Comparison of the Self Diffusion Coefficient of Polystyrene in Solution Estimated by Forced Rayleigh Scattering and Fluorescence Recovery after Photobleaching

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ABSTRACT: The self diffusion coefficient D_s of two labeled polystyrene (PS) samples in dibutyl phthalate (DBP) has been measured in the range of polymer concentration from 0.1 to 50 wt% using forced Rayleigh scattering (FRS) and fluorescence recovery after photobleaching (FRAP). 2-Nitro-4-carboxy-4'-dimethylaminostilbene and 4-glycyl-7-nitrobenzofurazan have been used as photochromic dyes for FRS and FRAP measurements, respectively. Good agreement has been found between two sets of D_s data in the range of D_s from 10^{-8} to 10^{-12} cm² s⁻¹ within experimental error. In the dilute region of $c \leq 1$ wt%, mutual diffusion coefficient D of unlabeled but otherwise the same PSs in DBP has been measured by dynamic light scattering. The D_s asymptotically approached D with decreasing polymer concentration.

KEY WORDS Self Diffusion Coefficient / Forced Rayleigh Scattering /
Fluorescence Recovery after Photobleaching / Polystyrene Solution /
Photobleachable Dye /

The self diffusion coefficient D_s is a most useful quantity for the study of slow chain dynamics in condensed polymeric systems.¹ Among various techniques proposed for D_s measurements,²⁻¹³ both forced Rayleigh scattering (FRS) and fluorescence recovery after photobleaching (FRAP) are methods which make use of photochromic dyes for labeling of polymers. The former method has been developed in the field of physical chemistry of synthetic polymers, while the latter has been invented in the field of biophysics.¹⁴⁻¹⁷ Both methods have merits and demerits. The FRS method usually provides an accurate estimate of D_s because of high diffracted intensity I_s detected at the Bragg angle and of a definite value of I_s at time $t \rightarrow \infty$, i.e., the base line

value. However, the complicated photochemical process of a dye sometimes gives rise to an anomalous I_s vs. t curve, which hinders unambiguous data analysis.^{18,21} The FRAP method is free from the dye problem, but an indefinite value of I_s at $t \rightarrow \infty$ makes it difficult to estimate D_s accurately.²² Furthermore it is troublesome in a strict sense that there is no standard material with a known D_s value with which accuracy of the either measurement can be examined. On the other hand, it is reasonable to consider that the two techniques should give an identical D_s value in principle, when applied to the same polymer-solvent system. Then good agreement for the absolute magnitude of D_s may be obtained. In spite of the importance of the two techniques, such com-

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parison has not been attempted yet.

Recently we constructed an apparatus with which both FRS and FRAP measurements can be performed with a slightly different experimental set up.²³ In this paper, we compare two sets of D_s data of narrow-distribution polystyrene in dibutyl phthalate over a wide range of concentration from 1 to 50 wt% obtained with the FRS and the FRAP methods. Dynamic light scattering measurements have been also made on unlabeled but otherwise the same polymer solutions in the dilute region of $c \leq 1$ wt%, and the mutual diffusion coefficient D data obtained have been compared with corresponding D_s data from FRS measurements. This experiment was aimed to confirm that, in the very dilute region, D_s should asymptotically approach D with decreasing polymer concentration.

EXPERIMENTAL

Materials

Two narrow distribution polystyrenes ($\bar{M}_w = 43900$; sample code F4 and $\bar{M}_w = 355000$; code F40, Toyo Soda) were used as samples in this study. The polymers were randomly chloromethylated in methylene dichloride with chloromethylmethylether and with zinc chloride as catalyst. The chloromethylated polymers were subsequently labeled with a photobleachable dye of 2-nitro-4-carboxy-4'-dimethylaminostilbene for FRS measurements and with a photobleachable fluorescent dye of 4-glycyl-7-nitrobenzofurazan for FRAP measurements, respectively. The detailed labeling procedure is described elsewhere.²⁴ GPC combined with low-angle light scattering measurements has shown that there is no change in either molecular weight or molecular weight distribution of the polymers during the labeling procedure.

Distilled dibutyl phthalate (DBP) was used as solvent for preparation of solutions to be measured. Weighed amounts of labeled PS and DBP were dissolved in sufficient amount of

CH_2Cl_2 , stirred for a couple of days to complete homogeneous mixing, and then filtered through a $0.45 \mu\text{m}$ pore size filter. Methylene-dichloride was subsequently evaporated very slowly from the solution with occasional stirring to obtain a test polymer solution. The polymer concentration c was determined by weighing. The range of c investigated was from 0.1 to 50 wt%.

