COMPARISON BETWEEN THE CLASSICAL AND QELSS DETERMINATION OF DIFFUSION COEFFICIENTS. SHARP POLYMER FRACTIONS

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Simple modification of the method of cumulants for an evaluation of QELSS data within the error limit of 2% in the case of narrow polymers is suggested. Model calculations of the deviations from the exponential decay due to polydispersity are presented, criteria of applicability of the 1st and 2nd cumulant fit at a given accuracy are derived and verified experimentally. The required agreement between the classical and QELSS diffusion constant is achieved if the QELSS measurements are made at concentrations where the effect of concentration dependence of the diffusion constant may be neglected.

Time resolved light scattering experiments are able to reveal various dynamic processes in solution. The technique, often referred to as quasielastic light scattering spectroscopy (QELSS), has in recent years become a standard tool used to determine diffusion coefficients in polymer solutions. Due to the nature of the QELSS experiment and since the technique lends itself to a high level of computerization, such determinations can be done very rapidly.

One important application of the diffusion coefficients of polymers is to use them for sample characterization either by themselves or, preferably, in combination with sedimentation data, to determine absolute molar masses according to the Svedberg formula. Some years ago we published[1-3] extensive precision data obtained by classical techniques (sedimentation, diffusion, viscosity) which characterized a set of very sharp polystyrene fractions over a wide range of molar masses. From these results it clearly emerged that velocity sedimentation and diffusion can be brought to such a perfection that molar masses accurate enough for calibration purposes could be obtained. The key measurement is the determination of diffusion coefficients where it is necessary to keep the experimental error within ±2%. The accuracy of molar masses of polymer standards is known to be a limiting factor in the calibra-

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tion of GPC column sets and has been recently reconsidered as the main limitation of the precision of the thermal field flow fractionation technique[4].

Since QELSS is such a common technique, it should obviously be valuable to compare carefully the precision and accuracy of QELSS data with the results of classical diffusion experiments, the more so as one can find in the literature indications of an overestimation of QELSS precision and also objections based on the poor reproducibility of data measured on the same sample in different laboratories. Let us note in this connection three different $D \propto M^{-\alpha}$ relations determined by QELSS (refs[5-7]) for standard polystyrenes in butanone. Using these relations we get for $M_w = 5 . 10^5$ diffusion coefficients $3{\cdot}55 . 10^{-7}$, $3{\cdot}43 . 10^{-7}$ and $2{\cdot}96 . 10^{-7}$, i.e., deviations of $+3{\cdot}5\%$ and $-13{\cdot}7\%$. Comparing the cyclohexane data measured by King et al.[8] for standard polystyrenes with M_w values $1{\cdot}1 . 10^5$ and $6{\cdot}7 . 10^5$ with the data of Han[9] for the same samples we get the differences $1{\cdot}7\%$ and $6{\cdot}7\%$, respectively.

Still having access to the samples used in our classical measurements, we decided to perform such a comparative investigation to establish whether the precision and accuracy of QELSS data is sufficient for the new method to replace the classical difusion methods in the high precision determination of molar masses of standard polymers by the Svedberg formula. The aim of this study is to select carefully the experimental conditions and to evaluate the computational approach based on cumulants, in view of the experimental peculiarities and effects of the molar mass distributions.

THEORETICAL

In the case of a polydisperse polymer in solution the electric field autocorrelation function is[10]

$$|g^{(1)}(\tau)| = \int_0^\infty G(\gamma) \exp(-\gamma\tau) \, d\gamma \,, \qquad (1)$$

where τ is the delay time and $\gamma = K^2 D$ is the decay rate, D being the diffusion coefficient. The scattering vector is $K = (4\pi n/2) \sin(\Theta/2)$, where λ is the wavelength of light in vacuum, n is the solvent refractive index, and Θ is the scattering angle. The normalized distribution function of decay rates, $G(\gamma) \, d\gamma$, is defined as a z-fraction of the total intensity scattered by molecules having γ within the increment $d\gamma$, i.e., in terms of z_i arguments as the general statistical weight[11] $G(\gamma) \, d\gamma = z_i/\sum z_i = c_i M_i/\sum c_i M_i$, c_i being the mass concentration. From this definition one directly obtains the relation between the distribution of decay rates relevant for the QELSS experiment and the mass-defined molar mass distribution $f(M) \, dM$,

