

Retention effects in thermal field-flow fractionation^a

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ABSTRACT

While the basis of the thermal field-flow fractionation (TFFF) method presumably is thermal diffusion, predicting retention for different polymer types in different solvents is generally not possible. To clarify the TFFF retention mechanism, we developed a data base for a range of experimental conditions. Retention is strongly a function of polymer and carrier solvent types, and is proportional to solvent viscosity. Studies suggest that polymer retention significantly increases when using a mixed carrier solvent that has a higher concentration of "poor" solvent relative to "good" solvent. Although no apparent correlation was found between retention and various physical properties of the system, more effective practical TFFF experiments can be done because of these studies.

INTRODUCTION

Quantitative molecular-weight distributions (MWD) of a wide range of organic-soluble¹⁻⁶ and certain water-soluble⁷ polymers are possible with the high-resolution separation method called thermal field-flow fractionation (TFFF). Because of higher resolution, TFFF generally has higher molecular-weight accuracy than the widely used size-exclusion (or gel permeation) method⁶. Especially attractive is the ability of TFFF to characterize fragile polymers of very high molecular weight^{4,8}.

TFFF separations typically use a single carrier liquid by applying a large thermal gradient across a very thin channel formed from two highly polished, parallel plates. Because of this temperature difference, sample components are pushed against one wall (the accumulation wall). Higher-molecular-weight (MW) materials are forced closer to the wall compared to lower-MW components. The essentially laminar flow profile created within the thin channel creates a very steep velocity profile near the walls. Higher-MW components that are closer to the accumulation wall are swept

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down the channel by slower flow streams near the wall. They lag behind and elute after lower-MW components. Thus, retention differences result from the different redistribution of solutes from fast- to slow-moving flow streams within the laminar flow profile.

Previous studies have shown that TFFF retention is dependent on the chemical composition and molecular size of the polymer investigated^{1,2,6,9}. TFFF retention allows the determination of thermal diffusion coefficients in different solvents¹⁰. Also reported is the effect of temperature on thermal diffusion, to provide a better basis for choosing optimum conditions for TFFF experiments¹¹. Still, even with these studies, the *a priori* prediction of TFFF retention characteristics of new or previously uncharacterized polymers is not possible.

The purpose of this study was to develop data for a range of polymer types and solvents. The expectation was that this information might lead to a better fundamental understanding of thermal diffusion and the TFFF retention process. Retention relationships were determined with the time-delay, exponential-decay programming method. After an initial period of constant temperature difference between the two channel faces, the temperature difference decreases exponentially during the separation. With this method, TFFF retention follows a simple log MW vs. solute retention time relationship that provides a convenient way to organize and analyze data.

The results of this study still do not allow quantitative predictability of polymer retention. Nevertheless, improved techniques for carrying out TFFF experiments resulted, with a clearer definition of some strengths and limits of TFFF.

EXPERIMENTAL

Equipment

The apparatus was essentially the same as that previously described⁵, with one exception. A Model 150 computer equipped with Model 5088-2057 and Model 61012A interface components (Hewlett-Packard, Palo Alto, CA, U.S.A.) remotely controlled the setpoint of the temperature controllers during programming. Instrument control was by in-house-developed Basic software. A Hewlett-Packard Model 18652A analog-to-digital converter digitized the outputs from a UV absorption detector (Model 783, Kratos Analytical Instruments, Ramsey, NJ, U.S.A.) and a refractive index detector (Optilab Model 5902, Tecator Instruments, Herndon, VA, U.S.A.). Signals were processed with a Hewlett-Packard Model 3357 LAS system equipped with in-house-developed software⁶. Time-delayed exponential-decay (TDE)-TFFF calibration data were log-linear regressed and plotted with a Vectra Model 45 personal computer (Hewlett-Packard), using Graphwriter II software (Lotus Development Corporation, Cambridge, MA, U.S.A.) and a Model 7475 plotter (Hewlett-Packard).

Reagents

HPLC-grade solvents were from Fisher Scientific (Fair Lawn, NJ, U.S.A.) or American Burdick and Jackson (Muskegon, MI, U.S.A.). Polymer MW standards were from Polymer Labs. (Amherst, MA, U.S.A.). All other materials were from DuPont.

THEORY OF RETENTION IN TIME-DELAYED EXPONENTIAL-DECAY TFFF

TDE-TFFF permits the separation of a wide range of polymer molecular weights with a single experiment in a convenient time. With this approach, the temperature difference between the plates decreases exponentially during the run⁵. Temperature programming speeds up the elution of higher MW components, reduces analysis time, and improves the detection of later-eluting species¹². Previous studies have shown some advantages of using time-delayed exponential-decay temperature programming in thermal field flow fractionation⁵⁻⁷. With this approach, uniform resolution and MW accuracy occurs over a wide dynamic MW range in a practical analysis time. A similar form of force field programming is used in sedimentation FFF^{13,14}.

In the TDE-TFFF experiment, the separation begins with an initial temperature difference between the two plates, $(\Delta T)_0$, that is held constant for a time τ . Then, ΔT decreases exponentially also with a time-constant τ . With this approach, retention times of well-retained solutes can be described by^{5,6}

$$t_R = \alpha\tau(\ln M) + \tau \ln[eV_0(\Delta T)_0/6F\beta\tau] \quad (1)$$

When $\beta = b/D_T$, where b is a constant and D_T is the thermal diffusion coefficient, then

$$t_R = \alpha\tau(\ln M) + \tau \ln\{[D_T/b][eV_0(\Delta T)_0/6F\tau]\} \quad (2)$$

where, t_R is the retention time of the eluted component; $\alpha \approx 0.6$ for random-coil polymer conformations; τ is the time-delay, exponential-decay time constant; M is the molecular weight; F is the flow-rate; V_0 is the volume of the separating channel; and $(\Delta T)_0$ is the initial temperature difference between the hot and cold blocks. Eqns. 1 and 2 suggest that a plot of retention time t_R vs. $\ln M$ is a straight-line relationship with a slope set by the time-delay/decay constant τ . Other operating parameters such as the flow-rate F and the initial temperature difference $(\Delta T)_0$ determines the intercept of this plot. The value of τ primarily controls the elution of sample components, with smaller influence by the other operating parameters.

