J. Phys. Chem. B **2002,** *106,* 8499−8503

ARTICLES

Solution Properties of Self-Assembled Amphiphilic Copolymers Determined by Isomerization Spectroscopy

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Received: September 25, 2001; In Final Form: June 27, 2002

Amphiphilic copolymers of hydrophobic Disperse Red 1 acrylate (DR1A) and hydrophilic acrylic acid (AA) were dissolved in THF and a H_2O/THF cosolvent series. The thermal cis-to-trans isomerization behavior of the azo groups was measured by means of flash photolysis to investigate self-assembled structure in solution. The rate constants observed for the azo-containing polymers permit an estimation of how the thermal isomerization is influenced by both polymer and solution properties. In pure THF the relaxation was observed to be the usual monoexponential decay. In mixed aqueous−organic solvent solutions however, all poly(DR1A-*co*-AA) samples showed a highly unusual biexponential rate decay, with both a slow isomerization process due to inhibited mobility of the azo groups, and a faster isomerization process for unaggregated azo groups. In these systems, the relaxation of the cis-isomer was fit well with a biexponential function, which clearly demonstrates two distinct local chromophore environments. The spectroscopic results of this study suggest that weakly aggregated states of the amphiphilic polymers in an aqueous−organic solution can be readily studied through the simple technique of isomerization spectroscopy to estimate the fractions of aggregated groups involved and their local environment.

Introduction

Polymers containing azo aromatic chromophores have attracted considerable attention in the past decade due to their potential application as photoactive materials for optical recording and switching, biosensing, microstructuring and micropatterning of surfaces, and the development of target-activated theraputics.[1−6] Recently, much interest has been given to the reversible photoswitching of chemical or physical properties of molecules, macromolecules, and biopolymers containing azobenzene groups, which provides a basis for the development of future bio-optical devices.[7] Key to all of these novel applications is the readily induced and reversible isomerization between trans and cis geometric isomers of the azo aromatic group, when exposed to light near its maximum absorbance. These photochemical isomerizations of azobenzene and its derivatives, as well as the thermal back-isomerization occurring under no irradiation, are well-studied phenomena.[8,9] Illustrated in Figure 1, the trans to cis conversion is photochemically driven, whereas the reconversion to the energetically favored trans isomer can be achieved either photochemically or thermally. Of importance to this work, the rates of thermal isomerization in solution are first order, highly reproducible, and have been shown to be very sensitive to the local environment surrounding the azobenzene molecule, such as the polarity and pH of the solvent, as well as ion and polymer concentration.[8,10,11] These high sensitivities have thus facilitated the use of azobenzene molecules as excellent probes of the local polymer environment. For example, these molecules have been used to determine sequence distribution in statistical copolymers,[11] free volume distributions in thin films,[12] and structural formation of micelles by block copolymers.[13]

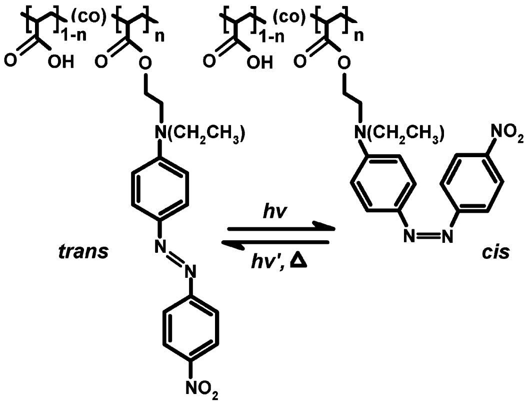

Figure 1. Chemical structure and geometric isomers of poly(DR1A-*co*-AA) where $n = 0.08, 0.14, 0.26, 0.35, 0.61, 0.71, 0.96$, and 1.0.

