confinement limit ($r_G < r_P$), a quantitative comparison with the results presented here was not attempted.

Registry No. Polyisoprene, 9003-31-0.

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Hydrophobic Attraction of Pyrene-End-Labeled Poly(ethylene glycol) in Water and Water–Methanol Mixtures

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ABSTRACT: Fluorescence stationary-state measurements of pyrene-end-labeled poly(ethylene glycols) (Py-PEG-Py) of three different molecular weights are presented. In aqueous solution the excimer to monomer intensity ratio, I_e/I_m, in the intramolecular excimer region decreases as the polymer molecular weight is increased. For Py-PEG-Py of PEG weight-average molecular weight of 4800, the I_e/I_m does not scale according to the Wilemski–Fixman theory based on a diffusion-controlled end-to-end cyclization process. Surprisingly, this Py-PEG-Py(4800) phase separates. We believe that these observations are due to a hydrophobic attraction between pyrene groups. The addition of methanol reduces the hydrophobic attraction by the preferential solvation of methanol molecules around the pyrene group.

Introduction

Fluorescence techniques have long been employed to investigate the dynamics of chemical species at the molecular level. There is a large amount of literature dealing with the studies of molecular dynamics of either synthetic or natural molecules with the aid of a variety of fluorescence techniques.[1] Among the various fluorescence methods, excimer formation has been frequently used to study polymer dynamics. In this technique an aromatic ring in an electronically excited state must encounter an identical chromophore in the ground state during the lifetime of the excited state. If the two chromophores are in the proper geometry they can form a sandwich-shaped excimer giving a broad emission at a lower energy than that of the individually excited aromatic ring. Examples of the application of excimer formation using probe molecules similar to those examined in this work are in the detection of the formation of association complexes between two different water-soluble polymers[2-4] and as probes of the end-to-end cyclization of polymer chains.[5-8] Among the many possible fluorescent chromophores, pyrene and its derivatives have been extensively used to study excimer formation due to their long excited-state lifetime and their spectral sensitivity to the medium examined.[9] The spectral sensitivity of pyrene molecules to the local environment has been employed to study micelle formation of surfactants or block copolymers[10-12] and polymer adsorption on surfaces.[13-16]

Traditionally, the introduction of fluorescent groups is considered a benign process to study intrinsic properties without perturbing the polymer chains. However, this may not be true for short polymer chains in a particular solvent. Cheung et al.[7] found anomalous behavior in the study of diffusion-controlled end-to-end cyclization using pyrene-end-labeled poly(ethylene glycols) in various solvents. They were able to correlate the measured excimer to monomer intensity ratio, I_e/I_m, with the inverse solvent viscosity in all solvents except water and methanol where

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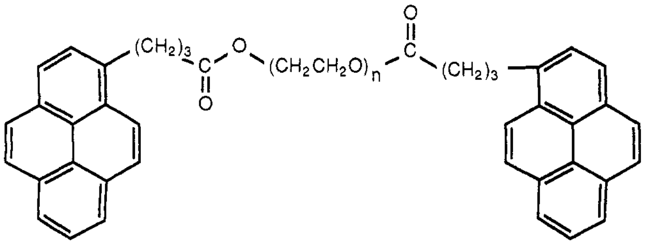
unusually high I_e/I_m were measured.

The purpose of this paper is to extend Cheung et al.'s work with a special emphasis on water and methanol as solvents to explain the deviation from diffusion-controlled end-to-end cyclization. The I_e/I_m were measured over a range of polymer concentrations and in water and water–methanol mixtures with the same polymer as that used by Cheung et al. but for three different molecular weights. We interpret the observed data in terms of a hydrophobic effect.

Experimental Section

1. **Material.** Poly(ethylene glycols) (PEG) having polydispersities less than 1.10 and weight-average molecular weights of 4800, 9200, and 11 000 were purchased from Polysciences Inc. 1-Pyrenebutyric acid (PBA) for pyrene-end-labeled PEG (Py-PEG-Py) synthesis, obtained from Eastman Kodak, was purified by recrystallization in dry toluene. *p*-Toluenesulfonic acid from Aldrich Chemical Co. served as purchased as the catalyst for the Py-PEG-Py synthesis. All solvents were Baker Chemical Co. spectroscopic grade and were used without further purification. Glass-distilled deionized water was the primary solvent for the fluorescence studies.

