Macromolecules **1997,** *30,* 2633−2641

Cyclization Rates for Two Points in the Interior of a Polymer Chain

Sam Lee and Mitchell A. Winnik*

Department of Chemistry and Erindale College, University of Toronto, Toronto, Ontario, Canada M5S 1A1

Received September 10, 1996; Revised Manuscript Received January 22, 1997[⊗]

ABSTRACT: The fluorescence spectra and decay curves of a series of poly(ethylene glycols), EO_x-Py-EO_{87}-Py-EO_x, were obtained in dilute solution. These pyrene-labeled polymers have a central chain of 87 (number average) monomer units separating the two pendant pyrene groups and two flanking end chains of equal length, x, where $x = 2$, 12, 17, or 47 (number average) monomer units. In toluene, tetrahydrofuran, and benzene, small decreases in the cyclization rates of these polymers were observed with increasing length of the end chains. The rate of ring opening in the chain interior was found to be twice as fast as ring opening at the chain ends. These results are consistent with theoretical predictions of interior cyclization and the effects of excluded volume, respectively. Rates of end-to-end cyclization were found to be slower than expected by comparison to the rates of interior cyclization. Also, dilute solutions of interior-labeled polymer show small amounts of pyrene dimers which are preassociated intramolecularly. These results are believed to arise from differences in the local concentration of pyrene in interior- and end-labeled polymers. In water, there is evidence of hydrophobic association of the pyrene groups, and the extent of preassociation decreases with increasing chain length.

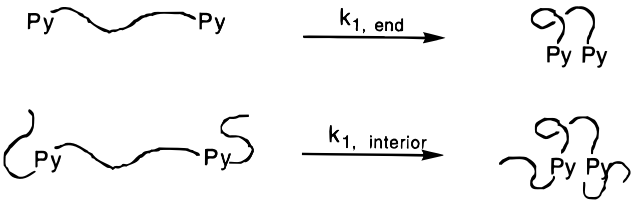
Introduction

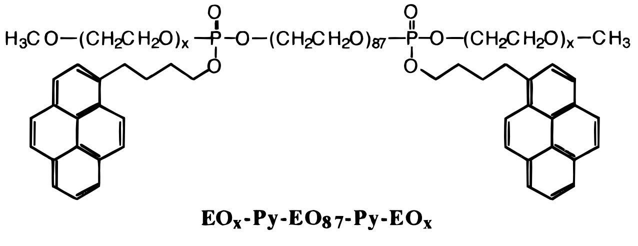
One of the last unexplored issues about polymers in dilute solution concerns the relative rates of motion of the chain ends compared to two segments separated by a similar contour length, but in the chain interior.[1] It is well-known that monomer units at the chain end undergo local motions more rapidly than their counterparts within the chain.[2] However, large-amplitude motion of sufficient scale to be described by Rouse and Zimm models is much more difficult to study experimentally. These are, for example, the motions leading to polymer cyclization.

For many years, we have studied a variety of linear polymers labeled with pyrene at the chain ends.[3] In these systems, the kinetics of intramolecular pyrene excimer formation provides information on the dynamics and thermodynamics of end-to-end cyclization. However, the case where the two excimer-forming chromophores occupy well-defined sites in the chain interior has never been examined. Excimer formation in these polymers involves the backbone-to-backbone cyclization of the chain. If a series of polymers labeled in the chain interior were available, their cyclization rates, $k_{1,\mathrm{interior}}$, compared with $k_{1,\mathrm{end}}$ of a corresponding end-labeled polymer would provide information on the extent to which the hydrodynamic drag of the tails may retard interior cyclization (Scheme 1).

Recently, we described the synthesis of a series of monodisperse poly(ethylene glycols) (PEGs), EO_x-Py-EO_{87}-Py-EO_x, which are characterized by a central chain of fixed length spanning the two pyrene labels.[4] The central chain in each sample is flanked by end chains (or tails) of a different length. These polymers allow us to determine how the rate of internal cyclization is affected by the molecular weight of the end chain.

Scheme 1





EO_x-Py-EO_{87}-Py-EO_x

There are theoretical studies of the conformation[5−7] and dynamics[1,8,9] of subchains located within the interior of the chain contour. In θ solvents, the dimensions of the central chain that spans the two labeled segments is independent of the length of the end chains. Under excluded-volume conditions (i.e., good solvents), most theories[5,6d,f] predict that the central chain swells more with increasing length of the end chains. Friedman and O'Shaughnessy have obtained scaling relationships for interior cyclization in the case where the relative position of the two cyclizing groups remains fixed; i.e., $N_{\mathrm{end1}}/N_{\mathrm{central}}$ and $N_{\mathrm{end2}}/N_{\mathrm{central}}$ remain constant while the total number of chain segments, $N = N_{\mathrm{central}} + N_{\mathrm{end1}} + N_{\mathrm{end2}}$, can change. Interior cyclization of polymers having the structure of our labeled PEGs—where N_{central} remains fixed while $N_{\mathrm{end1}} = N_{\mathrm{end2}}$ is varied—has been considered by Perico and Beggiato.[1,8] They found a small, but asymptotic decrease in k_1 with increasing length of the end chains. This effect becomes more pronounced as the capture strength of the fluorescent probe increases and also as the flexibility of the chain decreases.

This paper presents the results of our fluorescence studies of these labeled PEGs in dilute solutions with good solvents and compares their cyclization rates with the theoretical results of Perico and Beggiato (which refer to a θ solvent). The cyclization rate constants for these PEG bis(phosphates), EO_x-Py-EO_{87}-Py-EO_x, are also compared to those of an end-labeled diester, Py-

---

[⊗] Abstract published in *Advance ACS Abstracts,* March 15, 1997.

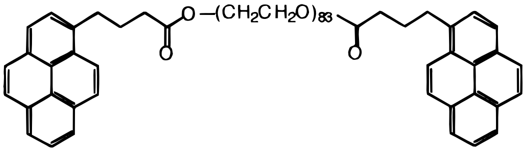
S0024-9297(96)01351-4 CCC: $14.00

Table 1. Molecular Weight Data for the Pyrene-Labeled Polymers Determined by MALDI Mass Spectrometry and NMR Spectroscopy

polymer[a]	M_w[b]	M_n[b]	M_n (central)	M_n (end)[e]	x[f]
Py-EO_{87}-Py	4269 ± 9	4232 ± 11	3658 ± 11[c]		0
EO_2-Py-EO_{87}-Py-EO_2	4714 ± 11	4692 ± 12	3823 ± 8[d]	89	2
EO_{12}-Py-EO_{87}-Py-EO_{12}	5628 ± 8	5607 ± 8	3823 ± 8[d]	571 ± 44	12
EO_{17}-Py-EO_{87}-Py-EO_{17}	5920 ± 6	5897 ± 7	3823 ± 8[d]	791 ± 110	17
EO_{47}-Py-EO_{87}-Py-EO_{47}	8427 ± 13	8409 ± 11	3823 ± 8[d]	2102 ± 187	47

[a] Subscripts, x, represent expected number average of oxyethylene units. In the case of $x = 2$, the end chain is 2-ethoxyethanol. [b] Determined by MALDI mass spectrometry of the labeled polymer. [c] Calculated from M_n of the labeled polymer. [d] Determined by MALDI mass spectrometry of the central chain before the end chains were chemically attached. [e] Determined by ^{1}H NMR of the end chain before it was chemically attached to both ends of the central chain. [f] Calculated from M_n (end).