The relationship in eqn. 2 provides a convenient and accurate method for calibrating and measuring the MWD of polymers over a wide MW range. The TDE-TFFF method permits convenient data handling for quantitative MWD analyses⁶. Accurate peak-position calibrations follow from the simple $\log MW$ vs. retention time relationship. Also, the valuable single- or two-broad-standard calibration procedures¹⁵ are readily used. Further, eqn. 2 predicts exact changes in the elution of components with variations in the various operating parameters such as τ , F and $(\Delta T)_0$. Nomographs simplify this calculation¹⁶.

The relationships in eqn. 2 are convenient to obtain certain physical data on polymers of interest. For example, eqn. 1 suggests that the slope of the log-linear TDE-TFFF calibration curve can be used to find the α value for a polymer in a particular solvent system. Since the slope = $1/\alpha\tau$, the α value is easily calculated, as indicated later in this paper. This α value is related to the well-known Mark-Houwink a constant. This constant is, in turn, related to polymer intrinsic viscosity by the expression¹⁵

$$[\eta] = KM^a \quad (3)$$

where $[\eta]$ is the polymer intrinsic viscosity in a particular solvent, K is a constant for the system, M is the polymer molecular weight, and a is the Mark-Houwink constant describing polymer conformation¹⁵. Now,

$$R_g \approx ([\eta]M)^{1/3} \quad (4)$$

and

$$R_g \approx M^{(1+a)/3} \quad (5)$$

where, R_g is the radius of gyration of the polymer molecule in the solvent. Because of these correlations,

$$D \approx 1/R_g \approx M^{-[(1+a)/3]} \quad (6)$$

and

$$D = bM^{-\alpha} \quad (7)$$

It can be calculated that

$$\alpha = [(1 + a)/3] \quad (8a)$$

and

$$a = (3\alpha - 1) \quad (8b)$$

where, D is the familiar solute-solvent diffusion coefficient, and the constant b is proportional to $1/\eta_{\text{solvent}}$. Therefore, the well-recognized Mark-Houwink a constant directly relates to the α value of eqns. 1 and 2. The α value also can be used to describe the conformational characteristics of a polymer molecule in a particular solvent.

RESULTS AND DISCUSSION

Effect of polymer type

Previous studies show that retention in TFFF varies with both polymer type and the solvent used in the separation^{1,2,6,16}. With TDE-TFFF this effect is shown by the somewhat constant slopes of the log-linear calibration plots for different random-coil polymers and for different organic solvents. However, the intercept of the calibration plots can vary^{5,6,16}. Fig. 1 illustrates these effects for four different sets of polymer standards fractionated with toluene as the mobile phase. All calibration curves were linear with essentially the same slopes. The same general effect occurs for the same polymer fractionated in different solvents, as discussed below.

Effect of carrier solvent type

We initially proposed that solvent effects on polymer solubility might be

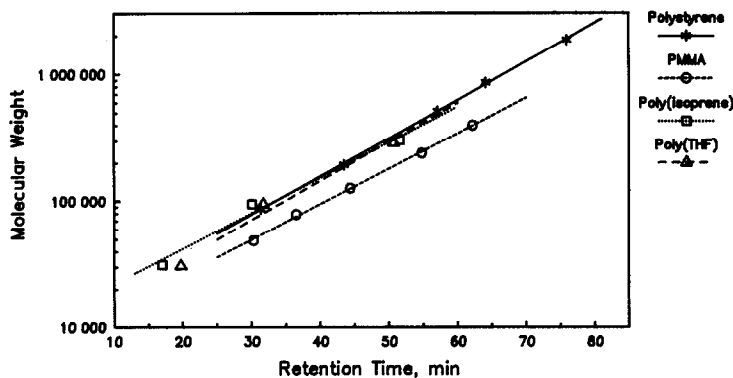


Fig. 1. TDE-TFFF calibration plots for several polymers. Carrier, toluene; flow-rate, 0.15 ml/min; channel, 132 μm ; Initial hot block temperature, 90°C; cold block temperature, 20°C; exponential time delay/constant, $\tau = 25.0$ min; sample, 50 or 100 μl of 1 mg/ml in carrier solvent; PMMA = poly(methyl methacrylate); poly(THF) = poly(tetrahydrofuran).

a central feature of TFFF retention phenomena. Our approach in defining this supposition was to develop a data base for polymer-solvent systems, in the hope that key elements regarding TFFF retention and thermal diffusion coefficients might be revealed. Fig. 2 illustrates the effect of solvent type on retention for polystyrene standards fractionated with a variety of solvents. Again, the slopes of the TDE-TFFF calibration plots are similar, suggesting approximate constancy of polymer conformation in these solvents. The greatest difference in retention was between toluene and chloroform, with the former showing the highest retention of these solvents. Fig. 3 shows similar data developed for the more polar polymer, poly(methyl methacrylate) (PMMA). For this system, greatest retention was in toluene, least in *N,N'*-dimethylformamide (DMF). The slope of the TDE-TFFF calibration plot appears somewhat dependent on the type of polymer and solvent. The slope also is characterized by the value of α from eqn. 1, as discussed later.