$$G(\gamma) \, d\gamma = \frac{1}{M_w} M \, f(M) \, dM \,, \qquad (2)$$

which enters into the classical diffusion experiment through the mass defined distribution of diffusion coefficients $g(D)\,dD$, noting simply that $f(M)\,dM = -g(D)\,dD$, M_w being the mass-average molar mass. Clearly, the difference in the definitions of $G(\gamma)$ and $g(D)$ results in a different average value of D determined by QELSS and by the classical diffusion. It will be shown below, however, that this difference does not exceed the experimental error in the case of extremely sharp polymer samples.

The measured intensity autocorrelation function $I^{(2)}(\tau)$ in a homodyne experiment relates to the electric field autocorrelation function as[10]

$$I^{(2)}(\tau) = B(1 + C^2|g^{(1)}(\tau)|^2)\,, \tag{3}$$

where B and C are constants. In the logarithmic form we get

$$\ln\left[\frac{I^{(2)}(\tau)}{B} - 1\right] = \ln \text{const.} + 2\ln|g^{(1)}(\tau)|\,. \tag{4}$$

For a monodisperse solution the homodyne autocorrelation function decays exponentially $g^{(1)}(\tau) = \exp(-\gamma\tau)$ and Eq. (*4*) represents a straight line.

In a polydisperse solution Eq. (*1*) may be analyzed by the method of cumulants[12,13],

$$\ln|g^{(1)}(\tau)| = \sum_{m=1}^{\infty} K_m(\gamma)(-\tau)^m/m!\,, \tag{5}$$

where $K_1 = \bar{\gamma} = \int_0^\infty \gamma\, G(\gamma)\, d\gamma = \mu_1$, i.e., the 1st algebraic moment of $G(\gamma)$ and higher cumulants are expressed in terms of central moments of $G(\gamma)$ as $K_2 = \mu_2'$, $K_3 = \mu_3'$, $K_4 = \mu_4' - 3\mu_2'^2$, μ_m' being defined by $\mu_m' = \int_0^\infty G(\gamma)(\gamma - \bar{\gamma})^m\, d\gamma$. Eq. (*5*) may be rewritten in the form

$$\ln|g^{(1)}(\tau)| = -\bar{\gamma}\tau[1 - R\bar{\gamma}\tau + Q(\bar{\gamma}\tau)^2 - S(\bar{\gamma}\tau)^3]\,, \tag{6}$$

where $R = \mu_2'/2\mu_1^2$, $Q = \mu_3'/6\mu_1^3$, $S = (\mu_4' - \mu_2'^2)/24\mu_1^4$. In this way the polydispersity terms (deviation from the straight line) are compared to unity and may be calculated using some specific forms of the distribution function. In practice, higher terms are omitted due to the fact that the least squares polynomial fitting procedure used[12] to calculate the cumulants will never yield the higher terms with sufficient accuracy. For a given distribution the question then arises whether one can specify the conditions under which a linear, quadratic or cubic approximation achieves the desired accuracy, in other words, over how many channels a straight line or a parabola adequately describes $\ln|g^{(1)}(\tau)|$ for a given degree and type of polydispersity.

This can be shown on the basis of Eq. (*6*) assuming some specific forms of the molar mass distribution. The m^{th} algebraic moment of $G(\gamma)$ is defined as

$$\mu_m = \int_0^\infty G(\gamma)\,\gamma^m\, d\gamma\,. \tag{7}$$

Noting that $\gamma(M) = K^2 D(M)$ and

$$D(M) = K_D M^{-\alpha}, \tag{8}$$

we get from Eq. (*2*) and Eq. (*7*)

$$\mu_m = \frac{(K^2 K_D)^m}{M_w} \int_0^\infty M^{1-m\alpha} f(M) \, dM. \tag{9}$$

