\text{P123}=0}^{\text{slow}}$ can be considered to be constant at acidic pH values within the experimental error ($R_{H,\text{P123}=0}^{\text{slow}} = 35 \pm 5\text{ nm}$ at pH 3 and $36 \pm 6\text{ nm}$ at pH 5), with an average value of $\langle R_{H,\text{P123}=0}^{\text{slow}} \rangle = 36\text{ nm}$. At

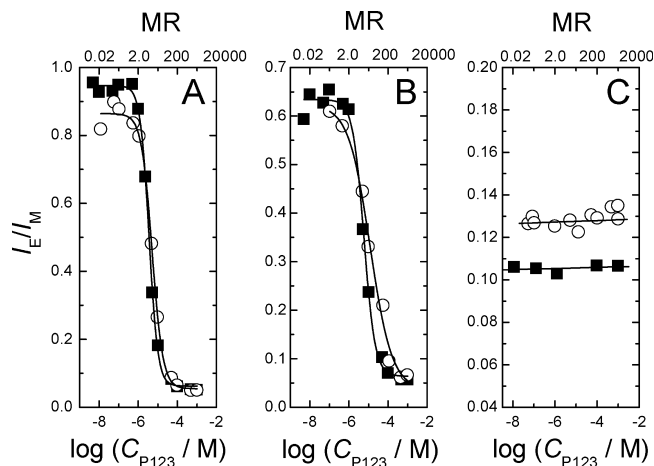


Figure 8. Dependence of the excimer-to-monomer ratio (I_E/I_M) on the P123 concentration with (○) and without (■) NaCl, at pH 3.6 (A), pH 5 (B), and (C) pH 9. At pH 3.6 and 5, the NaCl concentration used was 1 mM of NaCl, and at pH 9, it was 10 mM. The measurements were performed at 40 °C and with an excitation wavelength of 335 nm.

pH 9, the obtained $R_{H,P123=0}^{\text{slow}}$ value is 140 ± 20 nm, which is significantly larger than the value obtained at acidic pH. This is close to 135 nm, which obtained from the slow mode of the PAAMePy55 system at 10 mM without any P123. Furthermore, at all pH values, the presented errors were determined from the linear fits shown in Figure 7 assuming that the relative error of R_H to be the same as that of D_0 in each case.

Fluorescence Spectroscopy. Addition of NaCl. The association between the PAAMePy55 polyelectrolyte and the P123 block copolymer studied by fluorescence techniques was the subject of the previous paper.³⁹ The fluorescence results clearly showed the existence of association between the two polymers with a concomitant expansion of the polyelectrolyte chain, at acidic pH values. This observation is confirmed as shown above by the observed increase in the apparent hydrodynamic radius of the PAAMePy55–P123 complex when compared with pure PAAMePy55. In the DLS experiments, the presence of NaCl was required to screen electrostatic effects, and it is consequently of utmost importance to investigate the influence of NaCl on the nature of the intra- and intermolecular interactions involved.

The emission spectra of the PAAMePy55–P123 system display the characteristic pyrene emission with a shorter wavelength vibronically resolved band (monomer) coexisting with a broad emission band (excimer) at longer wavelengths.^{39,78} The excimer-to-monomer ratio (I_E/I_M) plotted as a function of the P123 concentration (readapted from ref 39) is depicted in Figure 8. At pH 3.6, the presence of 1.4 mM of NaCl has no significant effect on the I_E/I_M curves, meaning that the salt addition does not promote changes in the nature of the (hydrophobic intramolecular) interactions occurring in the system. At pH 5, a slightly different behavior is observed when salt is added, i.e., the range of P123 concentration at which the decrease of the I_E/I_M ratio occurs becomes broader. This observation means that the expansion of the PAAMePy55 polymer chain due to the presence of P123 block copolymer occurs in a more gradual manner; however, it has no effect in the obtained inflection point of the curve. The differences resulting from NaCl addition observed at pH 5 are due to the coexistence of protonated and deprotonated carboxylic groups within the PAAMePy55 polymer chain and will be discussed below.