For polymers in solution, or above their glass transition temperature, it was found that the thermal cis-to-trans isomerization of azobenzenes can always be characterized by first-order kinetics.[10−12,14] The only exceptions to first order kinetics thus far reported in the literature have been for azobenzenes in the solid (glassy) state, where a portion of the azobenzenes isomerize anomalously quickly, and this anomaly is attributed to a nonuniform distribution of free volume in the glassy state. For these anomalous solid-state systems the decay curves can

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10.1021/jp013619b CCC: $22.00

be fit with either a biexponential,[10] a triexponential,[11] or a continuous distribution of relaxation rates reflecting the irregularity of the local environment using a Williams-Watts function.[15] All previous observations of cis-to-trans thermal relaxations in solution however have been strictly through first order (monoexponential) decay, with one exception: Recently, Gille et al.[13] have reported biexponential rates observed for azobenzenes in monitoring the structural formation of block copolymer micelles in aqueous solutions. In their study, biexponential rates reflected that isomerization had occurred in different local environments in the micelle, and provided useful information of local microphase separation. To the best of our knowledge however, only monoexponential rates have been reported to date for dissolved azo small molecules, homopolymers, and statistical copolymers, where only one local environment would be expected. This report describes a second class of azo-containing polymers that exhibit unusual nonmonoexponential thermal relaxation in solution.

One of the most recently prepared classes of azobenzene host polymers (and the one of primary interest to this work) is water-soluble polyelectrolytes, where the electrooptic azo chromophores can be self-assembled onto surfaces from aqueous solution to act as simple mimics of light-harvesting and light-responsive biopolymers.[7] The inherent hydrophilic−hydrophobic properties of aliphatic or aromatic-modified polyelectrolytes is also an appealing route to the study of hydrophobic self-association (folding) of complex biopolymers in water.[16,17] As part of an ongoing effort to allow these chromophore-containing polymers to be self-assembled onto surfaces from aqueous solution, we have recently prepared statistical copolymers of azo aromatic monomers with ionic units, specifically Disperse Red 1 acrylate (DR1A) and acrylic acid (AA), whose structure is detailed in Figure 1. Preliminary characterization of the resulting copolymers in aqueous solution revealed anomalously high viscosities, which indirectly suggests a self-assembled association between polymer chains in water (solution aggregation). A larger-than-expected molecular size was also implied by the results of dynamic light scattering experiments, which is the subject of another paper in progress.[18] This paper describes our detailed spectroscopic studies of solutions of these self-associative copolymers, by isomerizing the azobenzene groups to confirm solution aggregation through the unusual non-monoexponential relaxation of the azo chromophores. Furthermore, the interesting phase behavior displayed by these polymers can be probed quantitatively through the simple technique of isomerization spectroscopy, as a function of solvent quality and hydrophilic−hydrophobic group ratio.

Experimental Section

Spectroscopic grade tetrahydrofuran (THF) was purchased from Fisher Scientific and was distilled in order to remove all traces of water. Distilled water was filtered with a Millipore Milli-Q system to 18 MΩ resistivity, and mixed solvent systems of H_2O/THF in various ratios were prepared by volume. A series of copolymers based on monomers Disperse Red 1 acrylate (DR1A) and acrylic acid (AA), having the structure shown in Figure 1, was obtained by free radical polymerization. The synthesis and characterization of these copolymers have been reported elsewhere and a summary of the relevant properties of this copolymer series is listed in Table 1.[18] The copolymers were dried in a vacuum oven at 65 °C for 12 h prior to use. The entire copolymer series was soluble up to the H_2O/THF v/v ratio of 1/4 used, and the pH of the filtered aqueous solutions was adjusted (from pH = 3 to pH = 11) with HCl and NaOH where required, both obtained from Fisher Scientific. The aqueous−organic copolymer solutions were adjusted to the appropriate ion concentrations with NaCl where required, which was also obtained from Fisher Scientific. All experiments were performed at pH = 5.5 (no added HCl, NaOH), and without added NaCl, unless otherwise stated.

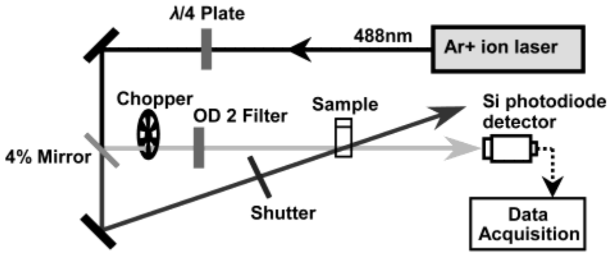

Figure 2. A schematic of the isomerization spectroscopy experimental setup used to determine the thermal cis-to-trans relaxation rate behavior of the copolymers in solution.