EO_{87}-Py, where the PEG chain is of the same length as the central chain of the bis(phosphates), to see how interior cyclization differs from end-to-end cyclization.

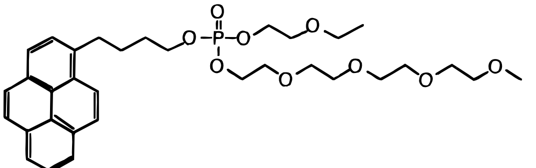


Py-EO87-Py

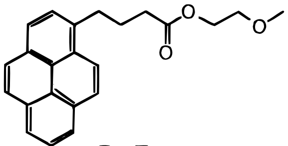
Experimental Section

Spectroscopic grade organic solvents (Caledon Laboratories) were freshly distilled under nitrogen prior to use. Tetrahydrofuran (THF) was refluxed and distilled from lithium aluminum hydride. Toluene was refluxed and distilled from sodium. Benzene was refluxed and distilled from a 50 wt % dispersion of sodium in paraffin. Distilled water was treated with a Millipore Milli-Q water system.

The synthesis and characterization of the bis(phosphates), EO_x-Py-EO_{87}-Py-EO_x, the diester, Py-EO_{87}-Py, and the Py-Phosphate model compound have been reported elsewhere.[4,10] The Py-Ester model compound was synthesized by esterification of pyrenylbutyric acid with 2-methoxyethanol. The molecular weights of the labeled polymers are summarized in Table 1.



Py-Phosphate

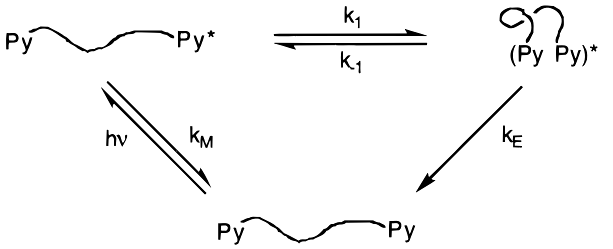


Py-Ester

The poly(ethylene glycol) used to construct the central chain in all of these polymers had a molecular weight of $M_n = 3841$ (MALDI),[4] which corresponds to a number average of 87 oxyethylene monomer units. Our characterization results indicated that the PEG chain in Py-EO_{87}-Py was only an average of 83 monomer units long.[10] However, this small difference is not great enough to affect the rate of cyclization. When k_1 for this polymer was corrected (using data from two previous investigations[11]) to the cyclization rate expected for a longer PEG chain with 87 monomer units, its value did not change.

Dilute solutions of these polymers and the model compounds—~(0.5−4) × 10^{-6} M to avoid the formation of intermolecular excimers—were prepared in 1 cm square Pyrex cells, and they were degassed by purging with prepurified argon (Canox, <3 ppm oxygen).

Scheme 2



Fluorescence spectra were recorded on a Spex Fluorolog 2 spectrometer. The emission spectra were not corrected for light scattering or for the wavelength sensitivity of the photodetector. Decay curves were obtained by the time-correlated single-photon-counting technique.[12] The excitation source was a coaxial flash lamp (Edinburgh Instruments, Model 199F) filled with deuterium. The excitation wavelength was selected by an ISA Instruments monochromator and that of the fluorescence by a Spex 1760 monochromator. The analyses of the decay curves were performed with the δ-pulse deconvolution method[13] using iterative weighted linear least-squares analysis to find the parameters of the fluorescence decay curves having the best fit. For the lifetime measurements, the excitation wavelength was 345 nm; the fluorescence from the pyrene "monomer" and excimer were monitored at 376 and 450 nm, respectively.

Data Analysis. The cyclization of pyrene end-labeled polymers has been described by Birks' scheme (Scheme 2).[14] k_1 is the cyclization rate constant, and k_{-1} describes the rate of excimer dissociation and ring opening. k_M and k_E are the reciprocal lifetimes of the excited pyrene monomer and excimer, respectively. Scheme 2 predicts that after a δ-pulse excitation, the intensity of the resulting monomer fluorescence, $I_M(t)$, should decay as the sum of two exponential terms, whereas the intensity of the excimer, $I_E(t)$, should increase and then decrease as the difference of the same two terms:

$$I_M(t) = a_1 \exp(-\lambda_1 t) + a_2 \exp(-\lambda_2 t) \tag{1}$$

$$I_E(t) = a_3 \exp(-\lambda_1 t) + a_4 \exp(-\lambda_2 t) \tag{2}$$

where

$$2\lambda_{1,2} = [X + Y \mp \{(Y - X)^2 + 4k_1k_{-1}\}^{1/2}] \tag{3}$$

$$X = k_M + k_1; \quad Y = k_E + k_{-1} \tag{4}$$

and

$$\frac{a_2}{a_1} = \frac{(X - \lambda_1)}{(\lambda_2 - X)}; \quad \frac{a_4}{a_3} = -1 \tag{5}$$

k_1, k_{-1}, and k_E can be calculated from the decay parameters, λ_1, λ_2, and a_2/a_1, once we know the monomer lifetime in the absence of quenching, $\tau_M = 1/k_M$. τ_M values were obtained from the monoexponential decay of dilute solutions (~5 × 10^{-7} M) of the corresponding model compound, Py-Phosphate or Py-Ester.

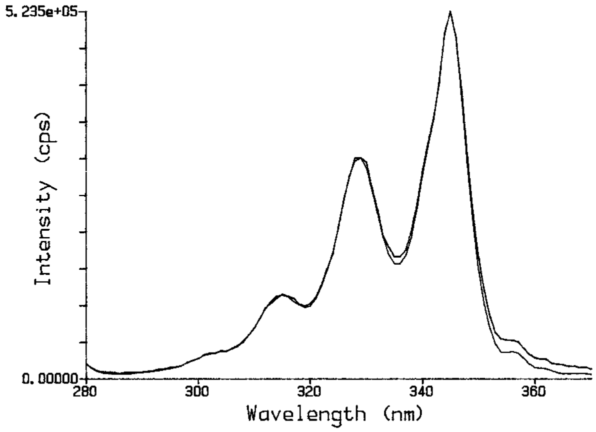


Figure 1. Excitation spectra of EO_{47}-Py-EO_{87}-Py-EO_{47} in THF, normalized to the peak at 345 nm. The spectrum showing enhanced absorption above ~350 nm was obtained by monitoring the excimer emission at 500 nm. The other spectrum was obtained from the monomer emission at 375 nm.

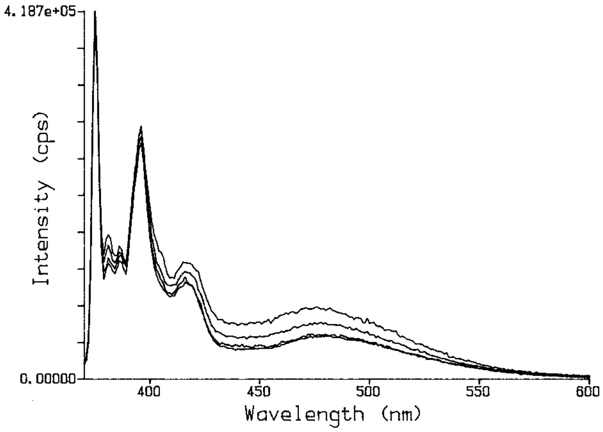


Figure 2. Fluorescence spectra of EO_{12}-Py-EO_{87}-Py-EO_{12} in THF, normalized to the 0−0 peak at 375 nm. From top to bottom in excimer intensity: $\lambda_{ex} = 365$, 360, 353, and 345 nm.