When the pH is increased (pH 9), the I_E/I_M ratio remains constant all over the P123 concentration range studied. Never-

theless, an increase in I_E/I_M values is observed in the presence of 10 mM of NaCl. At pH 9, the PAA chain is almost totally deprotonated, and in the state of an expanded conformation due to electrostatic repulsions between the COO^- groups. The presence of NaCl reduces the electrostatic repulsion and thus a more coiled polymer conformation is expected, reflected by an increase of the I_E/I_M ratio (i.e., an increase of the intramolecular pyrene-pyrene hydrophobic interactions).

Discussion

In the excellent review by Mandel,⁵² several important concentration intervals are discussed and necessary to take into account in order to understand the behavior of highly charged macromolecules in solution and the results from the light scattering investigations. At very low concentrations (dilute regime), when the concentration of the polyelectrolyte in solution (C) is much lower than the critical overlap concentration (denoted C^{**} in this study), i.e. $C \ll C^{**}$, the highly charged polyelectrolyte chains (or polyions) are stretched due to the osmotic swelling of the counterions. Due to the low concentration, the stiff polyions are unperturbed in solution and move freely. At a given concentration C^{*} , the rotational motion is hindered and a three-dimensional lattice is formed by the *rodlike* polyions. The concentration regime $C < C^{*}$ is seldom investigated by light scattering techniques due to the low scattering intensity of the solutions. Above C^{*} , the further addition of polyelectrolyte causes the lattice to melt (at a critical concentration C^*) due to charge screening in a similar way as addition of simple salt. This regime (i.e., $C^* < C < C^{**}$) is sometimes denoted as the transition regime of *semiflexible* polyions anticipating the semidilute concentration regime of *flexible* polyions, where the system is found when the concentration increases above C^{**} and where a transient network is formed.

The DLS measurements performed in the lower concentration part of the transition regime show single exponential correlation functions with one fast relaxation mode. The fast mode has been interpreted as a result of a coupled diffusion of polyions and counterions, see refs 53–56 and 68 and the references therein. Sedláč et al.⁵³ described this coupling as the motion of polyions and counterions in a common electrostatic field where the polyions experience an electric force due to the concentration fluctuations of the surrounding counterions. The fast mode diffusion coefficient (D_{fast}) has shown to be independent of the molecular weight and concentration of the polyelectrolyte,⁵³ but very dependent on the low molecular weight salt concentration⁶⁹ and of the degree of ionization of the polyelectrolyte chain.^{65,66,69} Therefore, it can be derived that D_{fast} is mainly determined by the fast diffusion rate of the counterions and that the hydrodynamics of the polyelectrolyte chain plays a minor role.

At higher concentrations in the transition regime (still below C^{**}), a slow mode may appear in the relaxation time distributions. The origin of this mode is still under debate and a number of proposals can be found in the literature. Among these, one seems more consensual and relates the slow mode with the collective diffusion of multichain domains ordered in solution as a result of strong intermolecular interactions, i.e. a static structure factor effect. Factors such as the polyelectrolyte molecular weight^{53,54,69} and concentration,^{54,69} ionic strength,^{62,65,69} solvent,^{57,64} temperature,^{57,66} stabilization time,^{61,63} and filtration procedures, made prior to the measurements,^{60,70} were shown to highly influence the formation of these domains.

Semidilute polyelectrolyte systems ($C > C^{**}$) behave similarly to that of uncharged polymer systems and a transient network is formed of the flexible overlapping polyions. The fast

mode, which is always diffusive, is now instead interpreted as the “gel mode” reflecting the network dynamics and a concerted motion of polymer chains, which in the framework of the blob model⁷¹ can be characterized by a cooperative diffusion coefficient.^{52,69} The slow relaxation mode becomes more pronounced in this more concentrated regime^{65,72} which, depending whether it is q dependent or not, may be interpreted in various ways, see e.g. ref 73 and the references therein. For some semidilute uncharged polymers in theta solvents⁴⁹ but also in good solvents,⁷⁴ the slow mode has been found to be connected to the disengagement relaxation of individual chains (i.e., viscoelastic relaxation) and it was then q independent. On the other hand, a q -dependent (q^2 or with a q dependence stronger than q^2) slow mode has been proposed to be associated with long-range concentration fluctuations of large domains (i.e., cluster relaxation) and can be predicted according to the coupling model of Ngai.⁷⁵ Such a q dependent behavior has been observed both for semidilute polyelectrolyte systems, e.g. refs 65, 69, 70, 72, and 76 as well as for semidilute systems of uncharged polymers, associating polymers and for gelling polymer systems.^{73,77}

