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Hydrodynamic Analysis of Well-Defined Flexible Linear Macromolecules of Low Molar Mass

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ABSTRACT: Samples of poly(styrene−diphenylethylene) copolymers of narrow molar mass distribution were studied by molecular hydrodynamic methods. The values of velocity sedimentation coefficient, frictional ratio, translational diffusion coefficient, and intrinsic viscosity were measured in toluene. It is shown that the method of velocity sedimentation, coupled with the numerical solution of the Lamm equation (Sedfit program), can be considered as an absolute method of molar mass determination. The correlations between the hydrodynamic values and the molar mass were obtained. A new Multi-HYDFIT program was applied to obtain the conformational parameters of poly(styrene−diphenylethylene) copolymer chains unaffected by intrachain excluded volume effects.

1. Introduction

Hydrodynamic methods and values, such as velocity sedimentation (s_0) and translational diffusion (D_0) coefficient as well as intrinsic viscosity ($[\eta]$), have played an important role in traditional polymer science, biochemistry and colloid science.[1−3] With synthetic polymers, however, interest in both the methods and their results in recent years have been scarce, despite the availability of modern analytical ultracentrifugation using the Beckman XLI analytical ultracentrifuge.[4] Therefore, the focus of software development for the treatment of automatically captured sedimentation data has been on the study of globular protein systems which are characterized by high fractal dimension (∼3) (similar to macroscopic particles), whereas linear macromolecules are characterized by fractal dimensions between 2 and 1 ("soft matter"). In fact, molecular hydrodynamics of the latter types of macromolecules generally are more sophisticated.[5,6] The question whether it is possible to determine adequately the sedimentation coefficients and frictional ratios of flexible linear chains with existing software therefore is an open one.

Despite the shortcomings described, there is an apparent need for studying and interpreting, by the available apparatus and software, the solution properties of synthetic homopolymers and copolymers and of more complex synthetic systems, such as metallo-supramolecular architectures and micelles[7−10] to obtain reliable information on their molar mass and size. Our present aim is to improve and optimize the measurement and the data analysis protocols and, by this, to transform the technique into a versatile and reliable-to-use procedure, in order to fully benefit from its advantages for studying nonglobular molecular systems. Regarding data analysis, the current paper presents an application of the recently developed Multi-HYDFIT program,[11] intended for the determination of conformational characteristics by a joint analysis of various properties of multiple samples. Applications of this methodology have been described for rigid or weakly bending rodlike molecules,[11] and stiff but long polysaccharide molecules.[12,13] The application reported here is the first one for flexible but short polymers.

2. Materials and Methods

In this contribution we report on the hydrodynamic study of well-defined terpyridine end-functionalized poly(styrene-*alt*-diphenylethylene) copolymers with low polydispersity index values ($M_w/M_n < 1.2$), synthesized via anionic polymerization. In order to grant the alternating nature of the synthesized polymers and in addition to obtain an effective termination with the terpyridine ligand, an excess of 1,1-diphenylethylene was used. The synthesis and characterization of the copolymers by ^{1}H NMR and UV−vis spectroscopy, MALDI−TOF mass spectrometry, GPC, elemental analysis, and preliminary hydrodynamic analysis were described in a previous contribution.[14] In particular the values of velocity sedimentation coefficients, frictional ratio and intrinsic viscosity, have been obtained. The traditional analysis of hydrodynamic data is made and the estimations of the equilibrium rigidity of polymeric chains are obtained.

Sedimentation velocity experiments were performed in a Beckman XLI analytical ultracentrifuge (ProteomeLab XLI protein characterization system) at a rotor speed of 55 000 as well as 40 000 rpm and at 20 °C, using the interference optics and Al double-sector cells of an optical path of 12 mm. The sedimentation experiments had an average duration of 10 h. Three concentrations of each sample in toluene were studied ($c_{\max}/c_{\min} \geq 3.5$).

All concentrations were in the zone of high dilutions where the Debye parameter $c[\eta]$[15] characterizing the degree of dilution was in the range $0.01 \leq c[\eta] \leq 0.1$.[16] These conditions of dilution are allowing reliable extrapolation to zero concentration and estimation also of the concentration parameters. The continuous

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particle size distribution c(s) of Sedfit[17,18] was used for data analysis. The frictional ratio value (f/f_{sph}), which is the weight-average frictional ratio of all species, was optimized executing a fit command. Different regularization methods were compared: the maximum entropy method with a confidence level F of 0.68, and the Tikhonov−Philips second derivative method with a higher confidence level (corresponding to a F-ratio of 1.1).

Translational diffusion was studied by the classical method of forming a boundary between the solution and the solvent.[2] The diffusion boundary was formed in glass cell of a thickness of h = 30 mm along the beam path, at an average solution concentration $c \cong 1$ mg/mL. The optical system used for recording the solution-solvent boundary in diffusion analysis was a Lebedev's polarizing interferometer.[19] The average time for boundary observation was 20−30 min. The values of the diffusion coefficient D obtained were assumed to be the values extrapolated to zero concentration. The correctness of this procedure has been confirmed by numerous investigations on the concentration dependence of D.[2] Translational diffusion coefficients were calculated from the equation

$$\sigma^2 = {\sigma_0}^2 + 2Dt \tag{1}$$

where σ^2 is the dispersion of the diffusion boundary calculated from the maximum ordinate and the area under the diffusion curve according to ref 2, ${\sigma_0}^2$ is the zero dispersion characterizing the quality of boundary formation, and t is the diffusion time. The intrinsic diffusion coefficient $[D]$, which depends only on the macromolecule properties and the temperature T (in our case 298 K), is calculated as follows:

$$[D] = D_0\eta_0/T \tag{2}$$

Viscosity measurements were conducted using an Ostwald viscometer. The respective flow times, τ_0 and t, were measured at 25 °C for the solvent and polymer solutions, with relative viscosities $\eta_r = t/\tau_0$ in the range of 1.2 to 2.2. This interval of η_r corresponds to different concentrations for different samples. For instance, for sample 1 the interval of concentration was equal to $1.2 \le c \times 10^2$ g/cm$^3 \le 4.3$, and for sample 7 it was equal to $4.5 \le c \times 10^2$ g/cm$^3 \le 16$. It should be noted that all solutions are in the range of the dilute ones (see footnote above).

The extrapolation to zero concentration was made by using both the Huggins and Kraemer equations, and the average values were considered as the value of intrinsic viscosity.