TABLE 1: Copolymer Composition and Polymer Properties for All Samples Studied

mol % DR1A	λ_{max} (nm)	MW (g/mol)	T_g (°C)
8	467	3000	92
14	466	2800	94
26	467	3200	93
35	467	3700	92
61	469	2500	-
71	470	2700	-
96	470	4200	-
100	468	3700	93

The experimental setup used for the isomerization measurements is illustrated in Figure 2 and is based on previously reported methods.[11,13] Approximately 30 μl copolymer samples (dye concentration 9×10^{-4} M) dissolved in various H_2O/THF mixtures were added to a quartz cuvette (transparent over the range of 375−600 nm studied) with an internal path length of 3 mm. The cuvette was capped to minimize evaporation of the solvent. The irradiation used was a circularly polarized beam of the 488 nm line of an argon laser at 425 mW/cm^2, for 1 s.

The first-order rate constants, k_a, for a monoexponential relaxation were determined from the time-dependent absorbance A_t and the absorbance A_0 at $t = 0$ (immediately after pumping), and A_∞ for infinite time (complete relaxation back to trans), by the usual data analysis techniques,[10−13] using the equation

$$(A_\infty - A_t) = (A_\infty - A_0)\exp(-k_a t) \qquad (1)$$

In cases where the data showed deviations from first-order kinetics, the experimental A_t relaxation curves were fit to the biexponential equation with 4 fitting parameters,[10]

$$(A_\infty - A_t) = A\exp(-k_a t) + B\exp(-k_b t) \qquad (2)$$

Similar to first-order kinetics, A_∞ and A_t are the absorbance at infinite time and at time t after the pump beam is switched off. A and B are coefficients and the normalization of these values are the relative weights associated with the processes having the rate constants k_a and k_b, respectively. A residual plot of the difference between and a least-squares fit demonstrated appropriateness of biexponential fitting. At least four pump/probe runs were repeated for each solution and the error analysis is based on the standard deviation of these runs to estimate the experimental uncertainty of the determined fitting parameters.

Results and Discussion

The acrylic acid portion of the copolymer has good solubility in both water and THF, but the aromatic DR1A group is

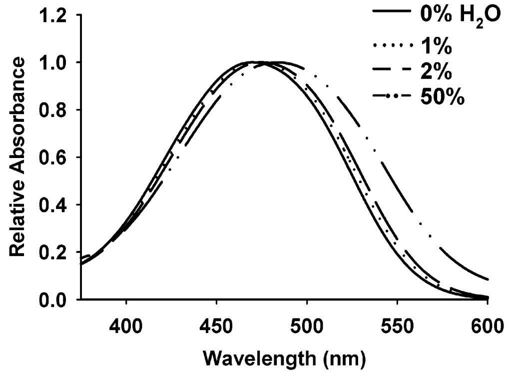

Figure 3. Absorbance spectra of poly(DR1A-*co*-AA) with 26% DR1A structural units observed with an increase of solvent polarity, by adding H_2O to THF.

insoluble in water and exhibits good solubility only in organic solvents such as THF and ether. THF solutions of poly(DR1A-*co*-AA) can be characterized by an absorption spectrum with an intense $\pi \rightarrow \pi^*$ transition centered at 468 ± 2 nm. Absorption spectra for the copolymer containing 26 mol % DR1A monomer at various solvent ratios of H_2O/THF are shown in Figure 3. These spectra show a shift of λ_{max} to longer wavelengths from 470 to 482 nm with an increase in v/v ratio of H_2O/THF from 0% to 50% H_2O, respectively. The red shift can be rationalized by a simple solvatochromic effect, which gives rise to such a shift in the absorption spectrum with gradual introduction of the more polar solvent. As suggested by Rau,[8] DR1 can be classified as pseudo-stilbene type azobenzene molecule based on spectroscopic and photochemical characteristics. These molecules contain electron-donating and electron-accepting ("push−pull") groups that effectively increase the energy of the N=N π-bonding orbital and lower the energy of the π^*-antibonding orbital, thus lowering the overall energy of the $\pi \rightarrow \pi^*$ transition. Polar solvents would be expected to stabilize the zwitterionic form of the molecule and contribute to the observed red shift.

Several λ_{max} absorption studies using UV−vis spectroscopy have demonstrated the tendency of some azo aromatic chromophores to form aggregates in the presence of water.[19,20] In these studies, aggregate formation of azobenzene phospholipids and small azo molecules in aqueous dispersions was attributed to dipole−dipole interactions of the azo compound and shown by either a red or a blue shift of the absorption maximum with increasing concentration of the chromophore. A marginal shift in the absorption maximum for the copolymers described here was observed, although it cannot be concluded that it arises from contributions other than solvatochromic effects. However, in addition to a red shift, a significant band broadening for the copolymers with increasing solvent polarity was observed.