The decay parameters were obtained from eqs 2 and 6. We determined from the characterization of Py-EO_{87}-Py that a

$$I_M(t) = a_1 \exp(-\lambda_1 t) + a_2 \exp(-\lambda_2 t) + a_M \exp(-\lambda_M t) \tag{6}$$

fraction of the chains was esterified at one end only.[10] We did not detect any singly labeled chains in the bis(phosphates), but we did find in all of those samples traces of an oxidized polymer (i.e. **15** and **16** in ref 4) containing a ketopyrene group.

$$f = \frac{a_1 + a_2}{a_1 + a_2 + 2a_M} \tag{7}$$

Fluorescence from these pyrenyl ketone groups is not detected under the conditions used for our fluorescence measurements. However, pyrene monomer fluorescence from the singly labeled chains in the diester and from the oxidized polymer chains in the bis(phosphates) appears as a minor, long-lived third component in the $I_M(t)$ decays. Hence, the monomer decays were fit to eq 6 instead of eq 1.[15]

The fraction f of chains in these polymer samples that contain two alkylpyrene groups can be estimated from the preexponential factors:[15b] Our $I_M(t)$ measurements indicated that ~80% of the chains in the Py-EO_{87}-Py diester sample were functionalized at both ends. For the bis(phosphates), f ranged from 87 to 95%, depending on the polymer. These results are consistent with our NMR and MALDI mass spectrometry characterization data.[4,10]

In the case of the bis(phosphates) EO_{12}-Py-EO_{87}-Py-EO_{12}, EO_{17}-Py-EO_{87}-Py-EO_{17}, and EO_{47}-Py-EO_{87}-Py-EO_{47}, the ratio a_4/a_3 from the $I_E(t)$ decays deviated significantly from −1 in all organic solvents when the above scheme was used. Also, the excitation spectra obtained by monitoring the excimer emission from these bis(phosphates) show slightly increased absorption at wavelengths greater than ~350 nm compared to those obtained by monitoring the monomer emission (Figure 1). This feature is accentuated by observing the emission spectra of these polymers as a function of excitation wavelength, λ_{ex} (Figure 2). The ratio of the excimer to the monomer fluorescence intensity (I_E/I_M) is constant up to $\lambda_{ex} = 350$ nm, and then it increases with longer wavelengths (Figure 3). These observations suggest the presence of a small amounts of preassociated dimers of pyrene in solution which give excimer directly upon excitation (or it is possible that some chains have the pyrene groups in close proximity so that those excimers are formed at a rate that is faster than the time resolution of our fluorescence measurements).[16]

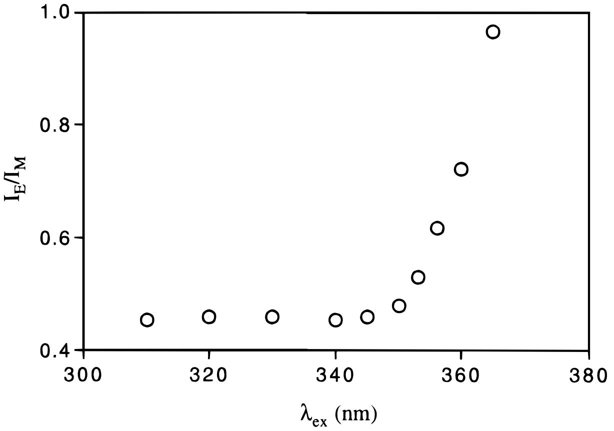
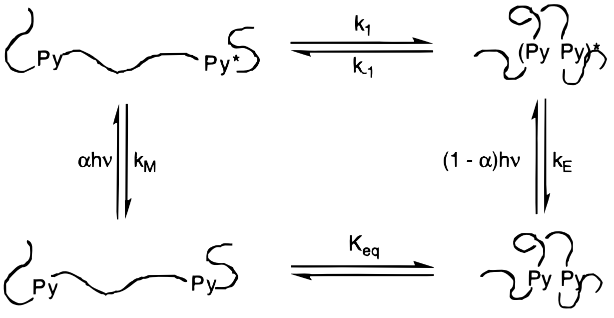


Figure 3. Intensity of excimer-to-monomer fluorescence (I_E/I_M) as a function of excitation wavelength (λ_{ex}) for EO_{12}-Py-EO_{87}-Py-EO_{12} in THF.

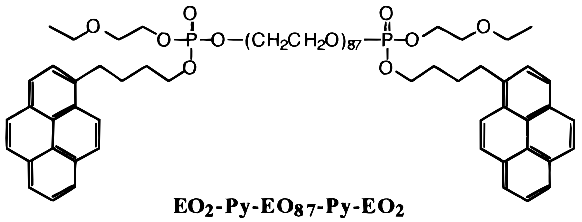
Scheme 3



In this case, Scheme 2 must be modified to consider an equilibrium between two light-absorbing species. In Scheme 3, α is the fraction of light absorbed by the nonassociated pyrenes and K_{eq} is the equilibrium constant between these two ground-state species. According to this scheme, eqs 1−4 remain valid, but the fraction of light absorbed by the preassociated dimers, (1 − α), changes the ratios of the preexponential factors from the $I_M(t)$ and $I_E(t)$ decays:[16,17]

$$\frac{a_2}{a_1} = \frac{\alpha(X - \lambda_1) - (1 - \alpha)k_{-1}}{\alpha(\lambda_2 - X) + (1 - \alpha)k_{-1}}; \quad \frac{a_4}{a_3} = \frac{(1 - \alpha)(Y - \lambda_1) - \alpha k_1}{(1 - \alpha)(\lambda_2 - Y) - \alpha k_1} \tag{8}$$

The rate constants in the organic solvents were calculated from the decay parameters using eqs 3, 4, and 8. In the absence of preassociation, α = 1 and the ratios in eq 8 reduce to those of eq 5. When the rate constants of the two labeled polymers that show no evidence for preassociation—Py-EO_{87}-Py and EO_2-Py-EO_{87}-Py-EO_2—are evaluated using Scheme 3, the results are identical, within experimental error, to those obtained by using Scheme 2.



EO_2-Py-EO_{87}-Py-EO_2

Results

Organic Solvents. The polymers were examined in THF, toluene, and benzene. An example of their fluorescence spectra is shown in Figure 4. It is possible to use the I_E/I_M ratios from the fluorescence spectra to compare rates of cyclization among a series of labeled polymers if the value k_{FE}/k_{FM} $(k_{-1} + k_E)$, where k_{FE} and k_{FM} are the rates of excimer and monomer fluorescence, respectively, is the same for all of these polymers and there is no preassociation.[14,15b] In these polymers, variations in their k_{-1} values and preassociation among some of the bis(phosphates) make such comparisons less reliable. However, these fluorescence spectra do suggest that the polymers with longer end chains have lower rates of cyclization. The rate constants obtained from analysis of the fluorescence decay measurements are summarized in Table 2.

Water. All of the labeled polymers have I_E/I_M ratios that are larger in water than in any of the organic solvents (Figure 5). This enhanced excimer formation in water is due to the hydrophobic association between the two pyrene groups on each polymer chain.[17] The strongest evidence for this effect is provided by the excitation spectra. Figure 6 shows a typical example. The excitation spectra obtained by monitoring the excimer emission of all polymers at 500 nm were red-shifted by 1 nm relative to those obtained by monitoring the monomer emission at 375 nm. These fluorescence results indicate that the extent of pyrene preassociation in water is much greater than that observed in organic solvents.