Using then the Schulz–Zimm distribution function[14]

$$f(M) = \frac{a^{b+1}}{\Gamma(b+1)} M^b \exp(-aM), \tag{10}$$

$(a, b > 0)$ and performing the integration in Eq. (*9*), we get

$$\mu_m = \frac{(K^2 K_D)^m a^{m\alpha} \Gamma(b - m\alpha + 2)}{\Gamma(b+2)}. \tag{11}$$

For the Pearson distribution[14]

$$f(M) = \frac{\beta^{\varepsilon - 1}}{\Gamma(\varepsilon - 1)} M^{-\varepsilon} \exp(-\beta/M) \tag{12}$$

we obtain similarly

$$\mu_m = \frac{(K^2 K_D)^m \Gamma(m\alpha + \varepsilon - 2)}{\Gamma(\varepsilon - 2) \beta^{m\alpha}}. \tag{13}$$

Since the arguments of the gamma functions in Eqs (*11*) and (*13*) must be positive, we see that $\varepsilon > 2$ in Eq. (*13*). A more complex situation arises with regard to Eq. (*11*), where higher moments exist only if $b - m\alpha + 2 > 0$. The R-, Q-, and S- terms in Eq. (*6*) may then be calculated, if the central moments are expressed in terms of the algebraic moments: $\mu'_2 = \mu_2 - \mu_1^2$, $\mu'_3 = \mu_3 - 3\mu_2\mu_1 + 2\mu_1^3$, $\mu'_4 = \mu_4 - 4\mu_3\mu_1 + 12\mu_2\mu_1^2 - 3\mu_2^2$.

It is seen after some algebra that R, Q, S depend solely on b, ε, α and on the corresponding gamma functions, and may be calculated for a given polydispersity index ($M_w/M_n = 1 - 1/b$ in the case of the Schulz–Zimm function[14] and $M_w/M_n = (\varepsilon - 1)/(\varepsilon - 2)$ in the case of the Pearson distribution) for a given solvent quality characterized by the value of α.

As soon as the coefficients R, Q, S are known, we can specify the conditions for the required precision of use of the single exponential (1st order cumulant fit) and/or a second order cumulant fit approximations. In this way, the problem of balancing the statistical and systematic errors[12] in cumulant expansion may be greatly simplified by using an error chosen a priori. We can use, similarly to our classical diffusion[1], the requirement of a 2% error and calculate the $(\bar{\gamma}\tau)_{\max}$ values as a function of the width of the molar mass distribution, still compatible with this error for the first or second order cumulant fit.

The corresponding simple conditions follow from Eq. (*6*): as a good approximation a linear condition for the single exponential fit

$$R(\bar{\gamma}\tau) \leqq 0{\cdot}02\,, \tag{14}$$

and a quadratic condition for the second order cumulant fit

$$0{\cdot}02[1 - R(\bar{\gamma}\tau)] \geqq Q(\bar{\gamma}\tau)^2\,. \tag{15}$$

EXPERIMENTAL

The standard polystyrene (Pressure Chem. Co., 5a) with M_w 498 000 and a polydispersity index $M_w/M_n < 1{\cdot}06$ was the same sample as that used in our previous classical diffusion work[1]. Toluene (Merck, B.R.D.) was reagent grade.

The fluorimeter cuvettes were thoroughly cleaned in nitric acid prior to use and then carefully washed with solvent filtered through a combined Presep silica cartridge (Tessek Ltd., Prague, Czechoslovakia) and Millex-SR 0·5 micron filter (Millipore Corp. Bedford, U.S.A.), a procedure which removes both particulate and dissolved polar contaminants. Solutions were prepared by weight, filtered in the same way, and put into fluorimeter cuvettes.

No special thermostating unit was used, but the room temperature (near to 25°C) was measured periodically and found to be fairly stable.