Py-PEG-Py samples were prepared by the direct esterification between hydroxy groups located at both ends of the PEG backbone and the carboxyl group of PBA following the method developed by Cuniberti and Perico.[17] The detailed procedures for synthesis and purification were described elsewhere.[4,17] The extent of labeling was confirmed by UV spectroscopy; the structure is shown below. From a comparison of UV spectra of the end-labeled polymer with that of methyl 1-pyrenebutyrate from Molecular Probes Inc. we concluded, within an accuracy of 5%, that the polymers were fully end labeled with pyrene groups. We denote the pyrene-tagged samples as Py-PEG-Py(4800), Py-PEG-Py(9200), and Py-PEG-Py(11 200), where the numbers in parentheses represent the weight-average molecular weights of the original untagged PEG samples as reported by Polysciences Inc.



2. **Sample Preparation.** The Py-PEG-Py solutions, of concentrations ranging from 1×10^{-7} mol/L to 1×10^{-3} mol/L, were prepared by the series dilution of stock solutions. Here the concentrations are based on the polymer molecular weight rather than that of the repeating unit. The stock solutions were obtained from dry solid-state Py-PEG-Py samples preserved in a vacuum chamber. Solutions were prepared immediately before the fluorescence measurement was taken to avoid hydrolysis of the labeled polymer.

3. **Fluorescence Measurement.** Photostationary-state spectra were measured on a SPEX Fluorolog 212 spectrofluorometer as reflections from the front face of the cell at an angle 22.5° from the incident light. The excitation wavelength was 343 nm, corresponding to the ^1L_a band of the pyrene ring, and the spectra were corrected for instrumental response from 360 to 600 nm. In addition, the samples were purged with nitrogen for at least 30 min before the measurement. Two spectral parameters were of particular interest: the excimer to monomer intensity ratio, I_e/I_m, calculated from envelope intensity measurements around 475–485 nm for the excimer (I_e) and at 376 nm for the monomer (I_m), and I_1/I_3, the relative intesities of first (376 nm) and third (386 nm) monomer fluorescence bands.

Fluorescence lifetime was measured on a single photon counting PRA (System 2000) instrument. Excitation was performed at 343 nm and excimer emission was monitored at 480 nm. Lifetimes and preexponential factors from multiexponential decay were determined by a nonlinear least-squares deconvolution method.

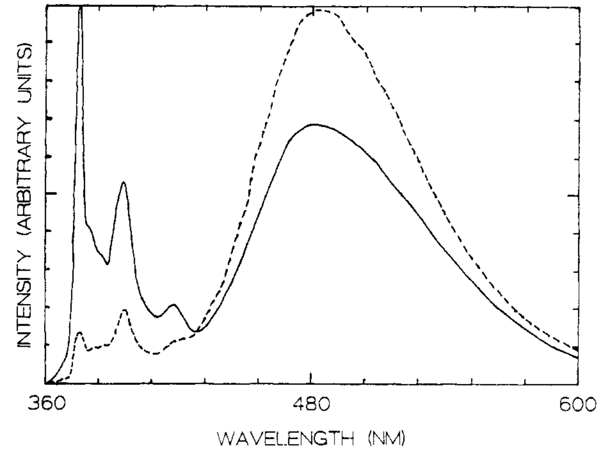


Figure 1. Fluorescence spectra of Py-PEG-Py(4800) in aqueous solution at different polymer concentrations at 298 K: solid line, 1×10^{-5} mol/L; dotted line, 5×10^{-4} mol/L (phase separated).

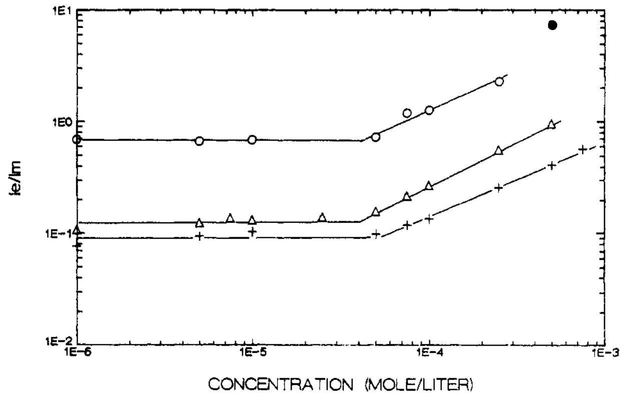


Figure 2. log–log plot of I_e/I_m ratios as a function of polymer concentration in deionized water for Py-PEG-Py samples of different molecular weights at 298 K: (○) Py-PEG-Py(4800); (△) Py-PEG-Py(9200); (+) Py-PEG-Py(11 200); (●) phase separation observed.