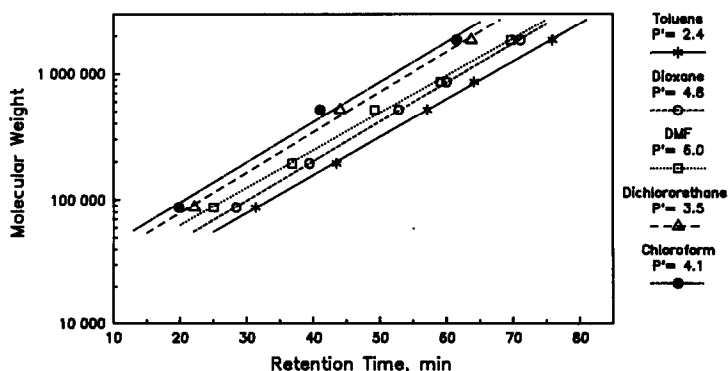


Fig. 2. Effect of carrier solvent on polystyrene calibration plots, Solute, polystyrene; other conditions as in Fig. 1; DMF = *N,N'*-dimethylformamide.

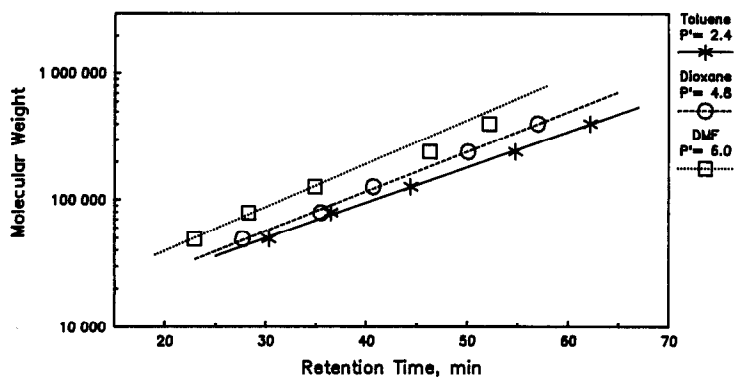


Fig. 3. Effect of carrier solvent on poly(methyl methacrylate) calibration plots. Solute, poly(methyl methacrylate standards); other conditions as in Fig. 1.

What is the retention change that occurs when a polymer is dissolved in solvents of highly different characteristics? The answer to this question depends on the particular solvent and the polymer of interest. Still, insight can be gained by observing the retention characteristics of a polymer that is soluble in both organic and aqueous systems. We have previously shown that retention occurs for certain water-soluble polymers, permitting their characterization by TFFF⁷. For example, poly(ethyleneoxide) shows TFFF retention in both organic and aqueous media. This polymer provides an interesting test case for studying retention changes as a function of mobile phase effects. Fig. 4 shows the TDE-TFFF calibration plots for poly(ethyleneoxide) standards in water and chloroform. In this instance, similar calibration plots occur in

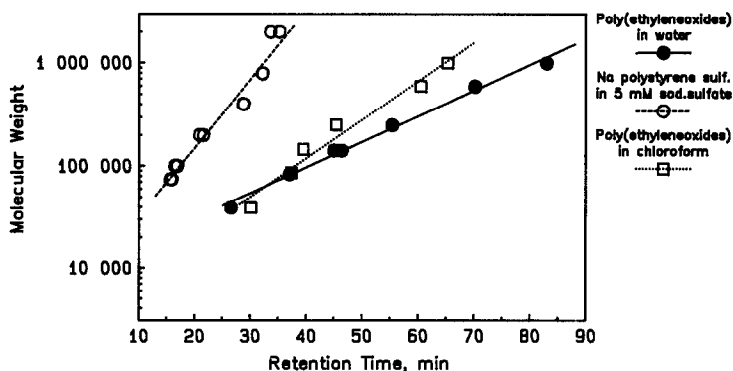


Fig. 4. Comparison of calibration plots for aqueous- and organic-soluble polymers. Poly(ethyleneoxide) standards: in water: 254 μ m channel; flow-rate, 0.30 ml/min; 25 μ l, 2 mg/ml each; in chloroform; 136 μ m channel; flow-rate, 0.15 ml/min; 50 μ l, 1 mg/ml each; sodium polystyrene sulfonate standards: 254 μ m channel; flow-rate, 0.30 ml/min; 25 μ l, 1 mg/ml each in 5 mM Tris-sodium sulfate, pH 7.5. Other conditions as in Fig. 1.

both solvents; retention also is similar in the very different solvents, water and chloroform.

However, strikingly different polymer retention differences occur for other water-soluble polymers. For example, sodium polystyrene sulfonate exhibits unusual retention characteristics in a very dilute salt solution⁷. The steep calibration curve found for this ionic polymer (Fig. 4), whose conformation should be rodlike in low ionic-strength media¹⁷, is decidedly uncharacteristic, as discussed below.

Data in this report and elsewhere^{9,18} support the contention that the mass of the solute and its normal diffusion characteristics primarily govern the MW dependency of TFFF retention. An MW increase decreases the diffusion coefficient and increases retention because normal diffusion cannot overcome the force pushing molecules to the accumulation wall. The result is that larger molecules remain in slower flow streams near the wall and elute later. One can speculate that, for this effect to occur, the force created by thermal diffusion to push components to the accumulation wall must be largely independent of solute mass (see also refs. 9, 10 and 18).