The light scattering apparatus comprised a coherent Super-Graphite CR-4 argon-ion laser (Coherent Laser Division, Palo Alto, CA, U.S.A.), equipped with and intra-cavity etalon and tuned to 514·5 nm, a Hamamatsu R 268 photomultiplier tube (Hamamatsu Photonics K. K., Japan), an amplifier-discriminator supplied by Lindmark Innovation AB, Sweden and a 64 channel Langley–Ford digital correlator (Langley–Ford Instruments, Amherst, MA, U.S.A.). The correlator was connected to a JET 80G computer (Jet Computer AB, Sweden) foron-line data manipulation and analysis. The optical components were mounted on a massive steel bench (Newport Research Corporation, CA 92708, U.S.A.). The laser beam was spatially filtered prior to focussing on the cuvette, and the scattered light was detected at an angle of 90° to the incident beam, after passing through a Glan–Thompson prism to remove any depolarized component. Bursts of light, if caused by dust straying into the beam, were detected by a photon counter (Lindmark Innovation AB, Sweden) and usually by the correlator's overflow counter as well, although this was less reliable. The computer was programmed to perform cumulant analysis; in our implementation of the method a polynomial in τ was fitted to the natural logarithm of the normalized photocurrent auto-correlation function (cf. Eqs (*4*), (*5*)). The algorithm uses the Gauss–Jordan elimination to calculate the cumulants and provide estimates of their standard errors. A plot of residuals was also made, which quickly revealed any systematic departure of the generated curve from the experimental data. The baseline, B, may be calculated from the average photon counting rate, or derived from the average value of channels representing long times — in our case, 8 channels delayed by 64 sample times. The two values should coincide in a correct experiment but differ if slowly decaying processes are occurring in the solution. In particular, dust gives rise to components of the function with long correlation times, both through heterodyning and through its slow motions.

RESULTS AND DISCUSSION

The dependence of the calculated reduced cumulant terms R, Q, S (Eq. (*6*)) on the width of molar mass distribution, expressed as $M_w/M_n = 1{\cdot}1 - 11{\cdot}0$, is shown in Fig. 1 for both the Schulz–Zimm and Pearson distributions for toluene as the solvent[1], $\alpha = 0{\cdot}57$. With α 0·5 and α 0·67 we get an analogous picture with values of R, Q, S lower and higher, respectively. (Tables of calculated values are available on request.) Bearing in mind that the direct least squares polynomial fitting procedure, suggested as the best route[12], is never reliable beyond the 4th degree, we can judge the interval of polydispersities where higher terms may be omitted from Fig. 1. We can see that the Q term becomes larger than R above $M_w/M_n = 5$ for the Schulz–Zimm distribution and for the Pearson curve all three R, Q, S approach unity at the same point. Thus, more than four terms are necessary to describe the polydispersity induced deviation of the experimental data from a single exponential fit for values of $\bar{\gamma}\tau \geqq 1$, i.e., with channel numbers $j \geqq 28$ for $\Delta t = 2 \,.\, 10^{-6}$ in our experiments. We believe that the polydispersity interval for a reliable use of the cumulant fit does not go beyond $M_w/M_n \sim 3$; accordingly, the other calculation procedures should be used[15] above this value as well as for bimodal samples. A magnified part of Fig. 1 in this polydispersity interval, useful for a comparison of the R, Q terms with calculated K_2, K_3 values is shown in Fig. 2. The generally accepted view that a detailed shape of the distribution function is not essential in the case of low polydispersities holds also in the case of QELSS below $M_w/M_n < 1{\cdot}3$. On the contrary, the use of the second cumulant (as K_2/K_1^2) has little meaning for, say, $M_w/M_n \sim 2$: We get the distribution width doubled for Schulz–Zimm in comparison to Pearson for the same value of the second cumulant. Let us note that there is one more difference in the QELSS behaviour of asymmetrical distribution. The more asymmetrical Pearson distribution gives much lower Q, S terms in comparison to the Schulz–Zimm for small polydispersities. The behaviour of the R term up to $M_w/M_n = 3$ for the three α values used is shown in Fig. 3. This figure is used to find the initial guess of polydispersity necessary for the determination of $(\bar{\gamma}\tau)_{\max}$ according to conditions (*14*), (*15*) if no prior knowledge of M_w/M_n is available. We also see from Fig. 3 that solvents with low values of α close to the Θ-solvent should be preferred, if high precision of D_z (z-average) is required.