However, Scheme 3 could not be used to evaluate the fluorescence lifetime data. All of the $I_E(t)$ measurements did have rise and decay components, but they could not be fitted to a difference of two exponential terms. Three exponentials were necessary for satisfactory fits to the data, and all of these exponential terms were different from those obtained from the $I_M(t)$ decays. This suggests that there is more than one rate describing excimer formation in these systems. It is possible that the chains are aggregating so that intermolecular excimers are being formed in addition to intramolecular ones. This possibility is being investigated further.

The mean decay time, $\langle\tau\rangle$ in eq 9, represents the average lifetime of the excited pyrene monomer before

$$\langle\tau\rangle = \frac{a_1\tau_1 + a_2\tau_2}{a_1 + a_2} \tag{9}$$

it returns to the ground state by fluorescence, radiationless deactivation, and quenching. Hence, it provides a qualitative description of pyrene quenching. Since we did not have a model to evaluate the decay data in water, rates of excimer formation were compared by using the mean decay times of the polymers. These values (Table 3) were calculated from the parameters obtained by fitting the $I_M(t)$ decays to eq 6. Typical $\langle\tau\rangle$ values for the polymers in organic solvents are also presented in Table 3 for comparison.

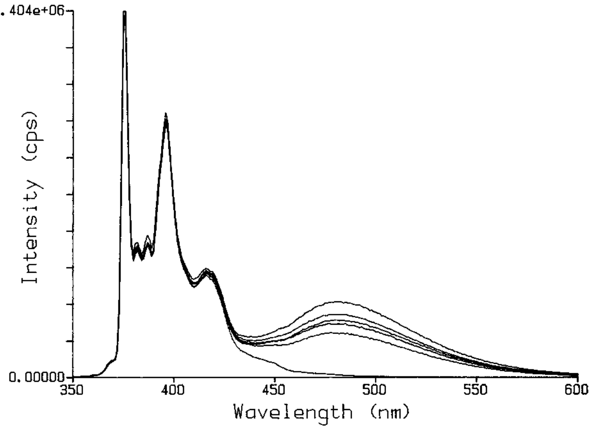


Figure 4. Fluorescence spectra of the labeled polymers in toluene, normalized to the 0–0 peak at 376 nm. λ_{ex} = 345 nm. From top to bottom in excimer intensity: Py-EO_{87}-Py, EO_{12}-Py-EO_{87}-Py-EO_{12}, EO_2-Py-EO_{87}-Py-EO_2, EO_{17}-Py-EO_{87}-Py-EO_{17}, and EO_{47}-Py-EO_{87}-Py-EO_{47}.

Discussion

Pyrene Preassociation in Organic Solvents. Our fluorescence spectra and the α values in Table 2 show that preassociated pyrenes can only be detected in solutions of the internally labeled PEGs and not in the end-labeled ones. At this time, it is not known why this effect only appears in the internally labeled PEGs. It is possible that such behavior could be associated with the fact that the pyrenes in these polymers are attached to the labeled segments via flexible side chains. This behavior might also indicate that the local concentration of two interior-labeled segments is greater than that of two end-labeled segments.

Effect of the Functional Group Linking the Pyrene to the Polymer. It is possible that, in fluorescence measurements using labeled polymers, the size of the probe may affect the cyclization rate through its frictional resistance to motion. Here, the molecular weight of the phosphate ester end groups of EO_2-Py-EO_{87}-Py-EO_2 is 1.5 times larger than the pyrenebutyrate ends of Py-EO_{87}-Py. Depending on the solvent, k_1 of Py-EO_{87}-Py is 33–47% larger than that of EO_2-Py-EO_{87}-Py-EO_2.

In previous investigations using polystyrene chains of greater molecular weight than the polymers studied here,[3c,18] the size of the end group had no observable effect on k_1. However, in these shorter and more flexible chains, the end groups represent 14 and 21% of the molar mass of Py-EO_{87}-Py and EO_2-Py-EO_{87}-Py-EO_2, respectively. The slower rates of cyclization of EO_2-Py-EO_{87}-Py-EO_2 relative to that of Py-EO_{87}-Py can therefore be due, in part, to the resistance to motion of the bulkier end groups of the bis(phosphate). It is less likely that the dangling 2-ethoxyethyl groups of the bis(phosphate) would present significant steric constraints to cyclization since the oxygen linkages in this side chain and in the phosphate group are highly flexible.

Differences between the Photophysical Behavior of Terminally and Internally Labeled Polymers. Stronger correlations exist between a pair of monomer units in the chain interior compared to two end units.[5] Figure 7 shows that the rates of excimer reversion for the internally labeled PEGs are about twice as fast as that of the end-labeled PEGs in organic solvents. This result illustrates the stronger excluded-volume effects that are operative on the internal segments of the chain compared to those near the ends.

Table 2. Rate Constants[a] and α Values for the Labeled Polymers in THF, Toluene, and Benzene According to Scheme 2

	THF	benzene	toluene
Py-EO$_{87}$-Py			
k_1	4.8 (±0.1) × 10^6	4.8 (±0.2) × 10^6	5.9 (±0.3) × 10^6
k_{-1}	5.4 (±0.4) × 10^6	3.9 (±0.6) × 10^6	3.9 (±0.4) × 10^6
k_E	1.8 (±0.1) × 10^7	1.9 (±0.1) × 10^7	1.9 (±0.1) × 10^7
α	0.99	0.98	0.98
EO$_2$-Py-EO$_{87}$-Py-EO$_2$			
k_1	3.4 (±0.2) × 10^6	3.6 (±0.3) × 10^6	4.0 (±0.3) × 10^6
k_{-1}	4.1 (±0.6) × 10^6	4.1 (±0.5) × 10^6	3.7 (±0.4) × 10^6
k_E	1.8 (±0.1) × 10^7	1.7 (±0.1) × 10^7	1.7 (±0.1) × 10^7
α	0.99	0.99	0.99
EO$_{12}$-Py-EO$_{87}$-Py-EO$_{12}$			
k_1	5.2 (±0.4) × 10^6	3.7 (±0.3) × 10^6	6.5 (±0.3) × 10^6
k_{-1}	9.8 (±0.2) × 10^6	7.8 (±0.6) × 10^6	10.4 (±0.6) × 10^6
k_E	1.3 (±0.1) × 10^7	1.6 (±0.1) × 10^7	1.6 (±0.1) × 10^7
α	0.92	0.93	0.89
EO$_{17}$-Py-EO$_{87}$-Py-EO$_{17}$			
k_1	4.6 (±0.2) × 10^6	4.4 (±0.3) × 10^6	4.6 (±0.3) × 10^6
k_{-1}	1.0 (±0.1) × 10^7	9.6 (±1.2) × 10^6	7.9 (±0.3) × 10^6
k_E	1.6 (±0.1) × 10^7	1.5 (±0.1) × 10^7	1.8 (±0.1) × 10^7
α	0.95	0.93	0.94
EO$_{47}$-Py-EO$_{87}$-Py-EO$_{47}$			
k_1	4.0 (±0.2) × 10^6	3.6 (±0.2) × 10^6	4.0 (±0.2) × 10^6
k_{-1}	8.9 (±0.4) × 10^6	9.3 (±0.3) × 10^6	7.1 (±0.3) × 10^6
k_E	1.4 (±0.1) × 10^7	1.5 (±0.1) × 10^7	1.6 (±0.1) × 10^7
α	0.94	0.94	0.94

[a] Rates are in units of s^{-1}. Rates are reported as the average of four or five replicate measurements. Error values represent one standard deviation in the data.