Duhamel et al.[21] have employed fluorescence probe spectroscopy to investigate the association between polymer systems composed of a water-soluble backbone onto which hydrophobic chromophores have been attached. These measurements were carried out in organic and aqueous solutions of poly(ethylene glycol)s made of a central chain spanning two pyrene labels with extended chains of various lengths. To minimize their contact with water, the hydrophobic pyrene segments of the polymer appear to associate with each other. These molecular interactions result in fluorescence decays which could not be explained within the theoretical framework of the classic Birks' scheme. Also using similar amphiphilic polymers to the work reported here, McCormick et al.[22] have examined the effects of microblock hydrophobicity on interpolymer association behavior on a series of terpolymers composed of acrylic acid, methacrylamide, and a hydrophobic monomer. Their study demonstrated the effects of environmental stimuli on the associating behavior of the copolymers which incorporate comonomers with hydrophilicity and pH responsiveness. In our studies, similar molecular interactions appear to induce the formation of self-assembled aggregates which distort the UV−Vis spectra and complicate the thermal cis-to-trans relaxation process. In this study, we are suggesting a simple and viable method to verify aggregate formation through measurements of isomerization rates for azo-containing polymers in organic and aqueous−organic mixtures, and to estimate relative populations of the chromophores involved.

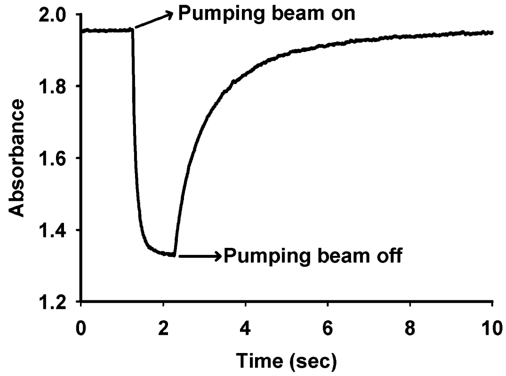

Figure 4. Typical time evolution for the absorbance of a poly(DR1A-*co*-AA) solution during flash photolysis.

We have investigated the thermal cis-to-trans isomerization in solution of the poly(DR1A-*co*-AA) copolymer series with variable azo content. In these experiments the solvent ratio of H_2O/THF was varied in order to analyze the effects of solvent quality on the kinetics of isomerization of DR1A in the copolymer series. Both the azo content and the H_2O/THF solvent ratio were found to display a strong influence on the isomerization, in contrast to the effects of varying either the salt or polymer concentration, or the solution pH (as discussed at the end of this section). Figure 4 illustrates a typical isomerization curve observed for the poly(DR1A-*co*-AA) copolymer with 26 mol % DR1A in THF. At the beginning of an experiment, the azo compound exists entirely in the energetically favored trans state. When the pumping beam is turned on (as indicated at $t =$ 1 s in Figure 4), isomerization is induced until the equilibrium photostationary state of both cis and trans is reached, and the net absorbance decreases due to the lower extinction coefficient of the cis form at 488 nm. Once the pumping beam is turned off, the azo compound thermally isomerizes back to the trans isomer. The reactions are completely reversible on subsequent flashes.

For the isomerization of DR1A contained in the copolymer series dissolved in pure THF (a good solvent for both groups), the relaxation can be fit using a monoexponential rate constant and is thus first order, confirming an equivalent local environment for each azo group. A typical linear first-order rate plot for the copolymer poly(DR1A-*co*-AA) containing 26 mol % DR1A structural units in pure THF is presented in Figure 5. Because copolymers in this series differ only in mol % of DR1A incorporated (ie: they have similar λ_{max}, MW, and T_g), a comparison between rate constants across cosolvent composition can reasonably be made, and should reflect only the influence of the solvent environment on the isomerization rates (and hence possible aggregation) of the copolymers. In pure THF, the first order rate constant was observed to be fairly constant for all copolymers in the series averaging $1.4\ s^{-1}$.