On the other hand, the data for sodium polystyrene sulfonate in Fig. 4 suggest that this picture is not so simple for some systems. At the ionic strength used, this compound extends to a rod-like conformation, with a resulting smaller diffusion coefficient than for non-ionic, random-coil polymers (*e.g.*, poly(ethyleneoxide) of the same MW in organic solvents. Based on this, one would expect that the poorer diffusion of the extended polymer would result in greater retention, not less retention, as in Fig. 4 (also, see discussion below). Clearly, the mechanism of TFFF retention is complex for some systems and not satisfactorily understood.

Effect of solvent strength

The variability of polymer TFFF retention in different solvents previously has been documented^{1,2,6,9}. Yet, explanations for the physical reasons for this variability are lacking, other than a change in solvent causes a change in thermal diffusion coefficients^{1,10,19}. Unfortunately, the molecular basis for thermal diffusion still is unknown. We need the means to calculate thermal diffusion coefficients in order to predict TFFF retention for new systems.

Data in Fig. 2 strongly suggest that solvent strength is not the main feature that controls solute retention in TFFF. Solvent strength P' values calculated according to Snyder²⁰ and shown on Fig. 2 show no relationship to retention characteristics. The weakest solvent, toluene, with a P' value of 2.4 shows highest retention. But, the strongest solvent, DMF, with a P' value of 6.0, causes intermediate retention. Dichloroethane and chloroform with intermediate P' values of 3.5 and 4.1, respectively, produce similar weaker retention.

The strength (P' values) of mixed "good" and "poor" solvents for the polymer also do not correlate with TDE-TFFF retention, as illustrated by the data in Fig. 5. Here, plots obtained for three "good" solvents diluted with an equal volume of heptane, a "poor" solvent, still show the general relationship of Fig. 2 for undiluted solvents. Mixed dioxane still shows the highest retention, and mixed chloroform the lowest retention, as in Fig. 2.

It appears, therefore, that solvent strength *per se* does not directly correlate with TFFF retention.

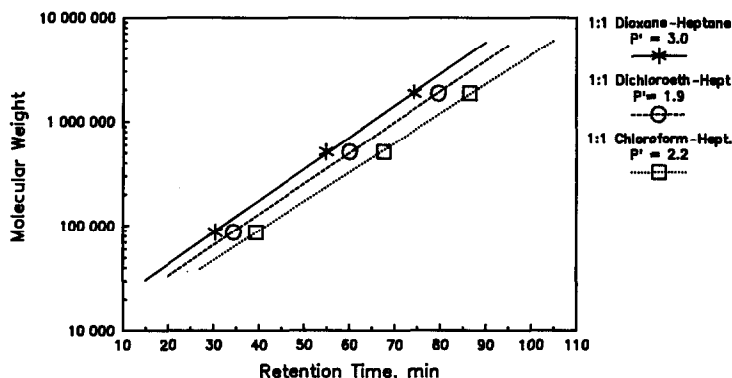


Fig. 5. Effect of carrier solvent strength on retention (different solvent types). Dioxane, dichloroethane, chloroform carrier solvents diluted with heptane. Conditions as in Fig. 1, except: polystyrene standards, 25 μ l, 1 mg/ml; initial hot block temperature, 90°C; initial cold block temperature, 20°C; final temperature of both blocks after programming, 35°C. Solvent strength P' values calculated according to Snyder²⁰.

Effect of solvent viscosity

Carrier solvent viscosity has a significant effect on TDE-TFFF retention. Fig. 6 illustrates this for two polystyrene MW standards fractionated in four pure solvents, dioxane ($\eta = 1.2$), cyclohexane ($\eta = 0.90$), dichloroethane ($\eta = 0.78$), and chloroform ($\eta = 0.53$). A straightline relationship appears for these systems, with a smaller slope for the plot of the higher-MW polymer. TFFF theory anticipates this trend, since retention is dependent on the relative strength of the interaction between the applied thermal force field and the opposing diffusion of the solute¹. As suggested by eqns. 2 and 7, retention time, t_R , in TDE-TFFF is proportional to $\ln(1/D)$. Therefore, as carrier solvent viscosity increases, the diffusion coefficient decreases for the polymer. Molecules with poorer diffusion cannot overcome the thermal force field. Retention then increases because polymer molecules are forced closer to the accumulation wall. Slower flow streams intercept molecules close to the accumulation wall so that they

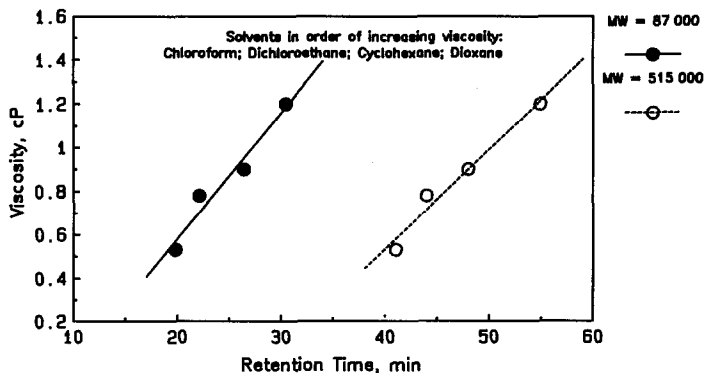


Fig. 6. Effect of carrier solvent viscosity on polystyrene retention (different solvent types). Conditions as in Fig. 5, with different dilutions with hexane; solutes, 87 000 and 515 000 MW polystyrene standards.