Density measurements were carried out in the density meter DMA 02 (Anton Paar, Graz, Austria) according to the procedure of Kratky et al.[20]

3. Theoretical Background

Sedfit Treatment of Velocity Sedimentation Data. The program Sedfit developed by P. Schuck[17,18] numerically solves the Lamm equation, the basic differential equation describing the coupled sedimentation and diffusion process.[21] In the $c(s)$ method, this is done for a large number of species with different sedimentation and diffusion coefficients. The result is a continuous sedimentation coefficient distribution which represents the best combination of species for matching the entire collected set of experimental concentration profiles. The numerical analysis is conducted under appropriate statistical criteria of goodness-of-fit. Two different regularization methods can be used: the maximum entropy or the Tikhonov−Philips second derivative method. A choice has to be made concerning the confidence level (F-ratio), which determines the magnitude of the regularization. The partial specific volume ($\bar{v}$), the solvent density (ρ_0), and solvent dynamic viscosity (η_0) are additional parameters required to calculate the sedimentation coefficient distributions. When studying synthetic polymer systems, the value of the partial specific volume or the buoyancy factor has to be measured experimentally.

In order to achieve an one-parametric distribution, a scaling law between the diffusion and sedimentation coefficient is invoked in the $c(s)$ method. Usually, the choice of the scaling law is based on $s \sim M^{2/3}$ for roughly globular particles, with the frictional ratio (f/f_{sph}) as a scaling parameter (f, frictional coefficient of the solute macromolecule; f_{sph}, frictional coefficient of a rigid sphere with the same "anhydrous" volume (free of solvent) as the macromolecule). Fitting for (f/f_{sph}) in a nonlinear regression will provide an estimate of the weight-average frictional ratio of all macromolecules in solution. Finally, the differential distribution ($\mathrm{d}c(s)/\mathrm{d}s$) of the sample is obtained and marked as $c(s)$, scaled such that the area under the $c(s)$ curve will give the loading concentration of the macromolecules between the minimum and maximum s-value occurring (expressed in number of fringes, J, in the case of interference optics). J is also used to calculate the refractive index increment: $(\Delta n/\Delta c) = J\lambda/Kcl$,[22] where λ is the wavelength (675 nm), K the magnifying coefficient, and l the optical path. With $K = 1$ and $l =$ 12 mm we obtain: $\Delta n/\Delta c = 5.625 \times 10^{-5}$ J/c.

Essential Hydrodynamic Values and Their Interpretation. The different hydrodynamic measurements can be expressed as intrinsic values, $[\eta]$, $[s]$, k_s, $[D]$, and $[f]$, which are independent of such common solvent properties as dynamic viscosity and density ($[\eta]$, intrinsic viscosity; $[s]$, $[D]$, and $[f]$, intrinsic coefficients of velocity sedimentation, translational diffusion, and translational friction, correspondingly; k_s, concentration coefficient or Gralen coefficient).[1,2] Each of them is related with common macromolecular characteristics, such as molar mass M and end-to-end distance $\langle h^2\rangle$ in the case of linear polymers:

$$[\eta] = \Phi\langle h^2\rangle^{3/2}/M \tag{4}$$

$$[s] \equiv s_0\eta_0/(1-\rho_0\,\bar{v}) = M/N_A P\langle h^2\rangle^{1/2} \tag{5}$$

$$k_s = B\langle h^2\rangle^{3/2}/M \tag{6}$$

$$[D] \equiv D_0\eta_0/T = k/P\langle h^2\rangle^{1/2} \tag{7}$$

$$[f] \equiv f_0/\eta_0 = P\langle h^2\rangle^{1/2} \tag{8}$$

where N_A is the Avogadro number, Φ and P are the Flory hydrodynamic parameters, and B is another hydrodynamic parameter.[23] The values of Φ and P are functions of the relative contour length (L/A) and relative diameter (d/A), tabulated in the case of the theory describing the translational friction of the worm-like cylinder, where L is the contour length of the macromolecule, A is the length of the statistical segment (Kuhn segment length),[24] and d is the diameter of the chain. These parameters (Φ and P) are also dependent on the thermodynamic quality of the solvent due to long-chain interactions. In the present study, copolymer chains are rather short, with $L/A \le 15$, and the term of the long-chain interactions will not be considered.

Combination of 5 and 7 leads to the Svedberg equation [1−3,25] used for the molar mass determination from the hydrodynamic data:

$$M_{sD} = (RT/(1-\bar{v}\rho_0))(s_0/D_0) = (N_A/(1-\bar{v}\rho_0))s_0 f_0 \tag{9}$$

The translational friction coefficient f_0 of the macromolecules may be expressed in our case in the following way:

$$f_0 = (f/f_{\mathrm{sph}})_0 f_{\mathrm{sph}} = 6\pi\eta_0(3M\bar{v}/4\pi N_{\mathrm{A}})^{1/3}(f/f_{\mathrm{sph}})_0 \qquad (10)$$

From eqs 7 and 10 it is possible to calculate the translational diffusion coefficient, and correspondingly, the intrinsic translational diffusion coefficient:

$$[D]_{\mathrm{sf}} = k/(9\pi 2^{1/2}((f/f_{\mathrm{sph}})_0)^{3/2}([s]\bar{v})^{1/2}) \qquad (11)$$

Linking, for each sample, s_0, f_0, and $(1 - v\rho_0)$ in the equation obtained from 9 and 11 allows one to obtain

$$M_{\mathrm{sf}} = 9\pi 2^{1/2} N_{\mathrm{A}}([s](f/f_{\mathrm{sph}})_0)^{3/2}\bar{v}^{1/2} \qquad (12)$$

This equation is the transformation of Svedberg equation. For $f/f_{\mathrm{sph}} = 1$ the equation is transformed into the relationship describing the hard spheres model.

Combining [s], [f] and [η] yields the hydrodynamic invariant, A_0:[2,26]

$$A_0 \equiv k(\Phi_0/100)^{1/3}P_0^{-1} = k(N_{\mathrm{A}}[s][\eta]/[f]^2)^{1/3} \qquad (13)$$

where k is Boltzmann's constant and [η] is expressed in 100 × cm^3/g.