Results

1. **Effect of Pyrene-Tagged Polymer Concentration on Fluorescence Spectra.** Typical fluorescence spectra at low concentration, below 1×10^{-5} mol/L, shown in Figure 1 by the solid line, correspond to the spectra reported elsewhere[7] for the same pyrene-end-labeled poly(ethylene glycol). These spectra comprise two parts: monomer emission bands and an excimer emission peak. Monomer emission consists of five characteristic fluorescence bands: three distinct peaks (376, 396, 418 nm) and two appearing as shoulders (381, 386 nm). These wavelengths are identical with those of 1-pyrene butyric acid.[4] Excimer emission is very broad and has a peak intensity around 480 nm. The wavelength of the excimer peak varies slightly with the polymer concentration and molecular weight. The excimer peak intensity increases as the molecular weight of tagged polymer decreases, in agreement with the results of Cuniberti and Perico[17] and Winnik et al.[5,6,8]

A plot of the logarithm of the excimer to monomer intensity ratio (I_e/I_m) against the logarithm of the polymer concentration is shown in Figure 2. We note that I_e/I_m for samples of different molecular weights show similar trends with concentration. We can clearly divide the concentration region examined here into two parts: a plateau region and a power-law region with a positive slope. From the extensive studies of Winnik et al.[5-8] we believe that the plateau region, where the I_e/I_m ratio is almost

Table I
Estimated End-to-End Distance for Py-PEG-Py Samples of Different Molecular Weights in Water at 303 K

mol wt, $\bar{M}_w$, g/mol	polydispersity, $[\bar{M}_w/\bar{M}_n]$	end-to-end dist, $[\bar{r}^2]^{1/2}$, Å
4800	1.05	57.7
9200	<1.10	79.9
11200	<1.10	88.1

Table II
Comparison of the Observed I_e/I_m in the Region of Intramolecular Formation and $(I_e/I_m)_{intra}$ Calculated from the Power-Law Region

	$(I_e/I_m)_{obsd}$	$(I_e/I_m)_{calcd}$	% dev
Py-PEG-Py(4800)	0.681	0.393	73.3
Py-PEG-Py(9200)	0.126	0.075	68.0
Py-PEG-Py(11 200)	0.091	0.067	35.8

constant, is primarily due to intramolecular excimer formation. This type of excimer formation is associated with the cyclization of pyrene groups labeled at both ends of a PEG chain[7] and the I_e/I_m ratios in this region for various molecular weights should be closely related to the end-to-end distance. The estimated end-to-end distances are listed in Table I. A quantitative analysis of I_e/I_m ratios in terms of the Wilemski–Fixman model[18-20] will be discussed in detail later.

The power-law region with a positive slope is due to the association of pyrene groups from different polymer chains as well as the cyclization of pyrene groups of a single chain. In order to separate intermolecular and intramolecular excimer formation using the measured I_e/I_m ratios in this region, the intermolecular contribution was assumed to be directly proportional to the concentration of pyrene molecules (or twice the polymer chain concentration). With this assumption we were able to calculate the I_e/I_m ratios in the region of intramolecular excimer formation giving rise to an exponent for the intermolecular region of unity: $(I_e/I_m)_{inter} = AC_p^B$ where $B = 1$. A logarithmic plot of I_e/I_m ratio for intermolecular excimer formation against polymer concentration is shown in Figure 3. The calculated I_e/I_m values in the intramolecular region for different polymer molecular weights are tabulated in Table II and compared with those observed experimentally. Notice that the difference between calculated and observed I_e/I_m ratios in the intramolecular excimer formation region decreases as the polymer molecular weight is increased.

A surprising result was observed for Py-PEG-Py(4800) at a concentration of 5×10^{-4} mol/L. As soon as the solution was prepared, it became turbid, eventually precipitating, leaving a dense liquid polymer phase at the bottom of the test tube. This phase separation was not observed for the other polymers of higher molecular weight even at concentrations up to 1×10^{-3} mol/L. The fluorescence spectrum of the phase-separated sample is shown as a dashed line in Figure 1. Notice the large excimer formation and the decrease in monomer intensities as well as the change in the ratio of the first and third monomer peaks, I_1/I_3.