In our study of the pure labeled and unlabeled PAA systems at pH 9, we were able to identify the slow and fast diffusion characteristics of polyelectrolyte systems (Figure 3). Hence, we believe that the polyelectrolyte systems of our study are in the higher concentration part of the transition regime (still below C^*). The formation of multichain domains is much more favorable at pH 9 and could not be avoided completely by small amounts of NaCl or the presence of P123 in the PAAMePy55 solutions at this pH (see Figures 3 and 4).

Effect of Salt. DLS measurements of the PAAMePy55 polymer were performed at three different pH values (3, 5, and 9), and strange effects can be observed in the relaxation time distributions of such systems as discussed above. Indeed, these effects are strictly related to electrostatic interactions (i.e., varying with the pH) and can be partially avoided by the presence of electrolytes, such as NaCl. Previous studies on the PAAMePy55 polymer⁷⁸ and on similar PAAMePy polymers ($M_n = 2000$ and $450\,000$ g/mol)⁷⁹ showed that their conformation in salt-free solutions depends on the balance between the hydrophobic (pyrene–pyrene) and electrostatic (i.e., degree of ionization) interactions. The degree of ionization (α) for PAA polymers is given by the following equation:^{79,80}

$$\text{pH} = 6.17 - 2 \log\left(\frac{1 - \alpha}{\alpha}\right) \quad (5)$$

The comparison of the titration curves presented in ref 79 shows that between pH 3 and 7 the curves of pyrene-labeled polymers are different from that of the untagged polymer, indicating that the hydrophobic labels influence the degree of deionization. At pH 3, the PAAMePy55 polymer chain is almost completely protonated ($\alpha \sim 0.025$, determined through the eq 5), i.e., the chain is in a collapsed state due to the absence of net charge and pyrene–pyrene interactions are predominant. By increasing the pH to 5, and consequently the degree of ionization of the PAAMePy55 chain ($\alpha \sim 0.2$), the entropy of the counterions increases, causing the polymer chain to expand due to the swelling osmotic pressure of the counterions (i.e., an increase in the volume occupied by the counterions).⁸¹ At pH 9, the PAAMePy55 chain is almost fully ionized ($\alpha \sim 0.96$) and the electrostatic interactions predominate over the hydrophobic interactions.

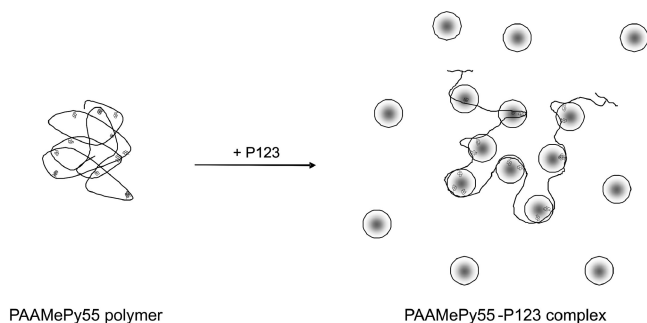
The presence of NaCl has been shown to promote the transition from an expanded to a collapsed state.⁸² However,