The calculated conditions for the use of the single exponential approximation according to Eq. (*14*) and of the second cumulant approximation (Eq. (*15*)) are plotted in Figs 4, 5, respectively, for both distributions and the α values chosen. In practice, the second cumulant fit (to prevent overfitting) is first applied to all experimental points to get an initial estimate of M_w/M_n (Fig. 3) and D_z. The $(\bar{\gamma}\tau)_{\max}$

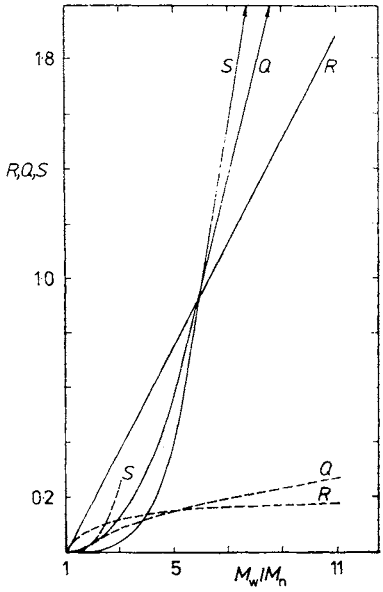

Fig. 1

The dependence of reduced cumulant terms R, Q, S on the width of molar mass distribution for the Schulz–Zimm (broken lines) and Pearson (solid lines) distributions (α = 0·57)

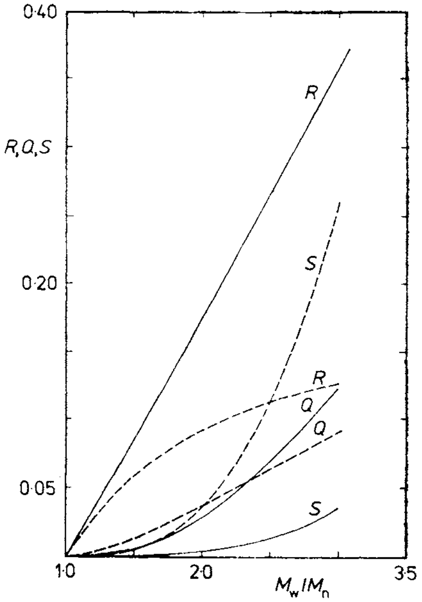

Fig. 2

The dependence of reduced cumulant term R, Q, S in the interval of M_w/M_n 1—3 for the Schulz–Zimm (broken lines) and Pearson (solid lines) distributions (a = 0·57)

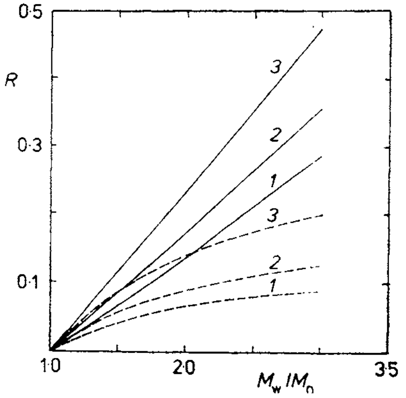

Fig. 3

The dependence of reduced cumulant term R on the width of molar mass distribution described by Schulz–Zimm (broken lines) and Pearson (solid lines) functions for different α values: *1* 0·50; *2* 0·57; *3* 0·67 values are then found from Figs 4, 5 and used in the final calculation. It is clear that some knowledge about the sample is required[15], but this is usually fulfilled to some degree in cases where one is interested in a precise determination of $M_{s,D}$ for calibration and/or other standardization purposes.

Another important problem is the choice of the B value in Eq. (*4*). We do not believe, in agreement with ref.[16], that the use of a forced fitting with an adjustable baseline is a relevant procedure. In an ideal experiment the statistical baseline (calculated by the correlator) B_1 is certainly justified and, if the sampling time is correctly chosen, it should coincide with the last eight delayed channel value B_2. Thus, the difference between B_1 and B_2, if any, should be used to check the deviation of the calculated D_z values, and this again should not exceed 2%.