Combining [s], [f] and k_{s} leads to the sedimentation parameter, β_{s}:[23,27]

$$\beta s \equiv B^{1/3}/P = (N_{\mathrm{A}}[s]k_{\mathrm{s}}/[f]^2)^{1/3} \qquad (14)$$

The ratios $\Phi^{1/3}/P$ and $B^{1/3}/P$, and thus A_0 and β_{s}, though theoretically not strictly constant, are experimentally found to be invariant within the incertitude of the measurements in the case of polymers of homologous structure but different contour lengths (molar masses). The mean destination of hydrodynamic invariants is to check the reasonable intercorrelations between the values which are differently related with size and molar mass (s, D, and [η]) and obtained by the different experimental techniques. The fact that the two hydrodynamic invariants are virtually constant makes it possible to estimate the molar mass of polymers from s_0 and [η], or from s_0 and k_{s} values.[2,23,28,29]

The limiting theoretical values of Φ and P for a Gaussian coil ($M \to \infty$), obtained after a preliminary averaging of the hydrodynamic Oseen tensor, are $\Phi_0 = 2.87 \times 10^{23}$ mol^{-1} and $P_0 = 5.11$.[5,30,31] This leads to $A_0 = 3.84 \times 10^{-10}$ g × cm^2 × s^{-2} × K^{-1} × mol$^{-1/3}$. The theoretical values of the Flory hydrodynamic parameters P_0 and Φ_0 are affected by the preaveraging of Oseen's hydrodynamic tensor, as studied by the Monte Carlo simulation method [32,33] and by renormalization group calculations,[34] leading to limiting values of 6.0 or 6.2 for P_0 and 2.5×10^{23} or 2.36×10^{23} for Φ_0, respectively. In these cases the theoretical values of A_0 will be 3.12×10^{-10} and 2.96×10^{-10}, correspondingly.

Global Multi-HYDFIT Analysis in Terms of the Wormlike Model. Although the chain skeleton of polystyrene copolymers—like that of other vinyl polymers—is essentially flexible, the copolymers can also be viewed as wormlike chains, due to their short length. The conformational parameters of wormlike chains are the persistence length, a (or the Kuhn segment length), the mass per unit length, M_L and the hydrodynamic diameter, d. The copolymers studied in this paper are of particular interest since, due to the bulkier side chains of the diphenyletylene units, the hydrodynamic diameter (the influence of which is usually neglected in long-chain hydrodynamics) may have an appreciable effect.

The analysis of the present data in terms of the wormlike model requires, first, a computational scheme relating solution properties to the conformational parameters. There is a classical theory by Yamakawa and Fujii,[31] which has the drawback of being based on the preaveraging approximation, which, as commented on in the previous section, will introduce important biases for rather flexible chains. Such defects are avoided in results based on a Monte Carlo simulation, coupled to rigorous hydrodynamics (the Monte Carlo, rigid-body approach proposed by Zimm and other authors[32,33]), carried out by Garcia Molina and García de la Torre some years ago,[35] whose results have been improved in recent recalculation). The Multi-HYDFIT program may optionally work with the classical Yamakawa−Fujii theory or with the newest Monte Carlo results.

The second piece in this kind of data analysis is the global fit of the various measured properties for various samples (molar mass, sedimentation, diffusion and intrinsic viscosity) to the predictions of the wormlike chain model, in order to identify the optimum values of the parameters a, M_L, and d. In the Multi-HYDFIT program,[11] this is done with the help of the so-called equivalent radii, a_X, which are the radii of a sphere that would have a given value of property X; for instance $a_T = f_t/6\pi\eta_0$ for the translational frictional properties (the Stokes radius), or $a_I = (3M[\eta]/10\pi N_{\mathrm{A}})^{1/3}$ for the intrinsic viscosity (which is similar but not identical to a_T). Then, for all the properties and all the samples, the square deviation of the experimental and calculated radii are evaluated, and combined in a quantity Δ^2 which equals the mean over all the properties and all the samples. Δ^2 (a, M_L, d) is a function of the three wormlike chain parameters, and it is treated by Multi-HYDFIT as the target function to be minimized in a search to find the best-fitting values of the parameters. Note that for the fitted best set of parameters, 100Δ can be considered as a typical percent relative deviation of the experimental and calculated equivalent radii. We also recall that Multi-HYDFIT makes a simulation to determine the uncertainties of the resulting parameters from user-supplied, or typical, uncertainties of the experimental data.

4. Results and Discussion

Velocity Sedimentation. For all samples the velocity sedimentation coefficients and frictional ratios were obtained by the $c(s)$ method implemented in Sedfit, as outlined above (Figure 1). For relatively high values of s this value can also be easily obtained in the classical way from the displacement of the midpoint (or maximum in the case of the differential distribution) of the boundary: $s = \Delta[\ln X]/\omega^2 t$, where X is the radial distance of the boundary from the rotation axis, ω is the angular velocity, and t is the time of sedimentation (Figure 2). The s-values calculated by this way coincide with those obtained for the corresponding concentrations by Sedfit.

Both the maximum entropy method and the Tikhonov−Philips second derivative method of regularization were applied during the treatments of velocity sedimentation data. The results virtually coincided (Table 1). The further interpretation used the average values. The concentration dependence was studied for all samples and followed the linear regression, in accordance with the relationship: $s^{-1} = s_0^{-1}(1 + k_{\mathrm{s}}c + ...)$, where s_0 is velocity sedimentation coefficient (at zero concentration) and k_{s} is the corresponding