according to the DLS measurements at acidic pH presented in this study, the addition of NaCl to the PAAMePy55–P123 system has little effect on the association between the PAAMePy55 polymer chain and the P123 block copolymer micelles, i.e. the measured hydrodynamic radii values were unaltered. The addition of NaCl was also reflected in the results from the steady-state fluorescence measurements on the PAAMePy55–P123 mixed system (Figure 8). The I_E/I_M ratio is affected by the presence of salt in the system at all pH values but the effect becomes progressively more pronounced as the pH (and α) is increased. At pH 3.6, minor effects are observed (only a slight increase in the I_E/I_M ratio in the low P123 concentration regime). At pH 5, the main effect is related with the fact that in the presence of salt, the concentration regime widens in which the association between P123 and PAAMePy55 occurs. A similar behavior was observed by Leyte et al.⁸³ in the investigation of the effect of Na_2CO_3 concentration on the poly(methacrylate) (PMA) conformational changes. They observed that an increase in the amount of salt results in an enlarged pH range where a conformational transition occurs. Later on, Leclercq et al.⁸⁴ reported the effect of salt on the thermodynamic properties of an aqueous solution of PMA and of a hydrophobically modified PMA and it was concluded that the presence of salt results in a variation of the conformational entropy change with a consequent increase in the stability of a coiled polymer conformation. Earlier ^{23}Na NMR studies have revealed that there is a strong association between the Na^+ ion and water.⁸⁵ In our system at pH 5, the presence of the Na^+ ion (by addition of NaCl) most likely results in the formation of complexes between the hydrated Na^+ ions and the COOH and COO^- groups, which leads to a decrease of the intramolecular repulsive electrostatic interactions. As a consequence of this, the intermolecular interaction between the PAAMePy55 chain and the P123 micelles (and the consequent change in the PAA conformation) is now less effective when compared with the situation without NaCl, due to a cooperative conformational transition from a globular to an expanded coil.⁸⁵

Complex Stoichiometry. It is noticed in Figure 7 that the diffusion coefficient of the complex does not change when the molar ratio is increased at acidic pH values, i.e. at a fixed P123 concentration D_{slow} is independent of PAAMePy55 concentration. This shows that the stoichiometry of the complex is independent of the P123 concentration once it is formed and additional P123 addition to the system only leads to more free P123 micelles. Furthermore, the hydrodynamic size of the complex at pH 3 and 5 seems to be the same. When comparing the average value of the hydrodynamic radius ($\langle R_{\text{H,P123=0}}^{\text{slow}} \rangle = 36$ nm) of the PAAMePy55–P123 complex at acidic pH values with the apparent radius of 18 nm of the pure PAAMePy55 polymer chain, a significant increase is observed. The observed increase corresponds to a polymer-coil volume¹ of ca. 8 times larger than in the absence of P123 micelles. Therefore, and considering the volume occupied by a P123 micelle (~ 4850 nm³), this increase implies that the stoichiometry of the complex is, on average, 1:42 PAAMePy55 chain:P123 spherical micelles (see Scheme 1). The pyrene groups of the PAAMePy55 polymer chain are in contact with the PEO corona of the P123 micelle according to the fluorescence data obtained previously.³⁹

The $R_{\text{H,P123=0}}^{\text{slow}}$ value obtained at pH 9 ($R_{\text{H,P123=0}}^{\text{slow}} = 140 \pm 20$ nm) from the slow relaxation mode (see Figure 4C) is too large to be attributed to the translational motion of a single PAAMePy55–P123 complex. Instead it is likely to be caused by the formation of multichain domains, i.e., the intermolecular repulsion exerted by the carboxylate ions (COO^-) force the

SCHEME 1: Schematic Representation of the PAAMePy55:P123 System in the High P123 Concentration Regime and at Acidic pH Values



stretched PAAMePy55 chains to diffuse as a group in a common electric field, appearing as one large object. The I_1/I_3 ratio and the fluorescence decay times³⁹ indicate that PAAMePy55 and P123 still associate at this high pH. There are two alternative explanations that can be put forward. First, we may interpret the multichain domains to be formed by PAAMePy55 chains that entrap some of the P123 micelles. The entrapped micelles are then close enough to associate with (or bind to) the polyelectrolyte chains, while the majority diffuse freely in the solution and do not interact with the PAAMePy55 chains. The randomly labeling of the PAAMePy55 chain could lead to an uneven distribution of the pyrene groups along the PAA chain, creating high and low labeled regions. These highly labeled regions constitute preferential sites of association for the P123 micelles at high pH 9. However, the association between P123 and PAAMePy55 at pH 9 is considered to be minor. This is confirmed by the very low relative amplitude of the slow mode compared to the fast mode in the relaxation time distributions (see Figure 4C). The amplitude of the slow mode is 0.05. This is a clear indication that the scattering of these large domains contributes very little to the total scattered intensity (only 5%) due to the fact that the relaxation time distributions are intensity distributions. Moreover, the total scattering intensity of a given sample is always higher at pH 3 than at pH 9. This means that with the PAAMePy55–P123 micellar complexes at pH 9 the domains are present at a very low number in the solution and thus give a little contribution to the total scattering intensity. It is thus difficult to conclude from the DLS results alone that the multichain domains consist of polyelectrolyte chains with entrapped P123 micelles. A comparison of the size of the polyelectrolyte domains in the absence and presence of P123 was not possible since their size is dependent on the polyelectrolyte concentration,^{53,69} and a DLS study of the pure PAAMePy55 system at such a low concentrations was not possible to perform due to the low scattering intensity. However, we can conclude that the multichain domains of the pure polyelectrolyte system are not destroyed by the presence of P123 block copolymer micelles.