Methods

The self diffusion coefficient D_s of labeled PS in solutions was measured using a computer-controlled apparatus recently constructed at our laboratory.²³ For the FRS experiment, monochromatic light of an argon-ion laser ($\lambda = 488 \text{ nm}$) and a He-Ne laser ($\lambda = 633 \text{ nm}$) were used as the writing and the reading beams, respectively. The optical configuration of the apparatus is similar to those of FRS instruments reported in the literature.^{2,4,8} Acquisition and analysis of diffracted light intensity data were conducted with a home-made data processor. The sample was sandwiched between two 1.5 cm diameter fused-silica optical windows, or put into a 1 cm square-shaped cell with width of 1–3 mm, which was fixed in the cell holder. The holder was thermoregulated within $\pm 0.02^\circ\text{C}$ by circulating a mixture of ethyleneglycol and water through an outer bath and an external thermostating bath.

For the FRAP experiment, a sample was deposited as a thin film on pattern mask (pitch 4–64 μm , 1 mm-square) made by electron-ray lithography on a 1.5 cm diameter fused-silica optical window. A laser line ($\lambda = 488 \text{ nm}$) with an output of 300 mW was used for photobleaching of the labeled PS at time $t = 0$ and also for fluorescence recovery measurements at a later time by decreasing the intensity by a factor of 10^5 . Since continuous illumination was found to photobleach the fluorescent dye even with such a low intensity, the samples were illuminated only for short intervals in detecting fluorescence signals from the samples

by use of an clock-controlled rotating shutter. All experiments were performed at $25 \pm 0.02^\circ\text{C}$.

Dynamic light scattering measurements on unlabeled but otherwise the same PS-DBP solutions were made at $25 \pm 0.01^\circ\text{C}$ with the instrument reported earlier.²⁵

RESULTS AND DISCUSSION

Data Analysis

Figure 1 shows the time profiles of diffracted light intensity I_d and fluorescence intensity I_f as an example of raw data obtained for a 5 wt% solution of PS with $\bar{M}_w = 355000$ in DBP with FRS and FRAP, respectively. The

solid curve in Figure 1(a) is a curve fitted with eq 1, the theoretical model function with a single decay rate Γ_d for FRS.

$$I_d = \{A \exp(-\Gamma_d t) + B\}^2 + C^2 \quad (1)$$

Here A is amplitude, B and C are contributions from coherent and incoherent background optical field, respectively. It is evident from the figure that eq 1 expresses the time dependence of I_d quite well. Indeed, normal residuals from the fit are very small and random as shown in the figure. Coherent scattered intensity from the polymer solutions was found to be negligibly small in comparison with the diffracted light intensity at $t=0$, i.e., a value of A , so that fitting by eq 1 with $B=0$

Figure 1. (a) Decay of diffracted light intensity I_d due to self-diffusion of labeled PS with $\bar{M}_w = 355000$ in DBP. Polymer concentration is 5 wt% and the grating spacing d is $8 \mu\text{m}$. The solid curve is a least-square fit with eq 1. (b) Recovery of fluorescence intensity I_f due to self diffusion of labeled PS with $\bar{M}_w = 355000$ in DBP. Polymer concentration is 5 wt% and the pitch of the striped pattern d is $8 \mu\text{m}$. The solid curve is a least-square fit with eq 2. Normal residuals obtained from each fit are shown in the bottom of the each figure.

gave only a few percent different value to Γ_d . Figure 1(b) shows a recovery profile of fluorescence intensity I_f . Data are more scattered in comparison with the FRS data shown in Figure 1(a) because of low irradiation intensity of the laser beam used for detection of I_f . The data were fitted with eq 2 in which higher frequency terms were neglected.

$$I_f = A' - B' \exp(-\Gamma_f t) \quad (2)$$

Here A' is the value of I_f in equilibrium and B' is the amount of I_f which is recovered due to self diffusion of labeled polymers. Normal residuals from the fit are shown in the bottom of the figure. Fitting by the solid curve in the figure gave Γ_f to an accuracy of about 30%. Inclusion of higher frequency terms did not improve the fitting. No PS-DBP solutions tested by either FRS or FRAP methods gave an anomalous I vs. t curve with a minimum and a maximum, and the curves could be successfully fitted with the corresponding eq 1 or 2.

Figure 2 shows that Γ_d and Γ_f thus obtained satisfy the proportionality of the decay rate to the square of the wave vector q ($=2\pi/d$, d , the pitch of the interference (FRS) or the striped pattern (FRAP)). It may be noted that q^2 was widely changed by a factor of 64 for FRAP measurements. D_s was estimated from eq 3 to an accuracy of 10 and 30% for FRS and FRAP, respectively.

$$D_s = \Gamma_i / q^2 \quad (i = d, f) \quad (3)$$

Values of D_s are given in Table I.

Normalized time correlation function data of scattered intensity from unlabeled PS solutions measured by dynamic light scattering (DLS) were analyzed using a single exponential type of decay function. The mutual diffusion coefficient D was estimated as the slope of the decay rate Γ vs. q^2 plot within an error of a few percent. Values of D are given in Table I.