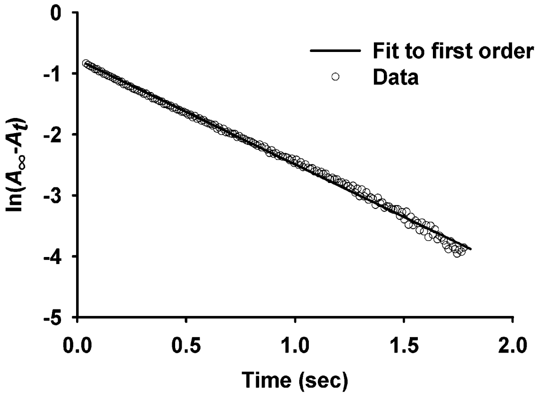

Figure 5. Monoexponential analysis of thermal isomerization spectra of poly(DR1A-*co*-AA) containing 26 mol % DR1A units in THF, to demonstrate a first-order relaxation.

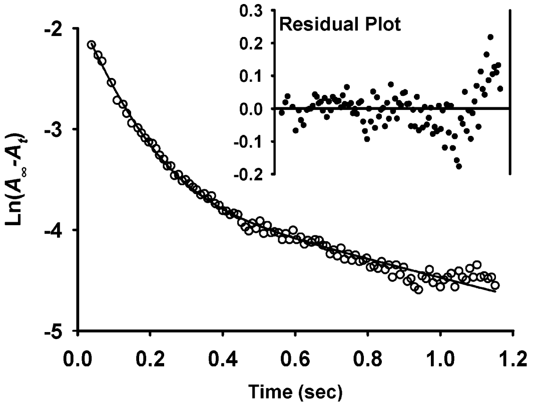

Figure 6. The decay of cis isomer fit to biexponential kinetics of poly(DR1A-*co*-AA) containing 26 mol % DR1A structural units in a 5% H_2O/THF solution. The inset is the residual plot between our least-squares four-parameter fit and the data.

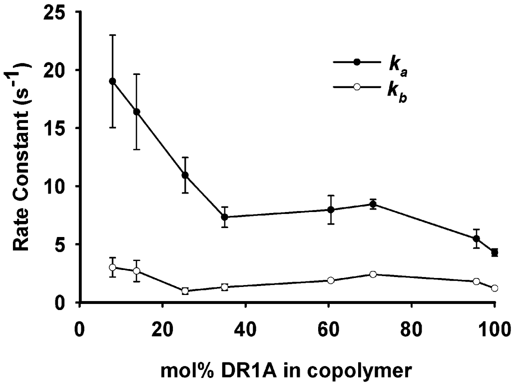

Figure 7. Biexponential rate constants obtained versus mol % DR1A structural units for copolymers poly(DR1A-*co*-AA) in a 5% H_2O/THF solution.

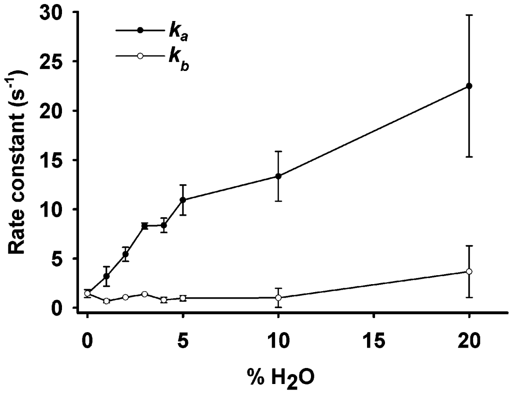

Figure 8. Rate constant versus % H_2O in a H_2O/THF solution of copolymers poly(DR1A-*co*-AA) containing 26 mol % DR1A structural units.

TABLE 2: Rate Constants and Amplitude Results from cis Relaxation Analysis of Poly(DR1A-*co*-AA) in a 5% Aqueous−Organic Solution

mol % DR1A	k_a (s^{-1})	A	k_b (s^{-1})	B
8	19.0 ± 4.0	0.61 ± 0.19	3.0 ± 0.8	0.34 ± 0.31
14	16.4 ± 3.2	0.66 ± 0.02	2.7 ± 0.9	0.34 ± 0.04
26	10.9 ± 1.5	0.83 ± 0.09	1.0 ± 0.3	0.17 ± 0.08
35	7.3 ± 0.9	0.73 ± 0.02	1.3 ± 0.3	0.27 ± 0.03
61	8.0 ± 1.2	0.30 ± 0.02	1.9 ± 0.1	0.70 ± 0.01
71	8.4 ± 0.6	0.63 ± 0.01	1.5 ± 0.2	0.37 ± 0.03
96	5.5 ± 0.8	0.32 ± 0.04	1.8 ± 0.1	0.68 ± 0.03
100	4.3 ± 0.3	0.74 ± 0.02	1.2 ± 0.1	0.26 ± 0.03