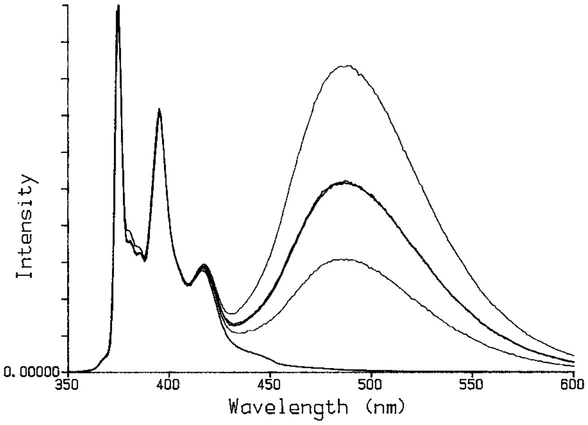


Figure 5. Fluorescence spectra of the labeled polymers in water, normalized to the 0–0 peak at 375 nm. λ_{ex} = 345 nm. From top to bottom in excimer intensity: EO$_2$-Py-EO$_{87}$-Py-EO$_2$, EO$_{12}$-Py-EO$_{87}$-Py-EO$_{12}$ and EO$_{17}$-Py-EO$_{87}$-Py-EO$_{17}$ (similar intensity), EO$_{47}$-Py-EO$_{87}$-Py-EO$_{47}$, and Py-Phosphate (no excimer).

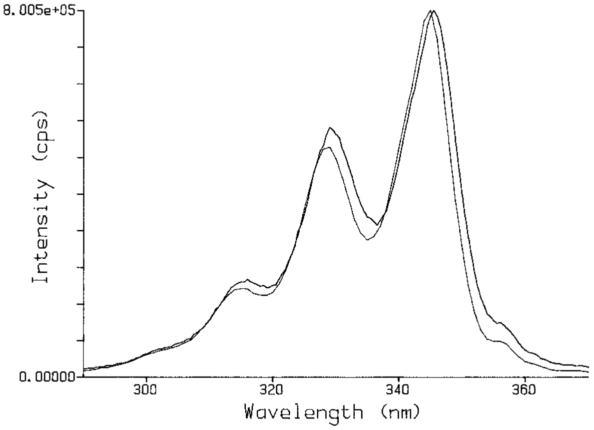


Figure 6. Excitation spectra of EO$_{47}$-Py-EO$_{87}$-Py-EO$_{47}$ in water, monitored at 375 and 500 nm. Both spectra are normalized to the peaks at 345 and 346 nm. The excimer excitation spectrum is red-shifted relative to that of the monomer.

Table 3. Mean Lifetimes, ⟨τ⟩, for the Labeled Polymers in Water (and in the Organic Solvents for Comparison)

	⟨τ⟩ (ns)			
polymer	water	toluene	THF	benzene
Py-EO$_{87}$-Py	6.9	103	121	108
EO$_2$-Py-EO$_{87}$-Py-EO$_2$	24.1	114	132	115
EO$_{12}$-Py-EO$_{87}$-Py-EO$_{12}$	41.5	113	134	126
EO$_{17}$-Py-EO$_{87}$-Py-EO$_{17}$	48.6	119	133	125
EO$_{47}$-Py-EO$_{87}$-Py-EO$_{47}$	60.9	123	141	132

With cyclization between the terminal points of a chain, the ends of two polymer strands are brought into proximity. Cyclization between two internally positioned monomer units brings the ends of four polymer strands together. The greater steric crowding in this latter case is responsible for additional repulsions at the point of ring closure.

In the poorest solvent for poly(ethylene glycol) examined here (toluene), k_{-1} for the internally labeled PEGs shows a slight decrease with longer end chains. This observation suggests that a reduction in solvent quality diminishes the excluded-volume effects more effectively between the inner parts of a chain. Such a conclusion is consistent with theoretical results which indicate that a subchain in a good solvent swells more when it is located nearer to the center of the polymer.[5,6d–f]

It should be noted that the attachment of chain segments two monomer units in length to the labeled ends of a polymer does not make a significant contribution to the correlation effects between those labeled units. For example, k_{-1} for EO$_2$-Py-EO$_{87}$-Py-EO$_2$, where the labeled segments are flanked by 2-ethoxyethyl tails, is smaller than that of the other bis(phosphates). This result would suggest that the large-amplitude motions characteristic of chain cyclization can be probed with any of the three monomer units closest to each chain end. Such a conclusion is also supported by a recent dynamic NMR study[19] of poly(ethylene oxides) in dilute aqueous solutions which found that the last three or four segments at each end of the chain have a dynamic behavior different from that of the main chain. Hence,

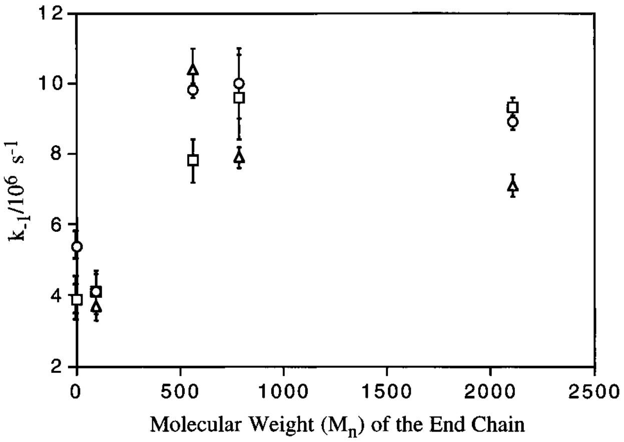


Figure 7. Plot of k_{-1} with error bars as a function of the number-average molecular weight (M_n) of the end chains of the labeled polymers in THF (○), benzene (□), and toluene (△). The points at $M_n = 0$ refer to Py-EO$_{87}$-Py.

Table 4. Equilibrium Constants for Cyclization, K_{cy}, for the Labeled Polymers in the Organic Solvents

	THF	benzene	toluene
Py-EO$_{87}$-Py	0.92	1.1	1.4
EO$_2$-Py-EO$_{87}$-Py-EO$_2$	0.83	0.88	1.1
EO$_{12}$-Py-EO$_{87}$-Py-EO$_{12}$	0.51	0.47	0.62
EO$_{17}$-Py-EO$_{87}$-Py-EO$_{17}$	0.46	0.43	0.57
EO$_{47}$-Py-EO$_{87}$-Py-EO$_{47}$	0.45	0.35	0.61

oxyethylene units are not expected to show dynamic behavior that is characteristic of the interior monomer segments until they are about four or five segments from the chain ends.

Equilibrium Constants for Cyclization. Table 4 lists the equilibrium constants for cyclization, $K_{cy} = k_1/k_{-1}$, in the organic solvents. K_{cy} provides a measure of the cyclization probability of these chains, so it is related to the conformational properties of the polymer. The most important feature in Table 4 is that K_{cy} accentuates the difference between the internal- and terminal-labeled chains. These K_{cy} values indicate that the fraction of internally cyclized chains in dilute solution is less than the fraction of terminally cyclized chains. This is another consequence of the stronger excluded-volume effects between two interior segments compared to that between the chain ends.