The pyrene I_1/I_3 ratio has served in previous work as a probe of the local environment, with I_1/I_3 being lower when the pyrene is in a more organic environment.[9,10] In general, asymmetric substitution will tend to decrease the sensitivity to the environment.[21] However, some sensitivity seems to remain in our 1-substituted pyrene as shown in Figure 4 where the I_1/I_3 ratios for the three samples are plotted against concentration. Py-PEG-Py(4800) shows a drastic drop in the I_1/I_3 ratio at the concentration where the phase separation was observed. This result is similar to the sudden I_1/I_3 ratio drop utilized to determine the critical micelle concentration (cmc) with pyrene probes.[10] In our situation, aggregation among polymer chains via pyrene groups would create a more organic environment. This aggregation is consistent with the high I_e/I_m ratio for the phase-separated sample.

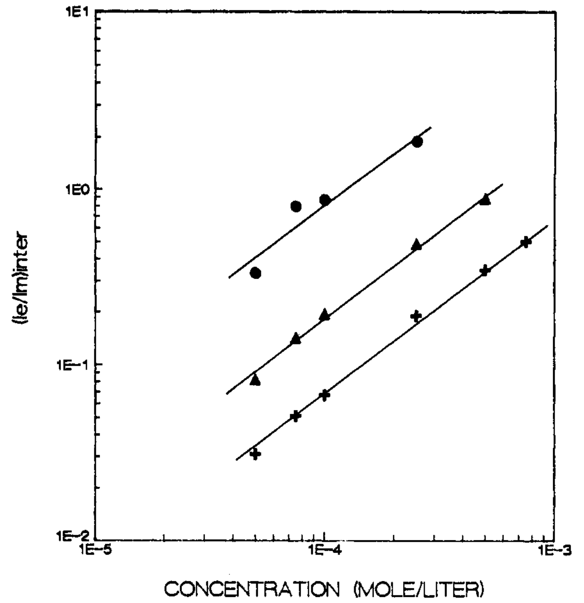


Figure 3. log–log plot of I_e/I_m ratios corrected for the intramolecular excimer formation as a function of polymer concentration for three Py-PEG-Py samples at 298 K: (●) Py-PEG-Py(4800); (▲) Py-PEG-Py(9200); (+) Py-PEG-Py(11 200).

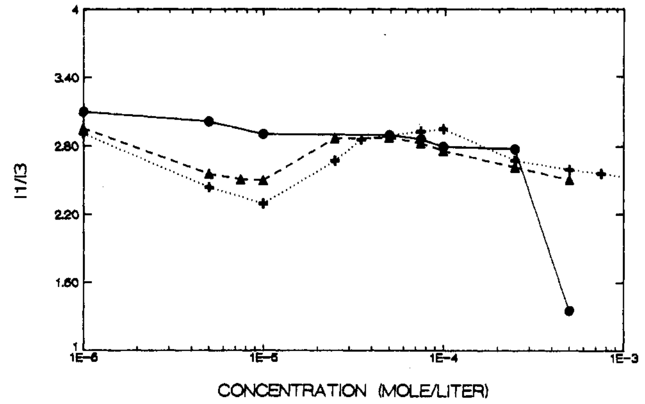


Figure 4. I_1/I_3 ratio as a function of polymer concentration for three Py-PEG-Py samples at 298 K in deionized water: (●) Py-PEG-Py(4800); (▲) Py-PEG-Py(9200); (+) Py-PEG-Py(11 200).

2. Excimer to Monomer Intensity Ratios (I_e/I_m) in the Water–Methanol Cosolvent System. We measured the I_e/I_m ratio as a function of concentration for Py-PEG-Py(4800) dissolved in water–methanol mixtures for several methanol contents. In the concentration range of 1×10^{-7} to 1×10^{-5} mol/L the I_e/I_m ratio is almost invariant with concentration for each water–methanol mixture, indicating that only intramolecular excimer formation is occurring. Similar behavior was observed for Py-PEG-Py(11 200). The I_e/I_m ratios at a fixed polymer concentration for the two molecular weights are shown as a function of methanol content in Figure 5. The I_e/I_m ratio decreases down to 90% methanol content with a slight increase in I_e/I_m for pure methanol. The I_e/I_m ratio for the higher molecular weight polymer shows a similar decrease with methanol content; however, both the initial value and percent decrease are substantially lower. For pyrene end-labeled polymer the formation of intramolecular excimers at room temperature should be diffusion