The addition of even a small quantity of H_2O to the THF however, yielded distinctly nonmonoexponential rate behavior, which is highly unusual for azobenzene chromophores in solution. We observed that for copolymers of poly(DR1A-*co*-AA) in H_2O/THF solutions, the decay of the cis isomer could not be fit with the monoexponential function (1), but instead by the biexponential function (2), as illustrated in Figure 6. Also illustrated in Figure 6 is a residual plot (inset), which confirms the appropriateness and sufficiency of a biexponential model. A biexponential model suggests two distinct local environments involved in the thermal cis-to-trans isomerization, with one portion of DR1A converting much more quickly and the remainder isomerizing more slowly.[10−12] This rate of the slow process (k_b) is fairly constant over the whole range of compositions, while the rate of the fast component (k_a) decreases significantly with the mol % DR1A content present in the copolymer, approaching the rate of isomerization observed for the pDR1A homopolymer. Figure 7 and Table 2 show the results of thermal isomerization of DR1A in the copolymer series dissolved in a 5% H_2O/THF solution. Also listed in Table 2 are the amplitudes *A* and *B* associated with each process of fast and slow isomerization, respectively. The comparison of the weight contributions, *A* and *B*, to each rate is not straightforward. While these results exhibit more scatter than the determined rate constants, it is evident that there are significant contributing populations to each rate for each sample. Furthermore, this two-state implication would also serve to rationalize the absorbance band broadening (Figure 3) observed for the copolymer series, where the results observed with increasing water content are in fact a summation of the absorbances of chromophores in each of the two states, aggregated and unaggregated.

Since water is a poor solvent for DR1A, the deviations from first-order kinetics in H_2O/THF mixtures are likely a consequence of an aggregate formation through hydrophobic molecular interactions, as the polymers lower their energy by folding the hydrophobic groups away from the polar solvent. These hydrophobic associations give rise to a reduced mobility of the side chain and a significantly different local solvent environment, which accounts for the slower isomerization. The local solvent environment of the fast component is more polar compared to that experienced by the aggregated azo groups, and accordingly a more rapid cis-to-trans thermal isomerization is observed. Furthermore, the fast rate component increases strongly with the H_2O content in the cosolvent consistent with solvatochromic effects indicated by increasing wavelengths of absorption maxima of the trans isomers. Figure 8 shows the two rate constants of thermal isomerization observed with

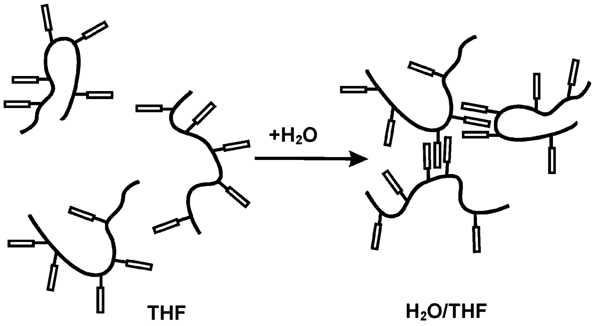

Figure 9. Suggested self-assembled aggregation behavior of amphiphilic copolymers studied in aqueous−organic solution, determined from isomerization rate analysis.

increasing solvent polarity for poly(DR1A-*co*-AA) containing 26 mol % DR1A. Since only the fast rate component markedly increased with the solvent polarity, this indicates a substantial change in solvent conditions for only one of the two environments of the chromophores, which we interpret with the model illustrated in Figure 9. Likewise, all the copolymers investigated demonstrated similar behavior to that illustrated in Figure 8. These isomerization results clearly suggest two distinct environments experienced by the chromophore in all copolymer compositions, only one of which appears to significantly experience the cosolvent environment enough to display solvatochromicity. Furthermore, this “ solution” rate component of the biexponential is the faster of the two with the slower rate constant being more similar to that usually observed in the solid state.[11] This behavior can be rationalized by the interchain aggregation depicted, where the azo aromatic groups have folded out of solution to behave more like solid state photoswitches, and are thus largely indifferent to the solvent environment. Isomerization rate constants (k_b) of this ‘solid’ component would then be expected to be largely independent of solvent polarity, as observed.