Rates of Internal Cyclization in Organic Solvents. Terminal cyclization rates are well-known to decrease with increasing molecular weight of the labeled polymer. Since the end-to-end separation, R_F, increases with chain length, the dependence of terminal cyclization rates on molecular weight is due largely to the effect of R_F rather than to the dynamics of the polymer coil.

In the case of internal cyclization with the polymers examined in this work, the two labeled segments in all of the bis(phosphates) are separated by the same number of monomer units. The average distance between these two labeled segments should, therefore, be the same for all of these bis(phosphates) under θ conditions. When excluded-volume effects are present, as would be expected from all of the solvents employed in this study, the polymer coil swells. This effect is molecular weight dependent, and the average distance, $\langle R_{ij}\rangle$, between the two phosphate groups of these internally labeled PEGs in good solvents is expected to increase with the length of the end chains.[5,6d-f] However, since the tails are shorter than the central chain and these are low molecular weight polymers, we do not expect $\langle R_{ij}\rangle$ to be affected significantly by the length of the end chains.

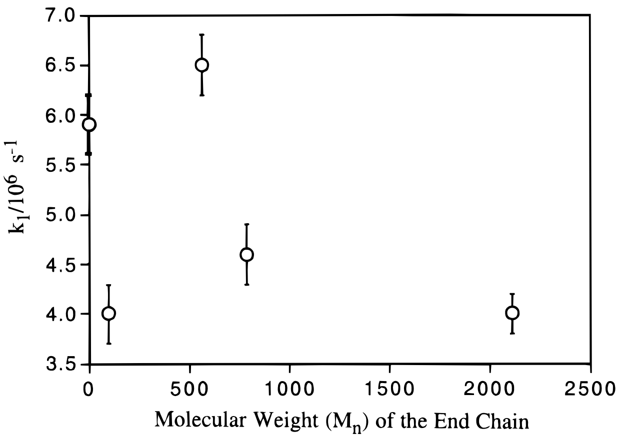


Figure 8. k_1 as a function of the number-average molecular weight (M_n) of the end chains of the labeled polymers in toluene. The point at $M_n = 0$ refers to Py-EO$_{87}$-Py.

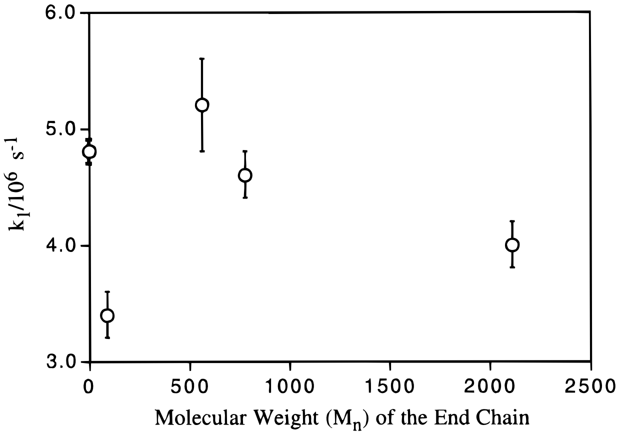


Figure 9. k_1 as a function of the number-average molecular weight (M_n) of the end chains of the labeled polymers in THF. The point at $M_n = 0$ refers to Py-EO$_{87}$-Py.

Nevertheless, the dependence of k_1 on $N_{\text{end chain}}$ can reflect differences in $\langle R_{ij}\rangle$ as well as in the dynamics of the polymer coil. Even though we expect the influence of the former to be less significant, the important point here is that these two effects act on k_1 in the same direction: longer end chains promote greater swelling of the central chain, and these longer end chains also exert greater drag on the interior groups undergoing cyclization. Both cause k_1 to decrease with increasing tail length.

Figures 8−10 plot k_1 as a function of the length of the end chains in the three solvents. In THF and toluene, k_1 decreases monotonically with increasing molecular weight of the end chains. The most noteworthy finding here is that this effect is small. A 3.8-fold increase in the length of the tails result in 23 and 38% decreases of k_1 in THF and toluene, respectively.

Perico and Beggiato have made some quantitative predictions about the relative changes in cyclization rates that can be expected for polymers where the two cyclizing segments are symmetrically displaced from the center of the chain.[8] Their theoretical calculations are consistent with the results obtained here. For example, the cyclization rate of a flexible polymer with 50 monomer units in the central chain and 20 monomer units in each of the end chains was calculated to be 6% smaller than that of the corresponding polymer containing only 10 monomer units in each of the tails. This

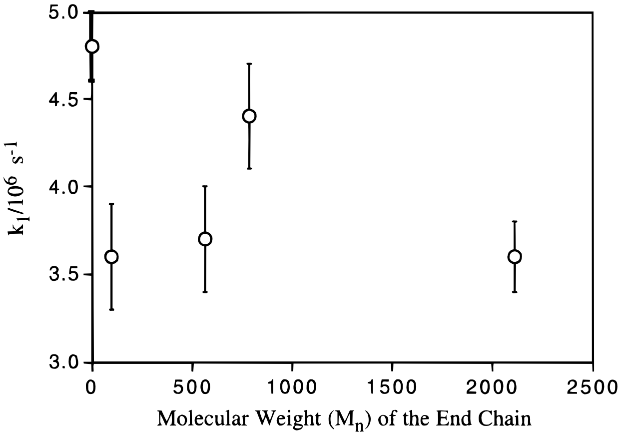


Figure 10. k_1 as a function of the number-average molecular weight (M_n) of the end chains of the labeled polymers in benzene. The point at $M_n = 0$ refers to Py-EO_{87}-Py.

decrease can be compared with that of two bis(phosphates) having similar relative lengths between the central and end chains: the difference between the k_1 of EO_{17}-Py-EO_{87}-Py-EO_{17} and EO_{47}-Py-EO_{87}-Py-EO_{47} ranges from 13 to 18%, depending on the solvent.

Perico and Beggiato also showed that the cyclization rate decreases to an asymptotic value with increasing tail length. This value is reached when the length of each end chain is somewhere between 0.5 and 2 times the length of the central chain, depending on the chain stiffness and on the capture strength of the two reactive groups. The trend of the cyclization rates shown in Figures 8−10 appears to support this prediction.

A simple rationalization for the slower motions of the internal segments relative to that of the chain ends is that two groups within the chain interior must drag their tails along in order to cyclize. This is a reasonable description when the end chains are much shorter than the central chain. Short tails affect k_1 largely through the frictional resistance to motion that they add to the cyclizing groups. In the other limit, where the end chains are much longer than the central chain, the motions of the two labeled segments are more restricted by the tails. In this case, the internal motions of the end chains become more important in bringing the cyclizing groups into proximity. Thus, a reasonable explanation for the asymptotic behavior of k_1, in the limit of very long end chains, is that monomer units along the tails that are remote from the labeled segments have little influence on the cooperative motions of the polymer that bring about the cyclization of the central chain.

Comparing Interior and End-to-End Cyclization Rates. The cyclization rate of a chain between two points is predicted to be faster with shorter end chains. However, the k_1 values for the "end-labeled" bis(phosphate) EO_2-Py-EO_{87}-Py-EO_2 is consistently small; in many cases this polymer cyclizes at a slower rate than the other internally labeled bis(phosphates). At this point, we do not have a clear understanding of why the measured end-to-end cyclization rate is slower than that of interior cyclization. We speculate that the local concentration of two interior-labeled segments is greater than that of two end-labeled segments.