Five sets of experiments were measured, three in the region of very low concentrations and two at a concentration of about 1 wt. %.

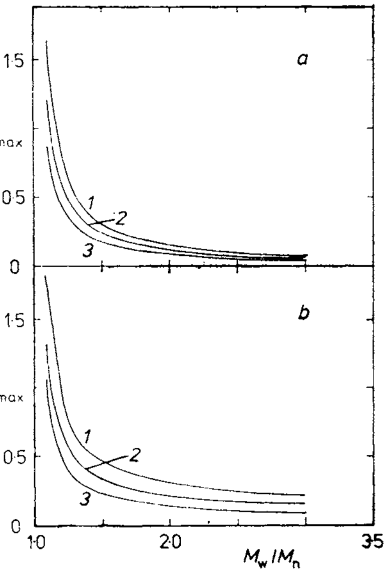

FIG. 4

The dependence of calculated values of $(\bar{\gamma}\tau)_{max}$ on M_w/M_n for the 1st order (single exponential) approximation. Molar mass distributions according to Pearson (*a*) and Schulz–Zimm (*b*). Values of α: *1* 0·5, *2* 0·57, *3* 0·67

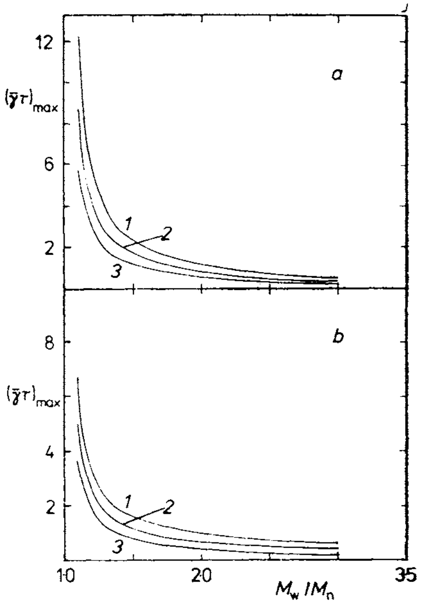

FIG. 5

The dependence of calculated values of $(\bar{\gamma}\tau)_{max}$ on M_w/M_n for the 2nd order cumulant approximation. Molar mass distributions according to Pearson (*a*) and Schulz–Zimm (*b*). Values of α: *1* 0·5, *2* 0·57, *3* 0·67

Three different sampling times, $\Delta\tau$, were used and the maximum channel numbers, $j_{\max}$, were calculated from

$$j_{\max} = \frac{(\bar{\gamma}\tau)_{\max}}{K^2\,\mathrm{D}\Delta\tau} \tag{16}$$

using the $(\bar{\gamma}\tau)_{\max}$ values from Figs 4, 5, taking a reasonable value of $M_w/M_n = 1{\cdot}1$ for the sample used, and assuming the classical diffusion coefficient[1] to be valid. We get $j_{\max}$ 53, 21 and 10 for $\Delta t \,.\, 10^6$ equal to 2, 5 and 10, respectively, as the condition for the 1st order fit whilst, in the case of the 2nd order approximation for the same $\Delta\tau \,.\, 10^6$ $j_{\max}$ equal to 197, 79 and 39, respectively. Both B_1 and B_2 baselines were used.

The reproducibility of individual experiments is seen from summarized results, evaluated for the 1st set in Table I. Using B_1 (B_2) all experiments yield values lying within 1·81% (4·92%) and 2·73% (2·93%) for the 1st and 2nd order approximation, respectively. Thus, the precision of the average D_z value can be safely accepted to be within 2%. It is also apparent that the effect of the differences in B in Table I is not significant within the reproducibility of D_z. As predicted, in all experiments we get lower values from the 1st order approximation than with the second cumulant plot, where only a very small error of opposite sign is expected. The average values calculated as in Table I based on the statistical baseline B_1 are presented in Table II