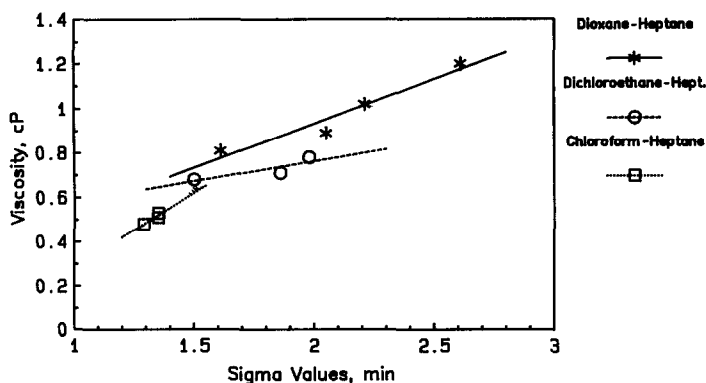


Fig. 7. Effect of solvent viscosity on band width. Solute, 87 000 MW polystyrene standard; sigma values = band standard deviation; conditions as in Fig. 5.

elute later. These results strongly suggest that thermal diffusion is not viscosity dependent, and that viscosity only affects conventional diffusion, which, in turn, strongly influences TFFF retention.

Note for the data in Fig. 5 that we used a "dual-mode" form of exponential temperature programming which was slightly different from previous experiments. To obtain these data the initial ΔT was 70°C , as usual, with the initial hot and cold block temperatures of 90 and 20°C , respectively. However, the cold block temperature was *not* held constant during the experiments. Using appropriate computer software, the temperature of *both* the hot and cold blocks was programmed to a final temperature of 35°C (hot block temperature decreased, cold block temperature increased). In this manner, the ΔT again was exponentially decreased with the desired τ value.

The results obtained with "dual-mode" exponential time delay/decay programming are similar to those of the traditional mode of holding the cold block temperature constant. But, sometimes there may be a practical advantage in dual-mode programming, since demands on precise temperature control are relaxed somewhat in experiments with small τ values (fast temperature decrease).

The viscosity of the carrier solvent also affects the standard deviation (width) of the solute peak, as might be predicted²¹. Fig. 7 shows that the peak width (in sigma values, min, where sigma is the standard deviation of the TDE-TFFF band) increases with increasing viscosity. This effect is because of poorer diffusion and increased resistance to mass transfer. Least change in sigma values occurs with the chloroform-heptane mixtures, since the viscosities of these two solvents are similar.

Effect of polymer solubility

All the solvents used to obtain the data in Fig. 2 are "good" solvents for polystyrene²². This leads to the question: Does the level of polymer solubility in the carrier solvent influence thermal diffusion and resultant TFFF retention? In an attempt to answer this question, we measured TDE-TFFF retention for polystyrene with "good" carrier solvents mixed with varying amounts of heptane, a "poor" solvent (actually, a non-solvent for polystyrene). Fig. 8 shows retention results from mixing heptane separately with dioxane. Increasing the concentration of the heptane diluent

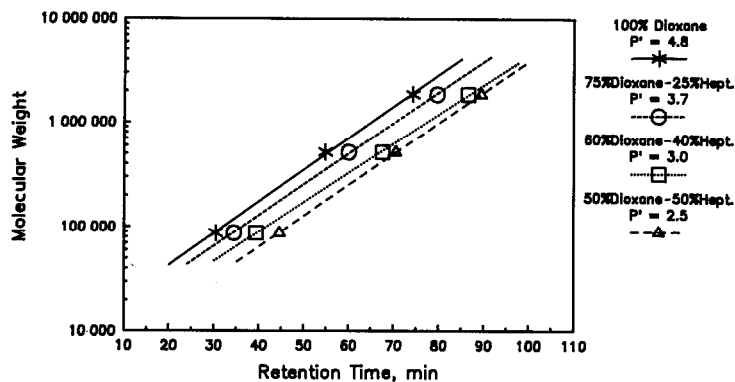


Fig. 8. Effect of solvent composition on retention. Solutes, polystyrene standards; dioxane diluted with heptane; conditions as in Fig. 1.

significantly increased retention. But, the slopes of the calibration curves remained essentially constant with increasing heptane content (potential decrease in polymer solubility).

Fig. 9 illustrates the increase in retention by diluting several "good" solvents with a "poor" solvent. Significant increases in polymer retention time occur as the volume fraction of heptane increases. Solubility of the polymer in the mixed solvent ultimately limits this effect. In the systems of Fig. 9, volume fractions of heptane exceeding about 0.5 precipitated the polymer and prevented TFFF characterization.

We believe that the results in Figs. 8 and 9 might be explained by changes in the thermal diffusion of the polymer, rather than by solvent power effects on molecular size and the normal diffusion coefficient. The reasoning is that the level of retention changes seen in Figs. 8 and 9 are much more than could be anticipated by changes in polymer diffusion due to solvation conformational changes. Increased diffusion is predicted for a decrease in polymer hydrodynamic radius due to less solvation

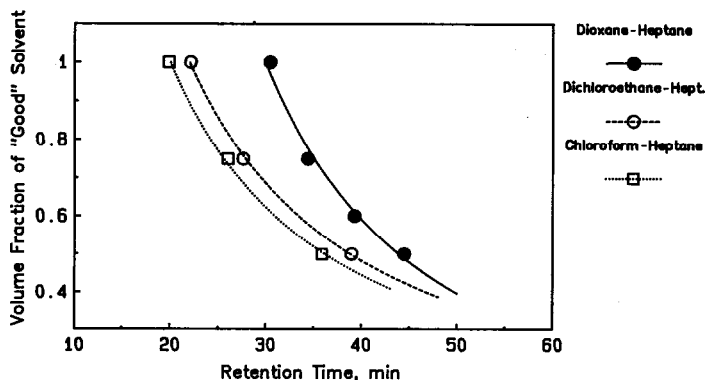


Fig. 9. Increased retention with higher "poor" solvent concentrations. Dioxane, dichloroethane and chloroform carrier solvents diluted with heptane; solute, 87 000 MW polystyrene standard; conditions as in Fig. 1.

resulting from diluting "good" solvents with a "poor" solvent. Such an increase in diffusion would reduce retention and, therefore, would not account for the retention increases observed in experiments with higher "poor" solvent concentrations.