Effect of the Solvent. End-to-end cyclization rates are well-known to be sensitive to the quality of the solvent. Poorer solvents decrease the coil dimensions and, more importantly, suppress excluded-volume effects. This leads to an enhancement in the k_1 values.

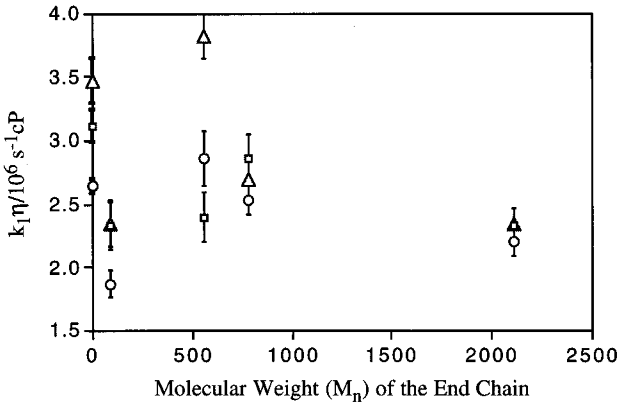


Figure 11. Cyclization rates corrected for solvent viscosity, $k_1\eta$, as a function of the number-average molecular weight (M_n) of the end chains of the labeled polymers in THF (○), toluene (△), and benzene (□). The points at $M_n = 0$ refer to Py-EO_{87}-Py.

The organic solvents used here are all good solvents for low molecular weight poly(ethylene glycol) at room temperature. The relative strength of these solvents for poly(ethylene glycol), based on their solubility parameters,[20] improves in the following order: toluene, THF, and benzene.

The cyclization rates corrected for solvent viscosity, $k_1\eta$, for all polymers are consistently faster in toluene compared with THF (Figure 11). This result agrees with the expected behavior of $k_1\eta$ with solvent quality. However, the sensitivity of these PEGs to the solvent appears to depend on the position of the two labeled segments in the chain. For the two end-labeled PEGs and for EO_{12}-Py-EO_{87}-Py-EO_{12}, their corrected cyclization rates are 1.3 times faster in toluene than in THF. With EO_{17}-Py-EO_{87}-Py-EO_{17} and EO_{47}-Py-EO_{87}-Py-EO_{47}, both $k_1\eta$ values are 1.1 times larger in toluene relative to THF. Long tails appear to have a greater affect on reducing the cyclization rate of the polymer when it is in a poorer solvent. This effect is probably related to the previously discussed tendency of the central chain in good solvents to swell more with increasing length of the end chain.

The equilibrium constants for cyclization of the labeled polymers in toluene are all larger than their corresponding K_{cy} values in THF (Table 4). This result is also consistent with our expectation that a larger fraction of the chains would be cyclized in the poorer solvent, toluene.

Even though benzene is the best of the three solvents for poly(ethylene glycol), the corrected cyclization rates of the labeled polymers in benzene are not consistently smaller than in THF or toluene. This result may be due to the interaction between this solvent and the functional groups that link the pyrene labels to the polymer. Benzene, being the least polar of the three solvents, is not expected to solvate the polar carboxylate and phosphate ester groups as effectively as THF or toluene. There is evidence from previous cyclization studies[15a] that k_1 can be affected by interactions between the solvent and the functional group used to attach the label to the polymer.

Fluorescence Behavior in Water. The fluorescence results in water can only be interpreted qualitatively, since we do not have a suitable model for analyzing the decay data. The results obtained here can be compared with those from a study by Duhamel et al.[17c] of pyrene end-labeled poly(ethylene glycol 8000)

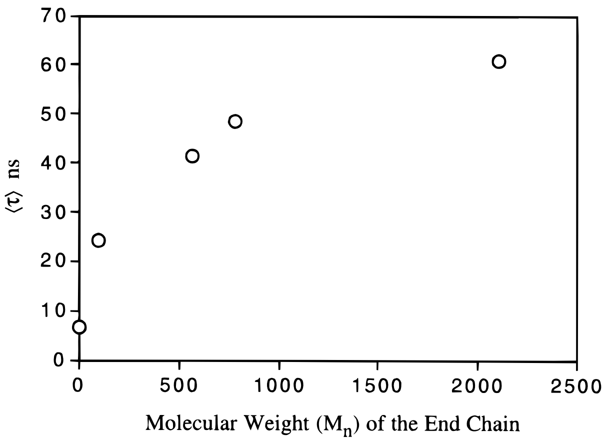


Figure 12. Mean lifetimes, $\langle\tau\rangle$, from the $I_M(t)$ decays as a function of the molecular weight of the tail segments of the labeled polymers in water. The point at $M_n = 0$ refers to Py-EO$_{87}$-Py.

in water. In those experiments, the end group was a PyCH$_2$O ether, which is less hydrophobic than the Py-(CH$_2$)$_4$O groups attached to the polymers examined here.

The $I_M(t)$ decays of dilute solutions of Py-PEG 8000-Py in water, reported by Duhamel et al.,[17c] were monoexponential. The $I_M(t)$ decays of the polymers in this work are not single exponential. Even the end-labeled PEGs, Py-EO$_{87}$-Py and EO$_2$-Py-EO$_{87}$-Py-EO$_2$, give complex monomer decays. The length of the chains separating the two pyrene labels in all of our PEGs are about half the length of poly(ethylene glycol 8000). With these shorter chains, the hydrophobic interactions between the pyrenes may be sufficient to cause some intermolecular association of the pyrene groups, so that we may be detecting a second rate of excimer formation due to aggregation of the chain ends. This possibility is supported by the work of Frank and co-workers, who found evidence for intermolecular association between pyrene end-labeled PEGs of M_w in the range 4800–11 200 in water.[17b]

Careful analysis of monomer and excimer decay profiles by Duhamel et al.,[17c] led them to deduce that ~7% of the Py-PEG 8000-Py chains in water have their two pyrene groups preassociated prior to excitation. The extent of preassociation in our shorter pyrene-labeled PEGs would be expected to be much greater, although we do not have enough information at this time to quantify the extent of association.

In systems with a relatively small fraction of preassociated pyrenes, mean monomer decay times are sensitive to the cyclization rate of those polymers whose pyrenes are not associated at the time of excitation. This presumes that excimers formed, either through cyclization or by direct excitation of preassociated pyrenes, do not have time to dissociate and make a significant contribution to the monomer fluorescence. Under these circumstances, ($\langle\tau\rangle^{-1} - \tau_o^{-1}$) is equal to the cyclization rate for these chains, where τ_o is the unquenched pyrene monomer decay time. In Figure 12 we plot $\langle\tau\rangle$ versus the length of tail for our five polymers. These $\langle\tau\rangle$ values increase with increasing tail length, becoming less dependent on the tail length with longer chains. This result indicates that the effect of tail length on k_1 is more pronounced with shorter chains. Another way of looking at the same data is that the cyclization rate of the chains with free pyrene end groups decreases as the fraction of the polymer mass representing the hydrophobic substituents decreases.