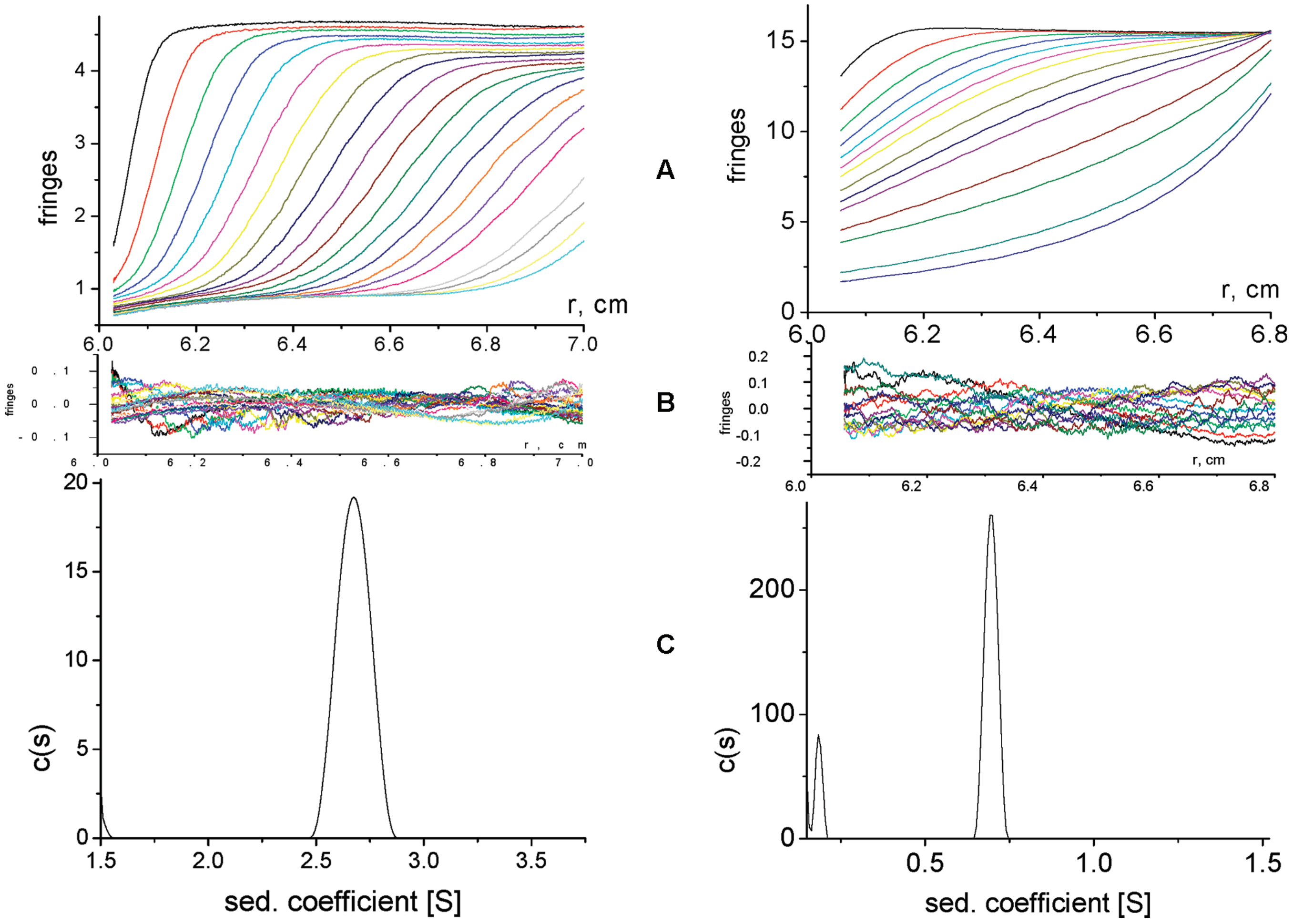

Figure 1. Raw sedimentation profiles of alternating styrene/diphenylethylene copolymers functionalized with terpyridine in toluene. (a) Sample 1 with c = 1.6 mg/cm^3; (b) sample 7 with c = 23 mg/cm^3. Key: (A) superposition of interference profiles; (B) corresponding residual plots; (C) distributions $c(s)$ of sedimentation coefficients, obtained with a regularization procedure with a confidence level of 0.90. In some cases an accessory minor peak around 0.1−0.15 S is observed (see part b); it was ignored in the further interpretation.

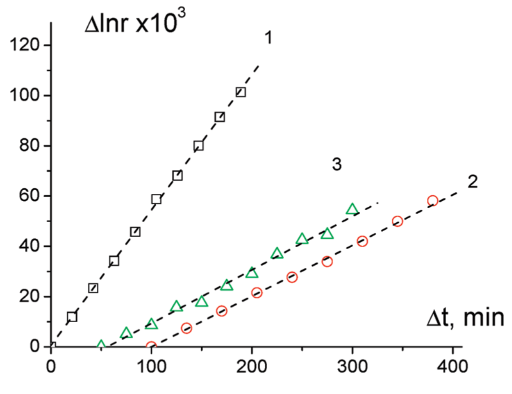

Figure 2. Dependencies of Δ[ln r] on sedimentation time Δt for two samples of the alternating styrene/diphenylethylene copolymers functionalized with terpyridine. Solvent: toluene. r is the position of the midpoint of the sedimenting boundary. The velocity sedimentation coefficient is determined from the slop of the dependencies Δ[ln r] vs Δt as s = Δ[ln r]/Δ$t\omega^2$, where $\omega = 2\pi n$ and n is number of rotations per second. Key: (1) n = 55 000 rpm, c = 1.63 mg/cm^3, s = 2.71 S; (2) n = 40 000 rpm, c = 7.79, s = 1.92 S; (3) n = 40 000 rpm, c = 2.02, s = 1.87 S. Numbers 1−3 correspond to the sample numbers in Table 1. Lines 3 and 2 are shifted with respect to line 1 along the time axis by 50 and 100 min, correspondingly.

concentration coefficient (Gralen coefficient). For more polydisperse samples (when $M_w/M_n \approx 2$) it is possible to extract the Gralen coefficient from the velocity sedimentation data of the sample. This approach is demonstrated in ref 36 and in the Sedanal program.[37,38]

Small shifts in the temperature of the sedimentation, diffusion and viscometric experiments were taken into account by comparing the corresponding characteristic values. It should be noted that, as toluene is an athermic solvent for such polymers (polystyrene, superpolystyrene), small differences in temperature will not change the size of a polymeric chain in a solution.

For homologous species, all the hydrodynamic values themselves and molar masses may be related through scaling relations of the Kuhn−Mark−Houwink-Sakurada type: $P_i = K_{ij}P_{j\neq i}^{b_{ij}}$ where P_i, $P_{j\neq i}$ are [η], s_0, D_0, k_s and P_j may also be M. For a homologous series the scaling indices are intercorrelated.[23,26,29]

A scaling relation linking s_0 and k_s of $k_s = 8.51 \times s_0^{0.93\pm0.13}$ was observed (Figure 3). In addition, it was established that the scaling indices b_{k_s} and b_s ($k_s = K_{sk_s}s_0^{bk_s}$ and $s_0 = K_sM^{b_s}$) are linked by the relation $b_{k_s} = (2-3b_s)/b_s$, which allows us to predict the b_s value.[29,39] In our case we could expect $b_s \approx 0.50$, which leads to an estimated molar mass range given by $M_{max}/M_{min} \approx (s_{max}/s_{min})^2 \approx 13$.