The results from above study may not significantly contribute to our quantitative understanding of thermal diffusion. However, they do furnish useful guidelines for increasing the scope of TFFF applicability for solving real problems in determining the molecular weight distribution of polymers. For example, the data of Figs. 8 and 9 suggest that a significant increase in polymer retention occurs by using mobile phases that result in very high thermal diffusion, here, by diluting "good" solvents with "poor" solvents for the polymer. This is a practical approach when additional retention is needed, especially if the V_0 channel "dead" volume peak interferes with low-MW components. Fig. 10 shows the significantly increased retention afforded by this technique for a mixture of polystyrene standards. Note the significant increase in resolution between the channel dead volume V_0 peak and the first-eluting 87 000 MW polystyrene standard peak, as heptane dilutes the carrier.

Effect of temperature difference, ΔT

Maintaining a constant temperature difference ΔT between the channel walls by varying the temperature level affects TFFF retention, as previously noted¹¹. Fig. 11 demonstrates this effect for an 87 000 MW polystyrene standard fractionated in cyclohexane with a ΔT of 30 and 40°C. Here, the polymer is more strongly retained at the lowest temperature range with the hot block at 50 and 60°C, respectively, and the cold block at 20°C (Fig. 11a and e). At the highest temperature levels (Fig. 11d and h), retention slightly decreases. The tailing, broad bands seen in Fig. 11a and e apparently are associated with poor solubility of polystyrene in cyclohexane at lower temperatures.

Correlation of retention with certain physical properties

Limited attempts to correlate TDE-TFFF retention with four pure carrier solvents (toluene, dichloroethane, dioxane and cyclohexane) with other physical properties produce no meaningful results. Fig. 12 shows that TDE-TFFF retention failed to correlate with carrier solvent thermal conductivity, heat capacity, surface

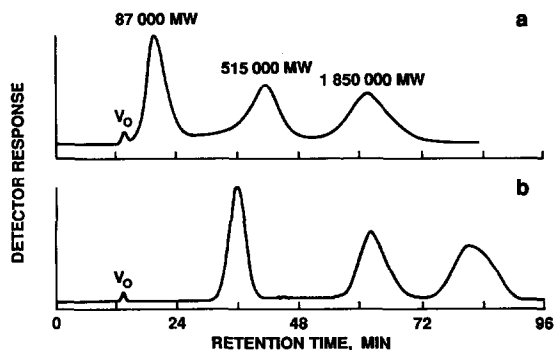


Fig. 10. Retention increase by diluting "good" carrier solvent with "poor" solvent. (a) 100% chloroform carrier; (b) chloroform-heptane (50:50) carrier; polystyrene standards; conditions as in Fig. 1.

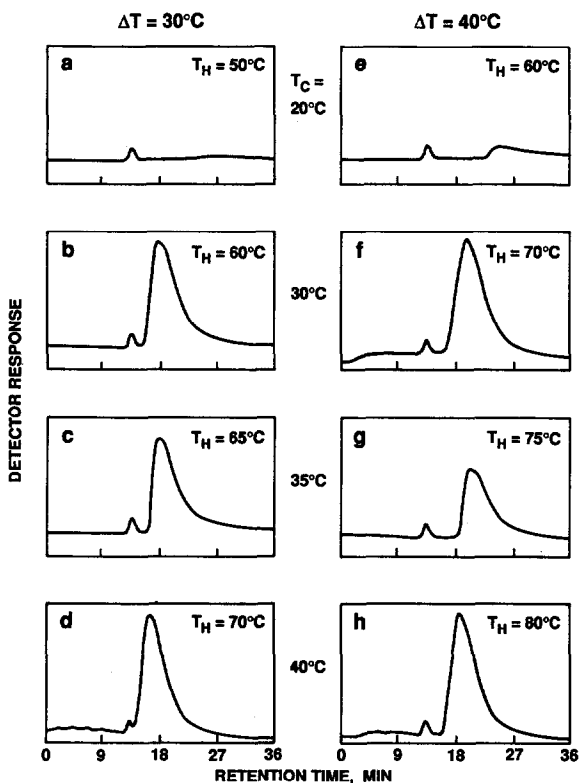


Fig. 11. Effect of level of ΔT on retention. Solute, 87 000 MW polystyrene; constant $\Delta T = 30$ and 40°C ; other conditions as in Fig. 1. T_H = Hot block temperature; T_C = cold block temperature.

tension, heat of formation, or solvent entropy data. While these results suggested no significant correlation with certain physical properties, only a limited data base was used. More complete documentation requires further studies. Another limitation is that the Fig. 12 data on the various physical parameters came from widespread reference sources that may be variable, incorrect or incomplete. It should be stressed, however, that TFFF is a superior method for measuring certain physical constants. Normal solute-solvent diffusion coefficients and thermal diffusion coefficients are readily determined, as described by Giddings and co-workers^{9,10}.

As described above, TDE-TFFF is a convenient method for measuring α values. This value relates to the Mark-Houwink a constant widely used in preparing universal calibrations in size-exclusion chromatography (SEC), as predicted by eqn. 8b. Table I summarizes the α values calculated from the slope of the calibration curves obtained during this study and from calibration curves given in a previous publication¹⁵. These α values also are compared with Mark-Houwink a values from the literature²³ for some polymer-solvent systems. In general, the correlation between a values from these two sources is reasonable, considering that literature a values themselves are quite variable.