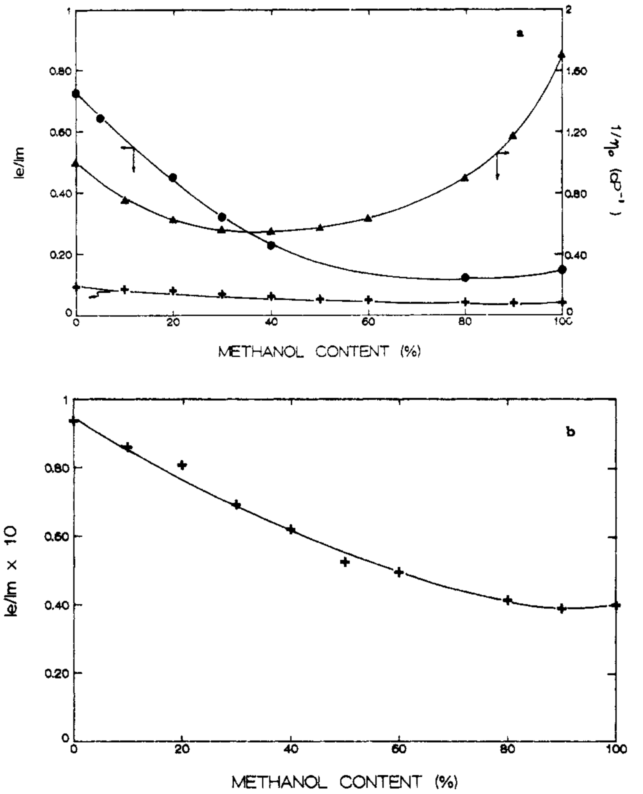


Figure 5. I_e/I_m ratio and the inverse viscosity with the addition of methanol. (a) Change of the I_e/I_m ratio as a function of methanol content at 298 K and change of the inverse viscosity of water–methanol mixture with the addition of methanol at 293 K:[30] (●) Py-PEG-Py(4800), concentration = 1 × 10^{-6} mol/L; (+) Py-PEG-Py(11 200), concentration = 5 × 10^{-6} mol/L; (▲) inverse viscosity of the solution. (b) I_e/I_m ratio as a function of methanol content for the Py-PEG-Py(11 200) sample plotted on an expanded scale.

limited, implying an inverse dependence on solvent viscosity.[7] The effect of methanol content on the solvent viscosity is also included in Figure 5. The dependence of I_e/I_m on the methanol content is not well correlated with the inverse cosolvent viscosity.

In order to illustrate this lack of correlation between I_e/I_m and the solvent viscosity more clearly, the solvent-viscosity-corrected I_e/I_m ratio $[(I_e/I_m)\eta_0]$ is plotted as a function of methanol content in Figure 6. For a diffusion-controlled process, $(I_e/I_m)\eta_0$ should be independent of the change in solvent viscosity.[22] From this figure we see that the behavior of the I_e/I_m ratios for intramolecular excimer formation in water–methanol mixture cannot be explained simply in terms of the diffusion-controlled end-to-end cyclization. We further notice that the decrease in $(I_e/I_m)\eta_0$ above 20% methanol is almost linear for the higher molecular weight polymer sample whereas $(I_e/I_m)\eta_0$ decays much more rapidly for the low molecular weight polymer. These results are interpreted in terms of preferential solvation in the next section.

Discussion

1. Pyrene-End-Labeled Poly(ethylene glycols) in Aqueous Solution. Experimentally measured I_e/I_m ratios in the intramolecular excimer formation region can be compared quantitatively with the cyclization rate constant (k_{cy}) predicted by the Wilemski–Fixman theory[18-20] and with other experimental results.[23,24] Wilemski and Fixman predict that for a nondraining Rouse chain k_{cy} is proportional to $N^{-3/2}$, where N is the number of repeat units in the chain. On the experimental side, Redpath and Winnik[23,24] have utilized fluorescence lifetime studies as well as photostationary-state measurements to show that k_{cy} is proportional to $N^{-1.6}$ in a Θ solvent. Since the I_e/I_m ratio is proportional to k_{cy},[6,7] we are able to compare the scaling of I_e/I_m with that expected for k_{cy} in Table III. Notice that for Py-PEG-Py(9200) and Py-PEG-Py(11 200) I_e/I_m scales according to the prediction while the measured I_e/I_m for Py-PEG-Py(4800) is much higher than the theory predicts. This implies that the end-to-end cyclization for Py-PEG-Py(4800) is not simply a diffusion-limited process as assumed in the Wilemski–Fixman theory. We attribute the unusually high I_e/I_m ratio to hydrophobic attractions between pyrene groups.