The second explanation refers to the fact that the PEO–PPO–PEO copolymers are polydisperse. We can not exclude that highly hydrophilic diblock PEO–PPO copolymers (with high PEO content) that may not be involved in the micelle formation at 40 °C can be present in the micellar P123 solution as single copolymer chains, which has been established in recent studies by Hvidt et al.^{30,35} In the mixed case, these diblock impurities can associate with the PAAMePy55 chains. This association will be reflected in the fluorescence data but not in the DLS data. We may then suggest that the multichain domains of the mixed system at pH 9 are the same as those of the pure

PAAMePy55 polymer system. The presence of diblocks does not affect the domains and hence the DLS data stay unchanged. At the present, we are not able to distinguish between these two scenarios unless the P123 sample is purified, which would be the next obvious step of this investigation.

Conclusions

The intermolecular association between a PAA polymer hydrophobically modified with pyrene (PAAMePy55) and the PEO–PPO–PEO triblock copolymer P123 was studied by dynamic light scattering and by steady-state fluorescence spectroscopy (the comprehensive fluorescence study is reported in ref 39). The previous fluorescence study showed that PAAMePy55 and P123 associate strongly at low pH, leading to the formation of PAAMePy55–P123 complexes, where the pyrene groups are located at the PPO/PEO interface of the P123 micelle.³⁹ Another important finding, resulting from the fluorescence investigations, is that no lowering of the cmc of P123 is observed upon addition of the PAAMePy55, thus showing that this polymer cannot promote the micellization of P123, i.e., the association occurs at P123 concentrations higher than the cmc of P123. This shows that the P123 micelles are first formed in solution and the association between the P123 micelles and the PAAMePy55 polymer chains is only effective after achieving a given P123/PAAMePy55 molar ratio. The degree of ionization and consequently the hydrophobicity (and flexibility) of the PAAMePy55 chain were also found to be of major importance for the association: the former is the driving force of the association and the latter determines the extension of the association. The DLS measurements performed in the low molar ratio regime and at acidic pH values showed that there is no change in the apparent hydrodynamic radius of the PAAMePy55 chain in the mixed system compared to that obtained for the pure PAAMePy55 chain. In the high molar ratio regime and at acidic pH, bimodal relaxation time distributions were obtained, which corresponded to the diffusion of free P123 micelles and of PAAMePy55–P123 complexes, respectively. From the DLS studies at acidic pH, the hydrodynamic size of PAAMePy55–P123 complex formed was determined, and which implied that the complex consists of one single PAAMePy55 chain with 42 P123 micelles associated to it. At pH 9, the relaxation time distributions revealed the presence of a short number of large domains formed by several PAAMePy55 chains, which are characteristic of polyelectrolyte systems in addition to free P123 micelles that are in the majority in number. The presence of P123 did not lead to the total disruption of these domains. Unless the P123 sample is purified, no final conclusions can be drawn of the structure of these domains since they may either originate from several PAAMePy55 chains with a few entrapped P123 micelles or several polyelectrolyte chains that associate with unassociated diblock impurities that are not involved in the micellization process of P123. However, we may conclude that the association between P123 and PAAMePy55 is minor at high pH as compared to acidic pH values.