The concentration sedimentation coefficient k_s may be compared also with the value of the intrinsic viscosity [η], which has the same dimension. The obtained average value of the dimensionless ratio k_s/[η] = 1.7 ± 0.3 is typical for linear flexible polymers.[40]

Table 1. Hydrodynamic Characteristics of Alternating Styrene/Diphenylethylene Copolymers Functionalized with Terpyridine in Toluene at 20 °C, As Obtained by Sedfit Treatment of the Data Acquired with a Beckman XLI Analytical Ultracentrifuge

		maximum entropy method			Tikhonov−Philips method			average values		
N	$c \times 10^2$ [g/cm^3]	s [S] (±0.04)	k_s [cm^3/g]	f/f_{sph} (±0.05)	s [S] (±0.04)	k_s [cm^3/g]	f/f_{sph} (±0.05)	s_0^{av} [S]	k_s^{av} [cm^3/g]	$(f/f_{sph})_0^{av}$
1	0.735	2.46	16 ± 2	2.31	2.44	17.7 ± 0.6	2.44			
	0.466	2.54		2.19	2.56		2.15			
	0.163	2.68		1.90	2.69		1.89			
	0	2.74		1.81	2.76		1.75	2.75	17	1.78
2	0.779	1.96	18 ± 7	1.94	1.92	21 ± 3	1.94			
	0.387	2.15		1.83	2.09		1.82			
	0.204	2.14		1.67	2.14		1.69			
	0	2.25		1.61	2.24		1.63	2.25	19	1.62
3	0.823	1.64	22 ± 2	1.48	1.59	20.3 ± 0.6	1.48			
	0.322	1.81		1.52	1.75		1.49			
	0.202	1.85		1.52	1.78		1.50			
	0	1.93		1.55	1.86		1.51	1.90	21	1.52
4	1.989	1.20	9 ± 1	1.68	1.15	12.2 ± 0.7	1.43			
	1.058	1.28		1.45	1.28		1.47			
	0.250	1.38		1.40	1.39		1.38			
	0	1.41		1.33	1.43		1.39	1.42	11	1.36
5	0.983	1.01	8 ± 3	1.31	0.95	10 ± 2	1.30			
	0.443	1.07		1.32	1.01		1.35			
	0.203	1.07		1.26	1.02		1.30			
	0	1.10		1.27	1.05		1.33	1.07	9	1.30
6	1.0995	0.76	8 ± 2	1.31	0.80	6 ± 2	1.24			
	0.7625	0.79		1.32	0.83		1.23			
	0.321	0.81		1.32	0.84		1.24			
	0	0.84		1.31	0.86		1.23	0.85	7	1.27
7	2.291	0.67	5 ± 1	1.21	0.63	8 ± 4	1.29			
	1.363	0.69		1.13	0.67		1.29			
	0.583	0.73		1.11	0.71		1.25			
	0	0.75		1.10	0.74		1.24	0.75	6	1.17

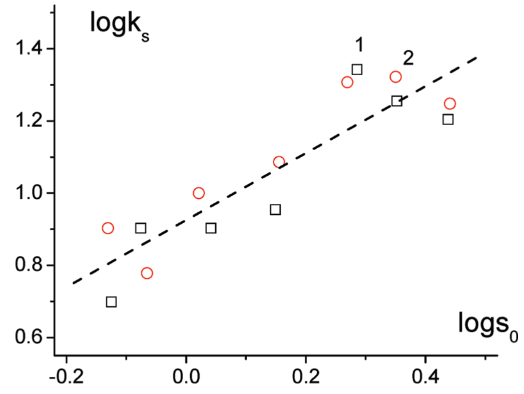

Figure 3. Double logarithmic plot of the concentration coefficient k_s vs s_0. The dashed line follows the relation k_s = 8.41 × $s_0^{0.93\pm0.13}$. Key: (1) s_0 and k_s values were obtained with the maximum entropy method; (2) these values were obtained with the Tikhonov−Philips method (Table 1).

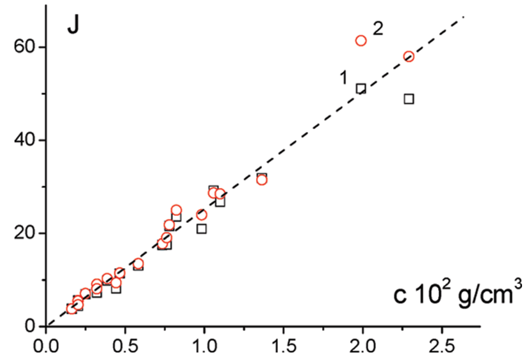

Figure 4. Total number of fringes J vs concentration of alternating styrene/diphenylethylene copolymers functionalized with terpyridine. Solvent: toluene. The refractive index increment was calculated according to the following relation: $\Delta n/\Delta c$ = 5.625 × 10^{-5}(J/c). The results were obtained with the maximum entropy method (1) or with the Tikhonov−Philips method (2).

Figure 4 shows the total number of fringes as a function of copolymer concentration, superimposed for all samples. From the linear plot we derive $\Delta n/\Delta c$ = (0.142 ± 0.003) cm^3/g for the terpyridine end-functionalized poly(styrene-*alt*-diphenylethylene) copolymers in toluene at λ = 675 nm. When compared to the corresponding value for polystyrene in toluene at λ = 633 nm, $\Delta n/\Delta c$ = (0.107 ± 0.004) cm^3/g,[41] the present value is higher. The difference is obviously related to the higher concentration of phenyl rings in the copolymer studied here.

The buoyancy factor $(1-\bar{v}\rho_0)$, which is required for the calculation of the molar mass from sedimentation velocity measurements, was determined from density measurements. For the four samples, a value of 0.266 ± 0.005 was obtained from the limiting slope of the plot of solution density versus solute concentration. From the buoyancy factor, the partial specific volume, which is a characteristic property of a system, was determined as $\bar{v}$ = (0.848 ± 0.005) cm^3/g.

Translational Diffusion Coefficient. The fitting values of (f/f_{sph}) were extrapolated to zero concentration following the linear approximation: $(f/f_{sph}) = (f/f_{sph})_0(1 + k_f c + ...)$. The concentration coefficient k_f has the same meaning as k_s but in most cases its experimental error is about two times larger than that of the k_s value. From the extrapolated value of $(f/f_{sph})_0$ the intrinsic translational diffusion coefficient [D] can be calculated. Its values were compared to those obtained with the classical technique of boundary formation (Figures 5 and 6). The virtual coincidence observed proofs that, for linear polymer chains with narrow length distribution, the $(f/f_{sph})_0$ value obtained from velocity sedimentation experiments by the use of the Sedfit program is equivalent to the data from classical diffusion measurements. Thus, for such polymers of low molar mass, velocity sedimentation experiments evaluated with the Sedfit program can be considered as an absolute method for the determination of molar mass.