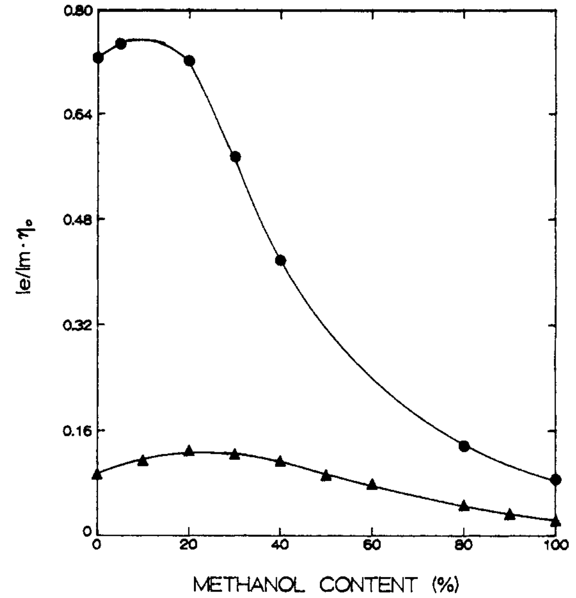


Figure 6. Solvent-viscosity-corrected I_e/I_m ratio as a function of methanol content in water–methanol mixtures at 298 K: (●) Py-PEG-Py(4800); (▲) Py-PEG-Py(11 200).

Table III
Relative Comparison of Measured I_e/I_m Values and Predicted k_{cy} Values

mol wt of Py-PEG-Py to be compared	$I_e/I_m(1)^a/I_e/I_m(2)$	$k_{cy}(1)^b/k_{cy}(2)$
9200 (1)–11 200 (2)	1.39	1.34
4800 (1)–9200 (2)	5.41	2.65
4800 (1)–11 200 (2)	7.48	3.56

[a] I_e/I_m values are taken from the plateau region in Figure 2. [b] $k_{cy} \propto N^{-3/2}$ where N is the length of the polymer chain.

The origin of the hydrophobic attraction is the reorientation of water molecules in the presence of dissolved nonpolar molecules.[25,26] The presence of a nonpolar molecule unable to form hydrogen bonds with water molecules causes the existing tetrahedrally coordinated water molecules to rearrange. The final structure of water molecules around a nonpolar molecule is a clathrate cage having more ordered water molecules than the bulk liquid. This is entropically very unfavorable, resulting in a large entropic contribution to the solvation free energy. As two nonpolar molecules approach one another they push ordered water molecules into the bulk, minimizing the volume of ordered water in the system and producing a net attraction. Israelachvili and Pashley[27] measured the hydrophobic force for linear alkyl chains with a crossed mica cylinder surface force measurement apparatus. They found an exponentially decaying attraction, corresponding to the theory proposed by Marcelja et al.,[28] of range comparable to that of London dispersion forces and of size exceeding that of the London interaction by an order of magnitude.

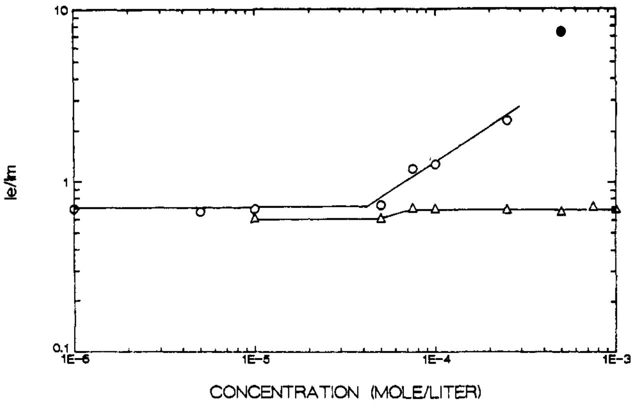


Figure 7. I_e/I_m ratio with total polymer concentration in deionized water: (○) 100% Py-PEG-Py(4800); (△) 1% Py-PEG-Py(4800) + 99% PEG(4800) mixture; (●) phase separation observed.