Because of experimental limitations, we estimate that the α values reported in

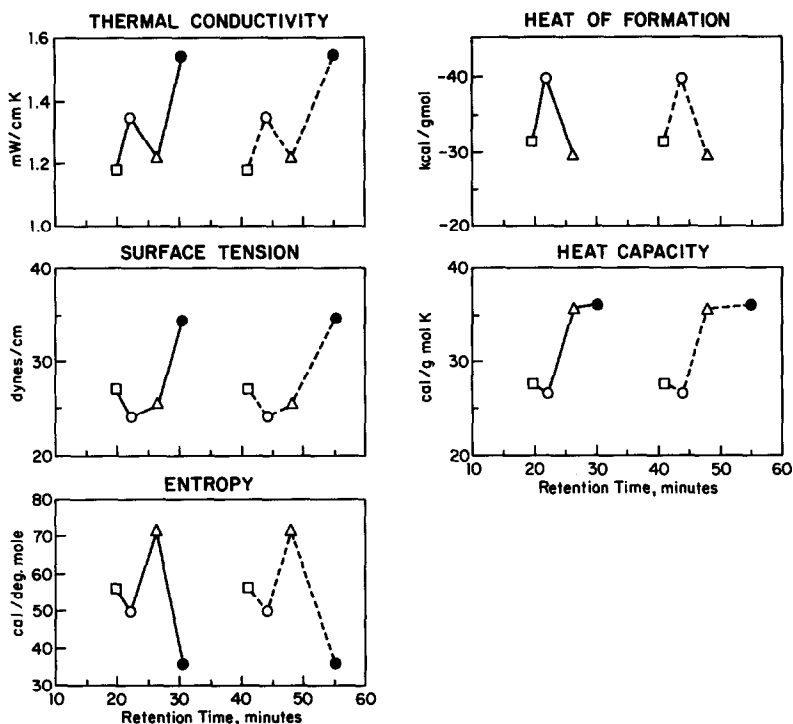


Fig. 12. Effect of physical parameters of carrier solvents on retention. Conditions as in Fig. 5; \square = chloroform; \circ = dichloroethane; \triangle = cyclohexane; \bullet = dioxane; physical data obtained from literature sources, based on 20°C, where appropriate.

Table I probably have a precision of about 0.08 (2σ) for all the polymers studied. (Variations are significantly less for some polymers, particularly those with narrow molecular weight distributions.) Therefore, the precision for calculated a values by this approach is three-fold poorer, or no better than about 0.24, since a is proportional to 3α (eqn. 8b). This level of experimental error makes it difficult to recognize slight changes in the slope of Fig. 8 plots that may result from polymer conformational changes by diluting a "good" solvent with a "poor" solvent.

The exception to correlation of the a values are data for sodium polystyrene sulfonate at the bottom of Table I. The near zero value calculated by TDE-TFFF for a is completely erroneous. This peculiar result is in keeping with the curious, very steep calibration curve for this polymer in Fig. 4. The rod-like structure for this polymer in low ionic-strength aqueous solution would be expected to produce a plot with a slope less than for poly(ethyleneoxide) (Fig. 4) in an aqueous solvent. Rod-like molecules should have a less-steep calibration plot because of the expected strong dependence of the diffusion coefficient on molecular weight. The data in Fig. 4 show just the opposite effect. The sodium polystyrene sulfonate plot is steeper, rather than less steep, than the calibration plot for a random coil molecule. Clearly, TFFF retention in aqueous media sometimes does not follow the same pattern as in organic solvents. This result suggests

TABLE I
COMPARISON OF TDE-TFFF α VALUES WITH MARK-HOUWINK CONSTANT a VALUES

Polymer	Solvent	Measured α	Calculated a value ^a	Literature a value ^b
Polystyrene	Toluene	0.58	0.73	0.79
	DMF	0.59	0.76	—
	Dioxane	0.56, 0.57	0.67, 0.72	0.69
	Dioxane-heptane (75:25)	0.59	0.77	—
	Dioxane-heptane (60:40)	0.62	0.86	—
	Dioxane-heptane (50:50)	0.59	0.76	—
	Dichloroethane	0.54	0.63	0.66
	Dichloroethane-heptane (75:25)	0.59	0.76	—
	Dichloroethane-heptane (50:50)	0.60	0.80	—
	Chloroform	0.55	0.64	0.76
	Chloroform-heptane (75:25)	0.62	0.86	—
	Chloroform-heptane (50:50)	0.60	0.80	—
	Poly(methyl methacrylate)	Toluene	0.62, 0.62	0.86, 0.86
Dioxane		0.55	0.66	—
DMF		0.58	0.75	—
Polyisoprene	Toluene	0.62, 0.54	0.85, 0.61	0.77
	Cyclohexane	0.61	0.82	0.70
Poly(tetrahydrofuran)	Toluene	0.54	0.63	0.78
Poly(ethyleneoxide)	Chloroform	0.58	0.74	0.50
	Water	0.70	1.10	0.78
Sodium polystyrene sulfonate	5 mM Na ₂ SO ₄	0.33	0(?)	0.93

^a From eqn. 8.

^b From ref. 19.

that a different retention TFFF mechanism may dominate for particular polymers in aqueous systems.