In this study of a polyelectrolyte-block copolymer system, we have shown that a good agreement (and complementary information) can be obtained from the DLS and fluorescence approaches. The concentration at which the two polymers begin to have a significant degree of association (even at high pH) could be easily detected in the fluorescence studies, and the data also indicated that the preferential location of the pyrene groups of the PAAMePy55 polymer is found enclosed in the P123 micelles.

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References and Notes

- (1) Evans, D. F.; Wennerström, H., *The Colloidal Domain: Where Physics, Chemistry, Biology, and Technology Meet*; Wiley-VCH: New York, 1999.
- (2) Jönsson, B.; Lindman, B.; Holmberg, K.; Kronberg, B. *Surfactants and Polymers in Aqueous Solution*, 2nd ed.; John Wiley & Sons: Chichester, 1998.
- (3) Schillén, K.; Anghel, D. F.; Miguel, M. D.; Lindman, B. *Langmuir* **2000**, *16*, 10528.
- (4) Fundin, J.; Hansson, P.; Brown, W.; Lidegran, I. *Macromolecules* **1997**, *30*, 1118.
- (5) Robb, I. D.; Stevenson, P. *Langmuir* **2000**, *16*, 7168.
- (6) Cole, M. L.; Whateley, T. L. *J. Colloid Interface Sci.* **1996**, *180*, 421.
- (7) Anghel, D. F.; Saito, S.; Baran, A. *Langmuir* **1998**, *14*, 5342.
- (8) D'Errico, G.; Ciccarelli, D.; Ortona, O.; Paduano, L.; Sartorio, R. *J. Colloid Interface Sci.* **2004**, *270*, 490.
- (9) Winnik, F. M.; Ringsdorf, H.; Venzmer, J. *Langmuir* **1991**, *7*, 912.
- (10) Khutoryanskiy, V. V.; Mun, G. A.; Nurkeeva, Z. S.; Dubolazov, A. V. *Polym. Int.* **2004**, *53*, 1382.
- (11) Chu, B. *Langmuir* **1995**, *11*, 414.
- (12) Chu, B.; Zhou, Z. In *Nonionic surfactants: polyoxyalkylene block copolymers*; Nace, V. M., Dekker, M., Eds.; Marcel-Dekker, Inc.: New York, 1996; p 67.
- (13) Alexandridis, P.; Hatton, T. A. *Colloids Surf. A* **1995**, *96*, 1.
- (14) Almgren, M.; Brown, W.; Hvidt, S. *Colloid Polym. Sci.* **1995**, *273*, 2.
- (15) Alexandridis, P.; Lindman, B. *Amphiphilic Block Copolymers: Self-Assembly and Applications*; Elsevier: Amsterdam, 1997.
- (16) Nagarajan, R.; Barry, M.; Ruckenstein, E. *Langmuir* **1986**, *2*, 210.
- (17) Brown, W.; Schillén, K.; Almgren, M.; Hvidt, S.; Bahadur, P. *J. Phys. Chem.* **1991**, *95*, 1850.
- (18) Brown, W.; Schillén, K.; Hvidt, S. *J. Phys. Chem.* **1992**, *96*, 6038.
- (19) Mortensen, K.; Brown, W. *Macromolecules* **1993**, *26*, 4128.
- (20) Glatter, O.; Scherf, G.; Schillén, K.; Brown, W. *Macromolecules* **1994**, *27*, 6046.
- (21) Pedersen, J.; Gerstenberg, M. *Macromolecules* **1996**, *29*, 1363.
- (22) Pedersen, J.; Gerstenberg, M. *Colloids Surf. A* **2003**, *213*, 175.
- (23) Goldmints, I.; von Gottberg, F. K.; Smith, K. A.; Hatton, T. A. *Langmuir* **1997**, *13*, 3659.
- (24) Goldmints, I.; Yu, G.; Booth, C.; Smith, K. A.; Hatton, T. A. *Langmuir* **1999**, *15*, 1651.