Our experiments can be interpreted in terms of hydrophobic interactions in the following manner. First, in the intramolecular excimer formation region Py-PEG-Py(4800) has an I_e/I_m value exceeding that predicted from the Wilemski–Fixman theory based solely on the diffusion-controlled process. The end-to-end distance of PEG(4800) is much smaller than that of the other PEG samples, as shown in Table I. Since the hydrophobic interactions decay exponentially with distance, we expect the hydrophobic interaction for Py-PEG-Py(4800) to be much stronger than for Py-PEG-Py of higher molecular weights. This is also supported by the fact that from Table II the observed I_e/I_m ratios in the intramolecular excimer formation region are higher than the calculated values, this tendency being most pronounced for the low molecular weight polymer.

Second, the phase separation occurred only for Py-PEG-Py(4800) at a high polymer concentration. In order to verify that this phase separation was not due to the self-aggregation of poly(ethylene glycols)[29] but due to the hydrophobic association among pyrene groups, we performed another experiment. The I_e/I_m ratio was measured over a range of polymer concentrations for a 1:99 mixture of Py-PEG-Py(4800) and PEG(4800) by weight. The results for the mixture are compared with the results of pure Py-PEG-Py(4800) in Figure 7. No phase separation was detected for the mixture even above 5×10^{-4} mol/L, proving that untagged PEG does not contribute to the phase separation. The nearly constant I_e/I_m value shows that there is no intermolecular excimer formation for the mixture. In other words, this indicates that the phase separation only occurs by the intermolecular association of pyrene groups.

Third, the substantial drop in I_1/I_3 for the phase-separated Py-PEG-Py(4800) also supports the hydrophobic association of pyrene groups, since such an association would create an environment enriched in organic species producing a low I_1/I_3 ratio.

2. Pyrene-End-Labeled Poly(ethylene glycols) in the Water–Methanol Cosolvent. The observations in the water–methanol cosolvent system can be interpreted in terms of preferential solvation. Since I_e/I_m is not correlated with the inverse viscosity of the solution in the region of the intramolecular excimer formation, we conclude that diffusion is not the only factor that influences the phenomena involved in this particular solvent system. As discussed earlier, there is an additional hydrophobic attraction due to the variation of water structure around the nonpolar pyrene groups for Py-PEG-Py in pure water. The addition of methanol, a less polar (dipole moment $D = 1.70$, dielectric constant $\epsilon = 32.7$) solvent than water ($D = 1.85$, $\epsilon = 78.54$)[9] most likely displaces water from hydrophobic pyrene groups disrupting the clathrate structure and reducing the hydrophobic attraction. The effect is more pronounced for Py-PEG-Py(4800) than Py-PEG-Py(11 200), as shown in Figure 5. The reduction of the effect of the hydrophobic interaction by the addition of methanol seems to persist up to 90% methanol content. The slight increase of I_e/I_m in pure methanol is probably due to the significant decrease in solvent viscosity. The observation from Figure 6 that the decrease in $(I_e/I_m)\eta_0$ in the region of methanol content above 20% is much faster for Py-PEG-Py(4800) than for Py-PEG-Py(11 200) also implies that the initial hydrophobic attraction is much stronger for low molecular weight polymer than for high molecular weight polymer and that this strong hydrophobic attaction is more strongly affected by the presence of methanol molecules. The paper by Cheung et al.[7] reported unusually high I_e/I_m in both water and methanol, uncorrelated with the other solvents tested. As an extension, Oyama et al.[4] found that the deviation of I_e/I_m for aqueous solutions from the value expected from the diffusion-controlled cyclization process was considerably larger for the shorter PEG chains. These observations are consistent with the hypothesis that enhanced intra- and intermolecular association is due to hydrophobic attractions between pyrene groups.