D_s obtained by FRS and FRAP and also D by DLS are plotted against polymer con-

Figure 2. Plots of relaxation rate Γ_i ($i = d, f$) against the square of the wave vector q . D_s is estimated as a slope of a straight line drawn in the figure for each c and M . (a) Γ_d from FRS. Sample and concentration are: 1, F40, 10 wt%; 2, F4, 30%; 3, F40, 15%; 4, F4, 40%. (b) Γ_f from FRAP. Sample and concentration are: 1, F40, 1 wt%; 2, F40, 2%; 3, F40, 5%; 4, F4, 20%; 5, F40, 10%; and 6, F4, 30%.

centration c in Figure 3 for comparison. In the dilute regime where c is less than 1 wt%, D of the both samples looks almost independent of c in the logarithmic scale, and a value at infinite dilution, *i.e.*, the translational diffusion coefficient D_0 , could be easily estimated as $2.83 \pm 0.04 \times 10^{-8}$ and $9.30 \pm 0.10 \times 10^{-9}$

Table I. D_s values of PS with $\bar{M}_w = 43900$ (F4) and 355000 (F40) in DBP estimated by FRS and FRAP, and D values of unlabeled but otherwise the same PSs in DBP by DLS at 25°C

$\bar{M}_w \times 10^{-4}$	$c/\text{wt}\%$ $D_s/\text{cm}^2 \text{s}^{-1}$		$c/\text{wt}\%$ $D_s/\text{cm}^2 \text{s}^{-1}$	
	by FRS		by FRAP	
4.39	0.100	2.5×10^{-8}		
	0.300	2.5×10^{-8}		
	0.995	2.3×10^{-8}	0.996	2.2×10^{-8}
	10.1	6.5×10^{-9}	9.67	5.3×10^{-9}
	19.9	9.6×10^{-10}	19.8	7.7×10^{-10}
	30.4	1.5×10^{-10}	29.8	1.5×10^{-10}
	39.6	2.6×10^{-11}	38.5	2.5×10^{-11}
	40.7	1.7×10^{-11}	40.8	1.9×10^{-11}
	49.9	1.7×10^{-12}	49.7	1.4×10^{-12}
35.5	0.098	1.0×10^{-8}		
	0.300	8.5×10^{-9}		
	1.06	8.3×10^{-9}	1.03	7.1×10^{-9}
	2.05	4.0×10^{-9}	2.01	3.0×10^{-9}
	5.38	1.1×10^{-9}	4.74	9.6×10^{-10}
	10.5	1.9×10^{-10}	10.1	2.1×10^{-10}
	20.0	1.3×10^{-11}	20.4	1.3×10^{-11}
$\bar{M}_w \times 10^{-4}$	$c/\text{wt}\%$ $D_s/\text{cm}^2 \text{s}^{-1}$			
	by DLS			
4.39	0.100	2.79×10^{-8}		
	0.300	2.91×10^{-8}		
	1.00	2.77×10^{-8}		
35.5	0.100	9.41×10^{-9}		
	0.296	9.87×10^{-9}		
	1.00	1.11×10^{-8}		

$\text{cm}^2 \text{s}^{-1}$ to PS molecule with $\bar{M}_w = 43900$ (F4) and 355000 (F40), respectively. D_s is almost equal to or slightly less than D in this concentration range as is expected to a polymer-good solvent system, and tends to approach asymptotically D_0 with decreasing concentration. Since DLS is a well-established technique for an accurate estimate of D_0 , good agreement between D_s and D_0 at infinite dilution indicates that D_s can be estimated to high accuracy with our instrument. The hydrodynamic radius R_H of the polymer in DBP can be calculated from values of D_0 and solvent viscosity by applying the Einstein-Stokes re-

Figure 3. Comparison of D_s estimated by FRS and FRAP, and D by DLS. Symbols: (□), FRS data; (○), FRAP data; and (●), DLS data. The right pip denotes F4 sample ($\bar{M}_w = 43900$) and the left F40 sample ($\bar{M}_w = 355000$).

lationship. It has been found that the dependence of R_H on molecular weight is closely represented by eq 4, which was determined for PS in good solvent in an earlier study.²⁶

$$R_H = 1.60 \times 10^{-9} \bar{M}_w^{0.55} \quad (4)$$

In the region of $c \geq 1$ wt%, two sets of D_s data for each PS sample are now compared. As a general trend, D_s values obtained by FRAP look a little smaller than corresponding values by FRS. Taking into account the experimental uncertainties involved in D_s values by FRAP, however, we may state that they are located on a single composite curve for each molecular weight. Thus we may conclude that correct values of D_s from 10^{-8} to $10^{-12} \text{cm}^2 \text{s}^{-1}$ can be obtained for the PS-DBP system with either FRS or FRAP.

The concentration dependence of D_s can not be described by a simple power law as is clear from Figure 3. Steep decrease in D_s of the low molecular weight PS at high concentration above 30 wt% is mainly due to increase in local

friction or decrease in free volume which essentially determines the mobility of a polymer segment in condensed system. On the other hand, the concentration dependence of D_s of the higher molecular weight PS should be interpreted as being due to the combined effects of local friction, hydrodynamic interaction and topological interaction.²⁷ Dependence of D_s on temperature, concentration and molecular weight in the concentrated regime will be discussed in detail in future papers.

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