Hydrodynamic Invariants. The intercorrelation between the experimental hydrodynamic values (s_0, D_0, $[\eta]$, and k_s) may be checked by calculation of the hydrodynamic invariant and sedimentation parameter values. In our case, the relationships for the two hydrodynamic invariants (eqs 13 and 14) can by easily transformed into the following relations which have a clear physical meaning:

$$A_0 = kN_A{}^{1/3}(9^2\pi^2 2)^{-1/3}\frac{([\eta]/\upsilon)^{1/3}}{(f/f_{\mathrm{sph}})_0} = 0.116 \times kN_A{}^{1/3}\frac{([\eta]/[\eta]_{\mathrm{sph}})^{1/3}}{(f/f_{\mathrm{sph}})_0} \quad (15)$$

$$\beta_s = N_A{}^{1/3}(9^2\pi^2 2)^{-1/3}\frac{(k_s/\upsilon)^{1/3}}{(f/f_{\mathrm{sph}})_0} = 0.162 \times N_A{}^{1/3}\frac{(k_s/k_{\mathrm{ssph}})^{1/3}}{(f/f_{\mathrm{sph}})_0} \quad (16)$$

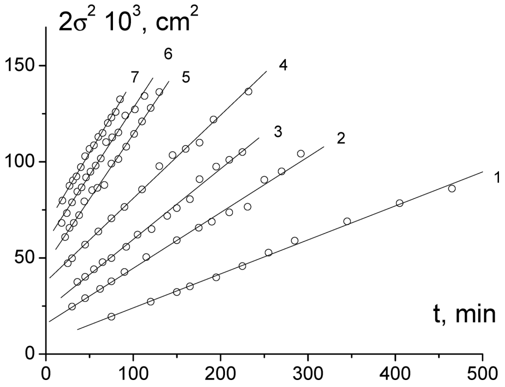

Figure 5. Dependence of the diffusion boundary dispersion $2\sigma^2$ vs time of diffusion t, for the samples of alternating styrene/diphenylethylene copolymers functionalized with terpyridine. Solvent: toluene. The numbers 1 to 7 correspond to the sample numbers in Table 1. The different curves were displaced from each other along the σ^2-axis by 10 cm^2.

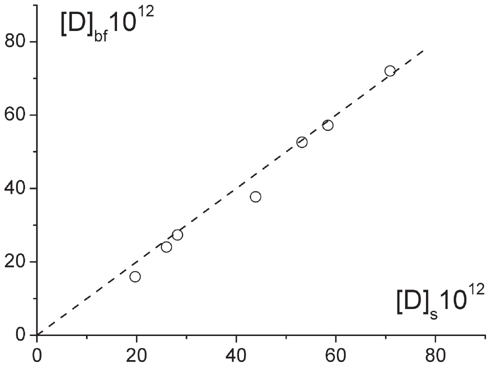

Figure 6. Comparison of the intrinsic diffusion coefficients $[D] \equiv D_0\eta_0/T$ obtained in two independent ways: from evaluating velocity sedimentation experiments by use of the Sedfit program ($[D]_s$ according to eq 11), and from separate experiments on the formation of a diffusion boundary ($[D]_{bf}$ according to Figure 5 and eqs 1 and 2). The dashed line has a slope of 1.

The numerators and denominators of these relations represent the ratios of the hydrodynamic values of the macromolecule and the corresponding values of a spherical particle. The numerators are the ratios of hydrodynamic values which are related to the volume of the macromolecules (intrinsic viscosity $[\eta]$ and Gralen coefficient k_s), and the denominators are the ratio of translational friction coefficients related with the linear size of macromolecule and sphere. The last terms of eqs 15 and 16 are obtained taking into account the known Einstein $[\eta] = 2.5\bar{\upsilon}$ and Burgers $k_{\mathrm{ssph}} = (55/8)\bar{\upsilon}$ expressions.[1,2,23] The more recent multiple scattering theory of the concentration dependence of the cooperative friction coefficients for suspensions of interacting spheres (k_{ssph}) yield a value of the numerical multiplier which is only 2% lower that the earlier value for the rigid sphere obtained by Burgers.[42]

The calculated values of A_0 and β_s are presented in Table 2. They do not change regularly with the change of molar mass of the samples, and their average values are characteristic for linear macromolecules.[2,26-29] The average value of A_0 is close to that calculated from the values of the hydrodynamic parameter Φ and P which were obtained without preaveraging Oseen's hydrodynamic tensor.

Molar Mass and Scaling Relationships. Molar masses were calculated using the Svedberg relationships[9,12] and are presented in Table 2. First we will compare the hydrodynamic values among themselves and also with the molar masses in the frame of Kuhn−Mark−Houwink-Sakurada relationships or the scaling relationships. Some of corresponding plots are presented in Figure 7.

The parameters of the scaling relationships are collected in Table 3. The first conclusion that follows from this set of scaling indices is that b_s, b_η, and $|b_D|$ are all close to 0.50. This finding may be explained by the low molar mass range of the studied copolymers. In this region of M the

Table 2. [a] **Hydrodynamic Characteristics, Molar Mass, and Hydrodynamic Invariants of Alternating Styrene/Diphenylethylene Copolymers Functionalized with Terpyridine in Toluene at 20 °C**

N	$[s] \times 10^{15}$ g/cm	$[\eta]$ cm^3/g	k_s cm^3/g	$[D]_s{}^b \times 10^{12}$ g cm/s^{2}K	$M_{fs} \times 10^{-3}$ g/mol	$\beta_s{}^b \times 10^{-7}$ mol$^{-1/3}$	$A_0{}^b \times 10^{10}$ g × cm^2/s^2 × K × mol$^{1/3}$	$[D]_{bf}{}^c \times 10^{12}$	$M_{sD} \times 10^{-3}$	$\beta_s{}^c \times 10^{-7}$	$A_0{}^c \times 10^{10}$
1	6.08	13.1	17	19.7	25.2	1.07	2.97	15.9	31.6	0.92	2.55
2	4.97	10.4	19	26.0	15.7	1.25	3.10	24.0	17.2	1.18	2.91
3	4.20	8.3	21	28.2	11.9	1.38	2.97	27.3	13.0	1.30	2.80
4	3.14	5.9	11	43.9	6.0	1.18	3.04	37.7	6.9	1.08	2.79
5	2.36	5.0	9	53.2	3.7	1.22	3.11	52.6	3.8	1.19	3.04
6	1.88	3.7	7	58.4	2.6	1.17	2.70	57.2	2.7	1.15	2.65
7	1.66	3.0	6	70.9	1.7	1.19	2.98	72.0	1.9	1.11	2.78
av.						1.21 ± 0.08	2.98 ± 0.09			1.13 ± 0.07	2.8 ± 0.1

[a] Some data in this table was taken from ref 14. [b] Intrinsic coefficient of translational diffusion calculated from $(f/f_{\mathrm{sph}})_0$ values which were obtained by analytical ultracentrifugation. [c] Intrinsic coefficient of translational diffusion obtained from experiments using boundary formation techniques.