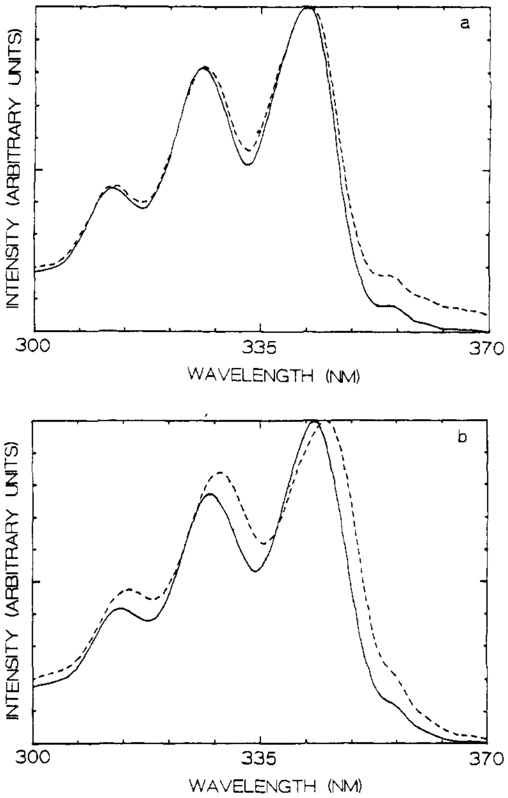


Figure 8. Normalized excitation spectra of Py-PEG-Py(4800): (a) in methanol and (b) in water; solid line = monomer emission, dashed line = excimer emission

In order to see the difference of excimer formation in either water or methanol, we examined the excitation spectra. Excitation spectra for Py-PEG-Py(4800) in each solvent are shown in Figure 8. In methanol, the excitation spectra of monomer emission monitored at 376 nm and excimer emission monitored at 500 nm are almost identical, implying that most of excimers form via a diffusion-controlled process. This also means that Birks' kinetic scheme[31] assuming diffusion-controlled excimer formation generally holds in this particular solvent. On the contrary,

Table IV
Results from the Lifetime Measurement of Excimer[a] for the Fluorescence of Py-PEG-Py(4800) at 298 K

MeOH %	A_1	τ^b	A_2	τ^c	$\chi^{2\,d}$
0	0.35	66.2	−0.05	83.7	1.55
100	0.88	79.9	−0.80	39.9	1.48

[a] $I_E(t) = A_1 \exp(-\lambda_1 t) + A_2 \exp(-\lambda_2 t)$. [b] $\tau_1 = \lambda_1^{-1}$ (ns). [c] $\tau_2 = \lambda_2^{-1}$ (ns). [d] Standard χ^2 displacement in the lifetime measurement. Time scale for fit, 350 ns.

the excitation spectra of excimer in water is red-shifted by about 2 nm with respect to that of monomer as shown in Figure 8b. This also corresponds to the results by Oyama et al.[4] and Winnik et al.[32] The red shift is due to the preformed ground-state excimer complex and we believe that it originates from the hydrophobic attraction between nonpolar pyrene molecules. Thus it is expected that Birks' scheme is not valid in this case.

We also measured the excimer lifetime of pyrene-end-labeled poly(ethylene glycol)(4800). We choose to present the excimer lifetime since it more closely represents the rate of end-to-end association on the pyrene-disubstituted polymer chain.[6] The excimer fluorescence data were fit with a double exponential and the decay times and preexponential factors are tabulated in Table IV. According to Birks' kinetic scheme[31] the ratio of preexponential factors, A_1/A_2, should be -1 for the initial condition at which the excimer concentration is zero at the time of excitation. As you notice from Table IV, the ratio A_1/A_2 in pure methanol is close to -1 implying that Py-PEG-Py(4800) in methanol generally follows Birks' scheme. Also notice that the lifetime values in methanol are in the same range as those from pyrene-end-labeled polystyrene of comparable chain length at Θ condition.[6] Conversely, the ratio A_1/A_2 in pure water is -7, which deviates from the expected -1, indicating that Birks' scheme is not valid in water. We also observed the growing-in component in excimer lifetime contrary to the results of Oyama et al.[33] and Winnik et al.[32] even though the contribution of that component to the total intensity is less than 15%. This is probably due to the narrow lamp profile of the hydrogen lamp of this work compared to that of the nitrogen lamp utilized in the other studies. From these lifetime measurements we infer that there are large portions of preformed excimers in water invalidating the initial condition made in Birks' scheme. Therefore, the existence of a preformed excimer in water is also consistent with our hypothesis that hydrophobic attraction between the pyrene tags exists in water.

Summary

Excimer formation in aqueous solutions of pyrene-end-labeled poly(ethylene glycols) of three molecular weights was studied as a function of polymer concentration. In Py-PEG-Py(4800) the excimer to monomer ratio, I_e/I_m, is much higher than predicted by the Wilemski–Fixman theory based on the diffusion-controlled end-to-end cyclization. Moreover, for this particular polymer phase separation occurred at high polymer concentration. We attribute this to a hydrophobic attraction between pyrene groups. The addition of methanol decreases the hydrophobic interaction by the preferential solvation of the pyrene groups by methanol. This effect is most obvious for Py-PEG-Py(4800) where the hydrophobic attraction is strongest.

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