- (25) Jansson, J.; Schillén, K.; Nilsson, M.; Söderman, O.; Fritz, G.; Bergmann, A.; Glatter, O. *J. Phys. Chem. B* **2005**, *109*, 7073.
- (26) Mortensen, K. *Polymers Adv. Technol.* **2001**, *12*, 2.
- (27) Wanka, G.; Hoffmann, H.; Ulbricht, W. *Macromolecules* **1994**, *27*, 4145.
- (28) Beezer, A.; Loh, W.; Mitchell, J.; Royall, P.; Smith, D.; Tute, M.; Armstrong, J.; Chowdhry, B.; Leharne, S.; Eagland, D.; Crowder, N. *Langmuir* **1994**, *10*, 4001.
- (29) da Silva, R. C.; Olofsson, G.; Schillén, K.; Loh, W. *J. Phys. Chem. B* **2002**, *106*, 1239.
- (30) Batsberg, W.; Ndoni, S.; Trandum, C.; Hvidt, S. *Macromolecules* **2004**, *37*, 2965.
- (31) Jansson, J.; Schillén, K.; Olofsson, G.; da Silva, R. C.; Loh, W. *J. Phys. Chem. B* **2004**, *108*, 82.
- (32) Nilsson, M.; Håkansson, B.; Söderman, O.; Topgaard, D. *Macromolecules* **2007**, *40*, 8250.
- (33) Linse, P. *Macromolecules* **1994**, *27*, 2685.
- (34) Linse, P. *Macromolecules* **1994**, *27*, 6404.
- (35) Hvidt, S.; Batsberg, W. *Int. J. Polym. Anal. Chem.* **2007**, *12*, 13.
- (36) Mortensen, K.; Batsberg, W.; Hvidt, S. *Macromolecules* **2008**, *41*, 1720.
- (37) Khutoryanskiy, V. V.; Dubolazov, A. V.; Nurkeeva, Z. S.; Mun, G. A. *Langmuir* **2004**, *20*, 3785.
- (38) Baranovsky, V.; Petrova, T.; Rashkov, I. *Eur. Polym. J.* **1991**, *27*, 1045.
- (39) Costa, T.; Schillén, K.; Miguel, M. G.; Lindman, B.; Seixas de Melo, J. *J. Phys. Chem. B* **2009**, *113*, 6194.
- (40) Löf, D.; Niemiec, A.; Schillén, K.; Loh, W.; Olofsson, G. *J. Phys. Chem. B* **2007**, *111*, 5911.
- (41) Löf, D.; Schillén, K.; Torres, M. F.; Müller, A. *J. Langmuir* **2007**, *23*, 11000.
- (42) Schillén, K.; Jansson, J.; Löf, D.; Costa, T. *J. Phys. Chem. B* **2008**, *112*, 5551.
- (43) Löf, D.; Tomšič, M.; Glatter, O.; Fritz-Popovski, G.; Schillén, K. *J. Phys. Chem. B*, ASAP. DOI: 10.1021/jp808442d.
- (44) Anghel, D. F.; Alderson, V.; Winnik, F. M.; Mizusaki, M.; Morishima, Y. *Polymer* **1998**, *39*, 3035.
- (45) Jakš, J. *Czech. J. Phys.* **1988**, *38*, 1305.
- (46) Berne, B. J.; Pecora, R. *Dynamic Light Scattering: with Applications to Chemistry, Biology and Physics*, 2nd ed.; Dover Publications, Inc.: New York, 2000.
- (47) Štěpánek, P. In *Dynamic Light Scattering: The Method and Some Applications*, Brown, W., Ed.; Oxford University Press: Oxford, 1993; p 177.
- (48) Johnsen, R. M.; Brown, W. In *Laser Light Scattering in Biochemistry*, Harding, S. E., Sattelle, D. B., Bloomfield, V. A., Eds.; The Royal Society of Chemistry, Thomas Graham House, Science Park: Cambridge, 1992.
- (49) Brown, W.; Nicolai, T. In *Dynamic Light Scattering: The Method and Some Applications*; Brown, W., Ed.; Oxford University Press: Oxford, 1993.
- (50) Ganguly, R.; Aswal, V. K.; Hassan, P. A. *J. Colloid Interface Sci.* **2007**, *315*, 683.