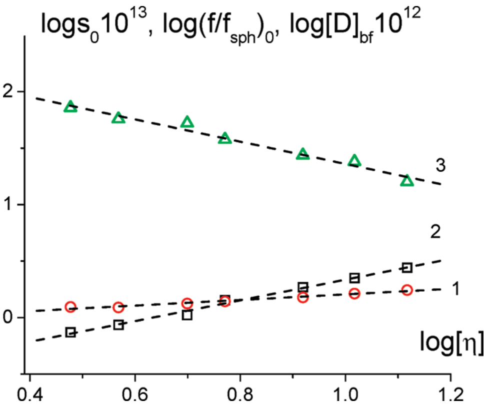

Figure 7. Double logarithmic plot of hydrodynamic values (Kuhn-Mark−Houwink-Sakurada plots) for the alternating styrene/diphenylethylene copolymers functionalized with terpyridine. Solvent: toluene. Key: (1) $(f/f_{\mathrm{sph}})_0$ vs [η], (2) s_0 vs [η], and (3) $[D]_{\mathrm{bf}}$ vs [η].

Table 3. Parameters of the Kuhn−Mark−Houwink−Sakurada (Scaling) Relationship for Alternating Styrene/Diphenylethylene Copolymers Functionalized with Terpyridine in Toluene

$P_i - P_j$[a]	$b_{ij} \pm \Delta b_{ij}$	$\log K_{ij} \pm \Delta[\log K_{ij}]$	K_{ij}	r_{ij}[b]
$s_0/[\eta]$	0.92 ± 0.04	$-(0.58 \pm 0.03)$	2.62×10^{-14}	0.9953
$(f/f_{\mathrm{sph}})_0/[\eta]$	0.25 ± 0.02	$-(0.041 \pm 0.015)$	0.91	0.9859
$[D]_{\mathrm{bf}}/[\eta]$	$-(0.99 \pm 0.07)$	2.35 ± 0.06	2.21×10^{-10}	−0.9869
k_s/s_0	0.93 ± 0.13	0.93 ± 0.03	8.51	0.8947
s_0/M	0.49 ± 0.02	$-(1.38 \pm 0.06)$	4.18×10^{-15}	0.9972
$(f/f_{\mathrm{sph}})_0$/M	0.16 ± 0.02	$-(0.47 \pm 0.06)$	0.338	0.9746
D_0/M	$-(0.53 \pm 0.02)$	3.59 ± 0.09	3.91×10^{-4}	−0.9955
[η]/M	0.53 ± 0.02	$-(1.24 \pm 0.08)$	5.76×10^{-2}	0.9959

[a] The properties P_i and P_j of all the samples are related by: $\log P_i = \log K_{ij} + b_{ij} \log P_j$. [b] r is the linear correlation coefficient.

excluded volume effects are not manifested.[43,44] Taking into account the experimental error of the scaling indices determination it may be concluded that they follow the intercorrelation $|b_{\mathrm{D}}| = (1 + b_\eta)/3$, $b_{\mathrm{D}} = -(b_{\mathrm{ff}} + (1/3))$, where b_{ff} is the scaling index in the relation of $f = K_{\mathrm{f}} M^{b_{\mathrm{ff}}}$. These are characteristic of a homologous series of linear polymers.

Global-Fit Estimation of Wormlike-Chain Conformation Parameters. Our further interpretation of hydrodynamic data is related with the estimation of gross conformational parameters corresponding to a wormlike-chain representation of the studied short-chain copolymers. Because the experimental data are in the range of low molar masses, excluded volume effects can be safely ignored,[30,31,45] and therefore, the Monte Carlo simulation of wormlike chains without excluded volume effects,[32] whose results are implemented in the Multi-HYDFIT analysis,[11] is applicable (we recall that the Yamakawa−Fujii also ignored the excluded-volume effects). It is also remarkable that, for our short oligomers, the ratio of contour length to the Kuhn segment length (as it can be deduced from the parameters that will we reported below) is in the range 1−10, which covers well the typical region of this ratio for wormlike chains, from weakly bending to appreciably coiled chains.

In order to judge the outcome of the global-fit results for the wormlike-chain parameters, it is convenient to make estimations that come easily from the chemical structure of the repeat units of the macromolecule. Thus, the thickness of the chain is about 0.8 nm at the styrene units and about 1.2 nm at the diphenylethylene units. Also M_{L} can be estimated with a L value corresponding to the fully stretched, all-trans chain skeleton. For the repeating unit, with chemical formula $C_{22}H_{20}$, the molar mass is 284.4 g/mol, and the length of its 4 C−C bonds in all trans conformation is $4 \times 0.154 \times \cos(35°) = 0.504$ nm, so that we obtain $M_L = 564.3$ g/(mol × nm). Furthermore, another estimation of the diameter can be made by equating the volume per unit length of the cylinder, $\pi(d/2)^2$, with that derived from the specific volume $M_L\bar{v}/N_{\mathrm{A}}$. For $M_L = 564.3$ g/(mol × nm) and $\bar{v} = 0.847$ cm^3/g we estimate $d = 1.0_1$ nm.

Table 4. Mass per unit length of chain (M_{L}), hydrodynamic diameter (d), persistence length (a) and target function Δ

wormlike theory/ data set	M_L, g/ (mol × nm)	d, nm	a, nm	100Δ (% diff)
Monte Carlo	**600 ± 100**	**1.1 ± 0.2**	**2.1 ± 0.4**	5.1
Monte Carlo	580 ± 40	1.0[a]	2.1 ± 0.2	5.9
Monte Carlo	564[a]	1.1 ± 0.1	2.0 ± 0.1	5.9
Yamakawa−Fujii	360 ± 30	0.53 ± 0.07	1.8 ± 0.2	12.8
Yamakawa−Fujii	400 ± 30	1.0[a]	2.8 ± 0.1	23.0
Yamakawa−Fujii	564[a]	0.59 ± 0.05	6.2 ± 1.6	26.3

[a] Corresponding fixed parameters, either M_L or d.