TABLE I

Results of calculation of D_z according to the 1st and 2nd cumulant approximation for set 1. Conditions: c 0·0703 wt. %, average counting rate 107 000 c/s, 23·9°C. Symbols are defined in the text (D_z values in cm^2 s^{-1})

		1st order		2nd order			
				B_1		B_2	
$\Delta\tau$ 10^6 s	ΔB %	B_1 $D_z \,.\, 10^7$	B_2 $D_z \,.\, 10^7$	$D_z \,.\, 10^7$	K_2/K_1^2	$D_z \,.\, 10^7$	K_2/K_1^2
5	0·012	1·868	1·843	1·902	0·02	1·986	0·10
10	0·004	1·875	1·838	1·924	0·03	2·028	0·13
2	0·042	1·892	1·910	1·955	0·05	1·987	0·07
2	0·045	1·878	1·897	1·933	0·05	1·983	0·08
2	0·099	1·858	1·900	1·953	0·07	1·990	0·08
2	0·101	1·887	1·932	1·947	0·05	1·969	0·04
2	0·038	1·886	1·903	1·946	0·05	1·993	0·08
Average	*0·048*	*1·878*	*1·910*	*1·933*	*0·046*	*1·991*	*0·083*

for all measured sets. Analogous calculations based on B_2 gave results not significantly different within the reproducibility, exactly as in the case of the 1st set. It follows from Table II that sets 1 – 3 behave in the same way; the scatter of D_z values is only slightly higher at the lowest concentration (set 3), the second cumulant fit gives reasonable values of K_2/K_1^2 (noting that the error estimate of K_2/K_1^2 is usually about ±30%) and the calculated deviation between the 1st and 2nd order agrees well with the theoretical prediction. This comparison also allows an internal check to be made of the autocorrelation curve when a different number of channels is used in both approximations ($\Delta\tau = 5$ and $10 \,.\, 10^{-6}$ in this case). As a general feature of the dilute solutions studied, we can say that a single Lorentzian fit applies within 2 – 4%, and with this precision the polydispersity effect can be neglected. This is not the case of sets 4, 5, however; neither K_2/K_1^2 nor the differences between the 1st and the 2nd order fit correspond to the expected value of 2%. It is not the aim of this paper to study QELSS in solutions close to the semidilute region; let us simply note that $c^* \sim 1{\cdot}5 \text{ g dl}^{-1}$ was determined[18] experimentally for approximately the same M_w in toluene and K_2/K_1^2 is known from the literature[16,19,20] to increase at higher concentrations.

The final values of D_z corrected to 25°C are summarized in Table III together with the average differences in baselines and the M_w/M_n values calculated on the basis of K_2/K_1^2. Jamieson[21] suggested that the baselines should differ by less than 0·1% in a reliable experiment; it follows from our data that the error of D_z resulting from such baseline uncertainty is then safely below 2%.

For a comparison of the classical diffusion data with the QELSS results at the ±2% error level, additional inspection of the classical diffusion is needed. It is

TABLE II

Average results of calculation of D_z according to the 1st and 2nd cumulant approximation based on the statistical baseline: ΔD_z difference of maximum and minimum value, Δ deviation of the 1st order to the 2nd order

Set	c wt. %	1st order $D_z \,.\, 10^7$ cm^2 s^{-1}	1st order ΔD_z %	2nd order $D_z \,.\, 10^7$ cm^2 s^{-1}	2nd order ΔD_z %	2nd order K_2/K_1^2	Δ %
1	0·0703	1·878	1·81	1·933	2·73	0·046	−3·25
2	0·0703	1·914	1·00	1·964	2·95	0·037	−2·54
3	0·0211	1·852	3·40	1·935	9·51	0·052	−4·20
4	1·037	2·911	3·20	3·369	7·83	0·108	−12·80
5	1·037	2·852	6·80	3·355	7·63	0·112	−13·40

desirable to check whether the possible error in the classical D has a positive or a negative sign.