Fluorescence decay measurements can be used to assess the relative strength of the hydrophobic attraction between the ends of the bis(phosphate) EO$_2$-Py-EO$_{87}$-Py-EO$_2$ and between the ends of the dicarboxylate Py-EO$_{87}$-Py. We found from the $I_E(t)$ decays that most of the excimers from Py-EO$_{87}$-Py are formed within 1 ns after the lamp pulse, while the growth in the excimer intensity of EO$_2$-Py-EO$_{87}$-Py-EO$_2$ occurs within the first 3 ns after its excitation. The mean lifetime of Py-EO$_{87}$-Py calculated from its $I_M(t)$ decay is also shorter than that of EO$_2$-Py-EO$_{87}$-Py-EO$_2$. The slower rate of excimer formation and of pyrene quenching in EO$_2$-Py-EO$_{87}$-Py-EO$_2$ compared to Py-EO$_{87}$-Py suggests that the phosphate group is more strongly solvated by water than the carboxylate. Despite this effect of the polar phosphate group, the strength of the hydrophobic interaction between the alkyl pyrenes in all of the bis(phosphates) is still large; $\langle\tau\rangle$ decreases by 0.5–0.2-fold when the medium is changed from an organic solvent to water (Table 3).

Comparing the interior- and end-labeled polymers, the mean lifetimes for the internally labeled bis(phosphates) in organic solvents are larger than their corresponding values in water by a factor of 2.0–3.2. With the end-labeled EO$_2$-Py-EO$_{87}$-Py-EO$_2$, the mean lifetimes in organic solvents are 4.7–5.5 times the value obtained in water. The smaller rates of quenching with the internally labeled PEGs compared with EO$_2$-Py-EO$_{87}$-Py-EO$_2$ suggest that the hydrophobic effect is not as strong, or the chain is locally less mobile, for polymers with the two hydrophobes in the chain interior relative to those situated at the chain ends.

The hydrophobic effect observed here is often explained as an ordering of water molecules around a nonpolar molecule. This is entropically unfavorable. The hydrophobic interaction between two or more such nonpolar species displaces some of this water into the bulk phase, resulting in a net reduction in the number of ordered water molecules. The results obtained above are consistent with this description; the volume of solvent associated with two polymer segments is greatest when these units are situated at the chain ends.

Conclusions

A series of poly(ethylene glycol) polymers were examined in which a common block of 87 EO units separates two pyrene groups and serves as the point of attachment for EO$_x$ tails of different lengths, where x = 2, 12, 17, and 47. Steady-state and fluorescence decay measurements of pyrene excimer formation and dissociation were employed to determine the cyclization and ring-opening rates of these polymers as a function of solvent and tail length. The effect of the length of the tail on the rate of cyclization was relatively small, as predicted theoretically by Perico and Beggiato.[1,8] Important differences between end-labeled and interior-labeled polymers were found on the rates of ring opening in different solvents and on the equilibrium constants for cyclization. These point to differences in correlation effects associated with excluded volume between chain ends and points in the interior of the polymer. In water, effects associated with hydrophobic association of the pyrene substituents have a strong effect on the excimer kinetics.

Acknowledgment. The authors thank NSERC Canada for their support of this research.

References and Notes

(1) Perico, A. *Acc. Chem. Res.* **1989**, *22*, 336.

(2) (a) Bailey, R. T.; North, A. M.; Pethrick, R. A. *Molecular Motion in High Polymers*; Clarendon Press: Oxford, U.K., 1981. (b) Tassin, J. F.; Monnerie, L.; Fetters, L. J. *Macromolecules* **1988**, *21*, 2404.

(3) (a) Winnik, M. A. *Acc. Chem. Res.* **1985**, *18*, 73. (b) Winnik, M. A. Spectroscopic Studies of Cyclization Dynamics and Equilibria. In *Cyclic Polymers*; Semlyn, J. A., Ed; Elsevier: London, 1986. (c) Winnik, M. A. Cyclization as a Reaction in Restricted Space. In *Molecular Dynamics in Restricted Geometries*; Klafter, J., Drake, J. M., Eds; Wiley & Sons: New York, 1989.

(4) Lee, S.; Winnik, M. A.; Whittal, R. M.; Li, L. *Macromolecules* **1996**, *29*, 3060.

(5) des Cloizeaux, J.; Jannink, G. *Polymers in Solution, Their Modelling and Structure*; Oxford Press: New York, 1989.

(6) (a) des Cloizeaux, J. *J. Phys. (Paris)* **1980**, *41*, 223. (b) Baumgärtner, A.; *Z. Phys. B* **1981**, *42*, 265. (c) Oono, Y.; Ohta, T. *Phys. Lett.* **1981**, *85A*, 480. (d) Mattice, W. L. *Macromolecules* **1981**, *14*, 1485, 1491. (e) Rubio, A. M.; Freire, J. J.; Bishop, M.; Clarke, J. H. R. *Macromolecules* **1993**, *26*, 4018. (f) Wittkop, M.; Kreitmeier, S.; Göritz, D. *Macromolecules* **1996**, *29*, 4754.

(7) Fujita, H. *Polymer Solutions*; Elsevier; Amsterdam, 1990.

(8) Perico, A.; Beggiato, M. *Macromolecules* **1990**, *23*, 797.

(9) Friedman, B.; O'Shaughnessy, B. *Macromolecules* **1993**, *26*, 4888.

(10) Whittal, R. M.; Li, L.; Lee, S.; Winnik, M. A. *Macromol. Rapid Commun.* **1996**, *17*, 59.

(11) (a) Cuniberti, C.; Perico, A. *Eur. Polym. J.* **1977**, *13*, 369. (b) Cheung, S.-T.; Winnik, M. A.; Redpath, A. E. C. *Makromol. Chem.* **1982**, *183*, 1815.

(12) O'Connor, D. V.; Phillips, D. *Time-Correlated Single Photon Counting*; Academic Press: London, 1984.

(13) Zuker, M.; Szabo, A. G.; Bramall, L.; Krajcarski, D. T.; Selinger, B. *Rev. Sci. Instrum.* **1985**, *56*, 14.

(14) Birks, J. B. *Photophysics of Aromatic Molecules*; Wiley & Sons: London, 1970.

(15) (a) Martinho, J. M. G.; Winnik, M. A.; *Macromolecules* **1986**, *19*, 2281. (b) Boileau, S. B.; Méchin, F.; Martinho, J. M. G.; Winnik, M. A. *Macromolecules* **1989**, *22*, 215. (c) Martinho, J. M. G.; Reis e Sousa, A. T.; Winnik, M. A. *Macromolecules* **1993**, *26*, 4484.

(16) Martinho, J. M. G.; Castanheira, E. M. S.; Reis e Sousa, A. T.; Saghbini, S.; André, J. C.; Winnik, M. A. *Macromolecules* **1995**, *28*, 1167.

(17) (a) Char, K.; Frank, C. W.; Gast, A. P.; Tang, W. T. *Macromolecules* **1987**, *20*, 1833. (b) Char, K.; Frank, C. W.; Gast, A. P. *Macromolecules* **1989**, *22*, 3177. (c) Duhamel, J.; Yekta, A., Hu, Y. Z.; Winnik, M. A. *Macromolecules* **1992**, *25*, 7024.

(18) Sinclair, A. M.; Winnik, M. A.; Beinert, G. *Macromolecules* **1985**, *18*, 1901.

(19) Bieze, T. W. N.; van der Maarel, J. R. C.; Eisenbach, C. D.; Leyte, J. C.; *Macromolecules* **1994**, *27*, 1355.

(20) Grulke, E. A. Solubility Parameter Values. In *Polymer Handbook,* 3rd ed.; Brandrup, J., Immergut, E. H., Eds; Wiley & Sons: New York, 1989.

MA961351U