- (51) Denkova, A. G.; Mendes, E.; Coppens, M.-O. *J. Phys. Chem. B* **2008**, *112*, 793.
- (52) Mandel, M. In *Dynamic Light Scattering: The Method and Some applications*; Brown, W., Ed.; Clarendon Press: Oxford, 1993.
- (53) Sedláč, M.; Amis, E. J. *J. Chem. Phys.* **1992**, *96*, 817.
- (54) Sedláč, M.; Amis, E. J. *J. Chem. Phys.* **1992**, *96*, 826.
- (55) Sedláč, M. *J. Chem. Phys.* **1997**, *107*, 10799.
- (56) Chang, R.; Yethiraj, A. *J. Chem. Phys.* **2002**, *116*, 5284.
- (57) Sedláč, M. *J. Chem. Phys.* **1994**, *101*, 10140.
- (58) Sedláč, M. *J. Chem. Phys.* **1997**, *107*, 10805.
- (59) Sedláč, M. *Langmuir* **1999**, *15*, 4045.
- (60) Sedláč, M. *J. Chem. Phys.* **2002**, *116*, 5236.
- (61) Sedláč, M. *J. Chem. Phys.* **2002**, *116*, 5246.
- (62) Sedláč, M. *J. Chem. Phys.* **2002**, *116*, 5256.
- (63) Sedláč, M. *J. Chem. Phys.* **2005**, *122*, 151102.
- (64) Sedláč, M.; Koňák, C.; Labsky, J. *Polymer* **1991**, *32*, 1688.
- (65) Sedláč, M.; Koňák, C.; Štěpánek, P.; Jakš, J. *Polymer* **1987**, *28*, 873.
- (66) Sedláč, M.; Koňák, C.; Štěpánek, P.; Jakš, J. *Polymer* **1990**, *31*, 253–257.
- (67) Adamczyk, Z.; Bratek, A.; Jachimska, B.; Jasinski, T.; Warszynski, P. *J. Phys. Chem. B* **2006**, *110*, 22426.
- (68) Tanahatue, J. J.; Kuil, M. E. *J. Phys. Chem. A* **1997**, *101*, 8389.
- (69) Förster, S.; Schmidt, M.; Antonietti, M. *Polymer* **1990**, *31*, 781.
- (70) Tanahatue, J. J.; Kuil, M. E. *J. Phys. Chem. B* **1997**, *101*, 10839.
- (71) de Gennes, P. G. *Scaling Concepts in Polymer Physics*; Cornell University Press: Ithaca, NY, 1979.
- (72) Buhler, E.; Rinaudo, M. *Macromolecules* **2000**, *33*, 2098.
- (73) Nyström, B.; Lindman, B. *Macromolecules* **1995**, *28*, 967.
- (74) Wang, C. H.; Zhang, X. Q. *Macromolecules* **1993**, *26*, 707.
- (75) Ngai, K. *Adv. Colloid Interface Sci.* **1996**, *64*, 1.
- (76) Tanahatue, J. J.; Kuil, M. E. *J. Phys. Chem. B* **1997**, *101*, 9233.
- (77) Kjøniksen, A.-L.; Nyström, B. *Macromolecules* **1996**, *29*, 7116.
- (78) Seixas de Melo, J.; Costa, T.; Miguel, M. D.; Lindman, B.; Schillén, K. *J. Phys. Chem. B* **2003**, *107*, 12605.
- (79) Seixas de Melo, J.; Francisco, A.; Costa, T.; Maçanita, A.; Gago, S.; Gonçalves, I. S. *PCCP* **2007**, *9*, 1370.
- (80) Katchalsky, A.; Spitnik, P. *J. Polym. Sci.* **1947**, *2*, 432.
- (81) Volkov, E. V.; Filippova, O. E.; Khokhlov, A. R. *Colloid J.* **2004**, *66*, 663.
- (82) Volkov, E. V.; Filippova, O. E.; Khokhlov, A. R. *Colloid J.* **2004**, *66*, 669.
- (83) Leyte, J. C.; Mandel, M. *J. Polym. Sci., Part A: Gen. Pap.* **1964**, *2*, 1879.
- (84) Leclercq, L.; Pollet, A.; Morcellet, M.; Martel, B. *Eur. Polym. J.* **1999**, *35*, 185.
- (85) Gustavsson, H.; Lindman, B.; Tornell, B. *Chem. Scripta* **1976**, *10*, 136.