As discussed earlier, we also investigated the dependence of the rate constants (k_a, k_b) and weights (A, B) of the biexponential isomerization on ionic strength, pH, and polymer concentration. No significant change in the rate behavior was observed when the ionic strength was increased (as tested up to 1 M NaCl), nor was there any significant change in these four fitting constants observed over a wide pH range, after adding HCl or NaOH (as tested from pH = 3 to pH = 11). Last, the complex rate behavior was similarly observed to be insensitive to increasing or decreasing the polymer concentration by 1 order of magnitude. The isomerization behavior thus appears to depend only on the fraction of hydrophobic units in the polymer and on the H_2O/THF cosolvent ratio.

Conclusion

Rate constants of the thermal cis-to-trans isomerization of DR1A in H_2O/THF solutions of random copolymers of poly(DR1A-*co*-AA) determined by means of flash photolysis show an unusual biexponential thermal relaxation of the cis isomer. This strongly suggests different local chromophore environments due to a self-assembled aggregation of the hydrophobic groups, an effect difficult to measure directly by other methods. The results of this study demonstrate the utility of isomerization spectroscopy in determining the microstructure of amphiphilic polymer solutions, which can assist in rationalizing macroscopic properties of copolymer organization in solution. Future work with these polymer systems will focus on using light scattering as a parallel technique to investigate the aggregation behavior, and to explore a reversible control over the aggregation using laser light.

Acknowledgment. The authors are grateful to Dr. N. Ahmad for preparing the copolymers used, and to K. Yager for electronics assistance. Funding for this project was provided by McGill University and NSERC Canada.

References and Notes

(1) Delaire, J. A.; Nakatani, K. *Chem. Rev.* **2000**, *100*, 1817.
(2) Natansohn, A.; Rochon, P. *Adv. Mater.* **1999**, *11*, 1387.
(3) Willner, I.; Rubin, S. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 367.
(4) Back, H.; Andrele, K.; Fuhramann, T.; Wendorf, J. *J. Phys. Chem.* **1996**, *100*, 4135.
(5) Carter, F. L.; Schultz, A.; Duckworth, D. In *Molecular Electronic Devices II*; Carter, F. L., Ed.; Marcel Dekker: New York, 1987; p 183.
(6) Osvaldo, P.; Adriano, F.; Angelini, N.; Lenci, F. *Acc. Chem. Res.* **2001**, *34*, 9.
(7) Willner, I. *Acc. Chem. Res.* **1997**, *30*, 347.
(8) Rau, H. in *Photochemistry and Photophysics*; Rabek, J. F., Ed.; CRC Press: Boca Raton, FL, 1990; Vol. 2, p 119.
(9) Kobayashi, T.; Degenkolb, E.; Rentzepis, P. M. *J. Phys. Chem.* **1979**, *83*, 2431.
(10) Paik, C. S.; Morawetz, H. *Macromolecules* **1972**, *5*, 171.
(11) Barrett, C. J.; Natansohn, A.; Rochon, P. *Macromolecules* **1994**, *27*, 4781.
(12) Eisenbach, C. D. *Makromol. Chem.* **1978**, *179*, 2489.
(13) Gille, K.; Knoll, H.; Rittig, F.; Fleischer, G.; Karger, J. *Langmuir* **1999**, *15*, 1059.
(14) Kumar, G.; Neckers, D. *Chem. Rev.* **1989**, *89*, 1915.
(15) Wiesner, U.; Antonietti, M.; Boeffel, C.; Spiess, H. W. *Makromol. Chem.* **1990**, *191*, 2133.
(16) Nelson, J. C.; Saven, J. G.; Moore, J. S.; Wolynes, P. G. *Science* **1997**, *277*, 1793.
(17) Esquenet, C.; Buhler, E. *Macromolecules* **2001**, *34*, 5287.
(18) Ahmad, N.; Norman, L. L.; Barrett, C. J. To be submitted for publication.
(19) Song, X.; Perlstein, J.; Whitten, D. G. *J. Am. Chem. Soc.* **1997**, *119*, 9144.
(20) Buwalda, R. T.; Jonker, J. M.; Engberts, J. B. F. N. *Langmuir* **1999**, *15*, 1083.
(21) Lee, S.; Duhamel, J. *Macromolecules* **1998**, *31*, 9193.
(22) Smith, G. L.; McCormick, C. L. *Macromolecules* **2001**, *34* 5579.