The classical diffusion data[22] are replotted in Fig. 6. The solid line is a polynomial fit to the data: $D(c) = 2{\cdot}0234 + 2{\cdot}1546c - 0{\cdot}293c^2$. Below c 0·5 g dl^{-1} we see that the scatter is slightly higher than ±2%. The value obtained by polynomial extrapolation may be compared with the diffusion coefficient calculated from $D = 3{\cdot}32\,.\,10^{-4}M_{s,D}^{-0.57}$ using $M_{s,D} = 452\,000$ as determined[1] previously. This relation $D \sim M$ is certainly valid and, when used $M_{s,D}$, "averages" the individual errors of s, D determinations. We get $D = 1{\cdot}98\,.\,10^{-7}$ indicating (see Fig. 6) that the value calculated by the polynomial fit is overestimated by about 2%. Values of D_z calculated for dilute solutions then agree quite well with the results of classical diffusion within the required accuracy. Let us note at this point that there is a difference in the definition of the average D values "seen" by both techniques; we will show in the

TABLE III

Summary of experimental data: c calculated from mass concentrations using density data[17]; D_z corrected to 25°C, average values for both baselines; M_w/M_n apparent polydispersity index from Fig. 2 for the Schulz–Zimm distribution

Set	No. of experiments	c g dl^{-1}	ΔB %	$D_z\,.\,10^7$ cm^2 s^{-1}	M_w/M_n
1	7	0·0606	0·048	1·995	1·25
2	4	0·0606	0·028	1·981	1·1
3	6	0·0182	0·017	1·987	1·2
4	5	0·8969	0·088	3·214	1·5
5	6	0·8969	0·076	3·294	1·33

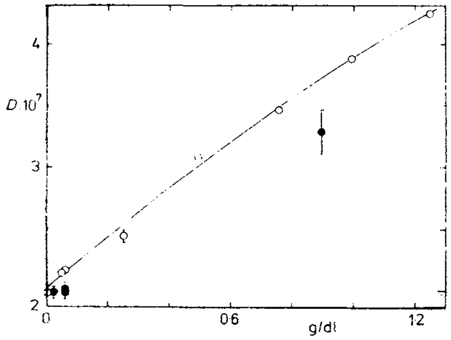

FIG. 6

Comparison of QELSS diffusion coefficient with the value obtained by classical diffusion: ● QELSS D_z, ○ classical diffusion data[22], ◐ calculated from $D \propto M$ relation[1] next paper[23] that D_z should be lower than the D-average from the Bryngdahl interferometer. The difference should not exceed 1% for $M_w/M_n \sim 1{\cdot}1$.

The D_z values for the higher concentration do not agree with the results of classical diffusion; the error bar indicates the maximum scatter of individual determinations. There is no way to force the data to obtain D high enough; a forced baseline value (selected to decrease apparent polydispersity) still results in lower D and the 1st order fit still cannot be used. Thus, a difference of about 13% between gradient and QELSS diffusion must be accepted at this concentration, apart from other differences discussed above.

The same five sets were measured also with unfiltered solutions. Although we do not present all data here, we believe that some practical consequences may be of interest. The data collected in dilute solutions showed a higher scatter, but should still be used to get reliable values of D_z, if some experiments with too different baselines and/or a negative second cumulant were omitted. Then value of K_2/K_1^2 and differences between 1st and 2nd order fit were also reliable. In more concentrated solutions it was almost impossible to collect data with sufficiently small baseline difference. Moreover, this difference increased with the duration of the experiment. The difference in the baselines apparently reflects the presence of dust (cf. ΔB in filtered solutions, sets 4, 5, Table III); the decrease in this difference with dilution in non-filtered solutions can be simply explained by the "dilution of dust" to a level where the influence of the rest of dust can be neglected, provided the duration of the experiment is not excessive.

In conclusion, we state that the simplest way to obtain comparable diffusion constants from QELSS and classical diffusion within the requested accuracy is to adjust the sample concentration to a value where the effect of the concentration dependence of D can be neglected (in the same way as in our classical work[1]). Then no extrapolation to zero concentration is necessary, and also the influence of dust decreases substantially.

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