Universal calibration curve for TFFF

The plots of Figs. 1-3 conclusively show that the intercept of TDE-TFFF log MW vs. retention time calibration plots vary with polymer and with solvent. This effect is also characteristic of SEC plots of log MW vs. retention volume, when polymer type or solvent varies. Similarly, different plots occur with log $[\eta]$ (intrinsic viscosity) vs. retention volume or retention time plots, respectively, for SEC and TDE-TFFF, as illustrated for the latter in Fig. 13. However, it is well-known in SEC that a plot of log $([\eta] \cdot M)$ vs. retention volume produces a universal calibration plot that is the same for all polymers and solvents¹⁶. This is not so in TDE-TFFF, as illustrated by the data in Fig. 14. Changes in polymer type and solvent produce significant changes in the intercepts of the calibration plots. This effect largely shows the change in polymer thermal diffusion coefficients. Therefore, the consequences of thermal diffusion are not normalized for the polymer hydrodynamic volume by using $([\eta] \cdot M)$ as a plotting parameter.

One means of obtaining an equivalent universal calibration in TDE-TFFF is to include the normal polymer diffusion coefficient and the polymer thermal diffusion

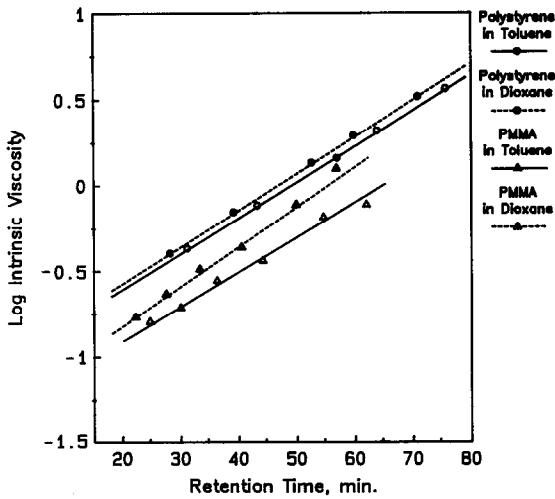


Fig. 13. Log intrinsic viscosity vs. retention time calibration plots. Solutes, narrow polystyrene and poly(methyl methacrylate) standards; conditions as in Fig. 1.

coefficient in the calibration relationship. Fig. 15 shows that this can be done by plotting polymer retention time vs. $\log(S_0 \cdot M)$, where the Soret coefficient²² S_0 is D_T/D (ref. 24). Ref. 10 supplied data for calculating the Soret coefficients in Fig. 15. Unfortunately, such data for polymers are not generally available, so that the approach of Fig. 15 now has limited practical utility.

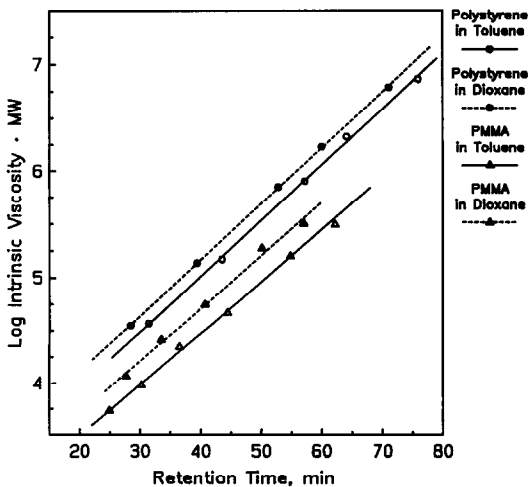


Fig. 14. $\log([\eta] \cdot M)$ vs. retention time calibration plots. Conditions as in Fig. 13.

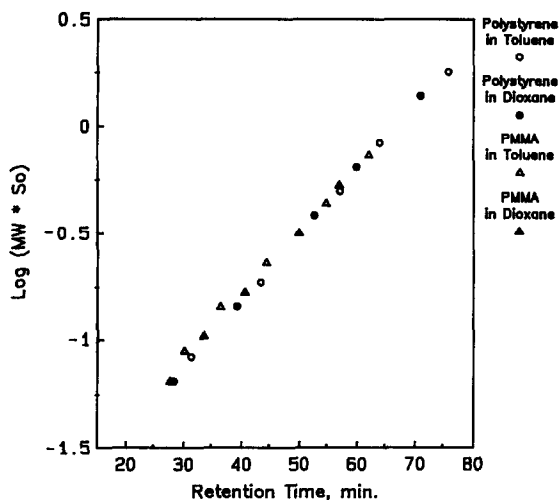


Fig. 15. Universal calibration plot for TDE-TFFF. Conditions as in Fig. 13; S_0 = Soret coefficient, D/D_T .

CONCLUSIONS

While thermal diffusion significantly influences TFFF retention, the mechanism for this effect is not well understood. Predicting TFFF retention is generally not possible for different polymer types in various solvents. We have studied TFFF retention over a range of experimental conditions, using the time-delay exponential-decay operating mode. Here, after an initial period of constant temperature difference between the two faces of the channel, the temperature decreases exponentially during the separation. A plot of log molecular weight vs. polymer retention time shows a linear relationship that provides a convenient way to organize and interpret TFFF retention data. Polymer and carrier solvent types strongly influence TFFF retention. Retention also appears directly proportional to solvent viscosity. Diluting "good" polymer solvents with certain "poor" solvents significantly increases polymer retention, presumably because of large changes in polymer thermal diffusion coefficients. No obvious correlation exists between TFFF retention and carrier solvent thermal conductivity, heat capacity, heat of formation, surface tension and solvent entropy. These studies still do not allow a quantitative prediction of TFFF retention for unknown systems, but they do provide a basis for conducting more effective TFFF experiments. For example, diluting a "good" solvent with a "poor" solvent provides a convenient way of increasing the retention of low molecular weight components. This is especially useful in resolving lightly-retained components from the channel dead volume V_0 peak.

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