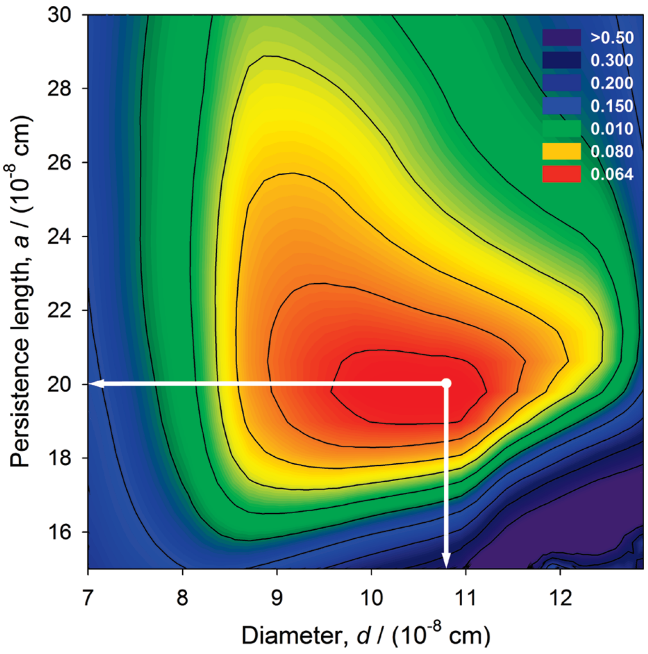

Figure 8. Contour plots of the score function △, obtained by Monte Carlo calculations on a wormlike chain of varying persistence length, a, and hydrodynamic diameter, d, with a fixed value of $M_L = 564$ g/(mol nm).

Multi-HYDFIT includes a simulation of the uncertainties in the resulting parameters arising from the expected range of errors of the experimental data, which were assigned—rather conservatively—as follows: 5% for translational properties, 8% for the intrinsic viscosity and 10% for the molar mass. The most critical fit is that to determine simultaneously the three parameters a, M_{L}, and d. Alternatively one could fix either M_{L} or d, assigning the above-mentioned chemical estimate, and floating only the two other parameters. In such cases, the fixed parameter is indicated in the Table 4 by footnote[a]. This kind of analysis is useful to visualize the quality of the fits in contour plots of the Δ score function vs. two of the parameters. Examples of such plots are presented in Figures 8 and 9. It is clear that the minimum that determines the optimum parameters is well-defined.

The global analysis in Multi-HYDFIT can be carried out alternatively using the classical Yamakawa−Fujii (YF) theory as the source of the theoretical results. Indeed this was the only choice before the introduction of our new Monte

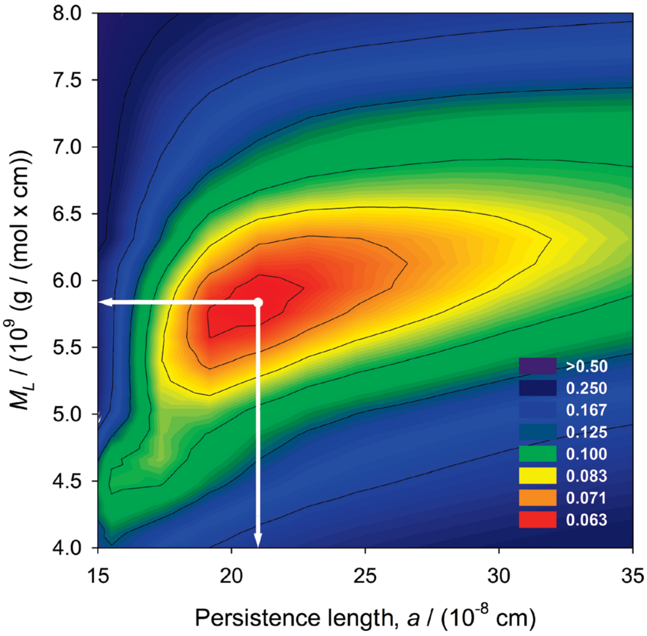

Figure 9. Contour plots of the score function Δ, obtained by Monte Carlo calculations on a wormlike chain of varying mass per unit length, M_L, and persistence length, a, with a fixed value of $d = 1.0$ nm.

Carlo (MC) results, while now they are both options of the program. The implementation of the new simulation results removes possible defects from the preaveraged hydrodynamic interaction in the YF treatment, and includes a better description of the effect of the hydrodynamic diameter with, as mentioned above, is relevant for our short and thick chains. Table 4 reveals that the global fit is much better using the MC predictions than the YF ones; the goodness of the fit, as measured by Δ, is better with the MC treatment. The typical errors in the prediction of the equivalent radii are about 5% and 13% with the MC and YF treatments, respectively. Clearly, the persistence length of the poly (styrene-*alt*-diphenylethylene) copolymers is about $a \approx 2.0$ nm. However, while the predictions derived from the MC treatment for the mass per unit length, $M_L \approx 600$ g/(mol nm), and diameter, $a \approx 1.0$ nm, agree very well with the above-described estimations based on the chemical structure and the specific volume of the polymer, the results using the YF theory seem rather deviated. The calculated persistence length of the poly (styrene-*alt*-diphenylethylene) copolymers is about two times higher than the value of the persistence length corresponding to the polystyrene chains, which can be explained by the restricted internal rotation due to the extra phenyl rings in the copolymer chains.

Conclusion

We used the interference optics of the Beckman XLI analytical ultracentrifuge for the study of the velocity sedimentation of samples of terpyridine-functionalized poly(styrene-*alt*-diphenylethylene) copolymers with narrow molar mass distribution. The sedimentation data were treated with the Sedfit program which numerically solves the Lamm equation. The Sedfit program can determine both the velocity sedimentation coefficient and the frictional ratio. The latter parameter is unambiguously related to the translational diffusion coefficient. The adequacy of its determination by the Sedfit program was checked by an independent experimental determination of the translational diffusion coefficient. As a consequence, velocity sedimentation experiments evaluated by the use of the Sedfit program may be considered as a self-sufficient method for the determination of the molecular characteristics of linear polymers with narrow molar mass distributions. The recently developed Multi-HYDFIT program performing a joint analysis of different transport properties of multiple samples allows the adequate estimation of the conformational characteristics of short flexible chains without the volume effects. This work also provides an adequate test of the ability of the recently developed Multi-HYDFIT program for the joint analysis of different transport properties of multiple samples, coupled to the MC simulation results for the wormlike chains that improve the classical YF theory. Indeed this first and successful application to the global-fit methodology to short, synthetic oligomers reveals its interest beyond the typical biopolymer systems to which it had been applied previously.

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