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Thermal field flow fractionation of polytetrahydrofuran

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Abstract

The possibility to fractionate polytetrahydrofuran (PTHF) samples effectively with Thermal Field Flow Fractionation has been investigated. Retention measurements of four standards of different molecular mass were performed in eight organic solvents. When methylethylketone or dioxane were used no retention was found. Highest retention for a given molecular mass was obtained with the solvents toluene and ethylbenzene. However, in ethyl acetate the highest separation speed was observed. Plate height measurements showed that both the thermal and molecular diffusion coefficients of the PTHF standards are very high in ethyl acetate. According to theory, this combination should result in a high separation speed. A baseline separation of two PTHF standards with molecular masses of 67 000 and 282 300 could be obtained in 8 minutes when ethyl acetate was used as the solvent.

1. Introduction

Field Flow Fractionation (FFF) was introduced in the 1960's by Giddings as a fractionation method for polymers and particles [1]. With the use of an external field perpendicular to the laminar flow of a carrier liquid in an open channel, macromolecules are concentrated at one of the channel walls. Due to the high velocity gradient near the channel walls retention and separation are accomplished. A number of different external fields or gradients have been employed to obtain retention in FFF. On the basis of field or gradient used, different subtechniques are distinguished in FFF. The major subtechniques are Thermal, Sedimentation [2], Flow [3] and Electrical [4] FFF.

In Thermal Field Flow Fractionation (ThFFF) a large temperature difference is established across the channel thickness. Due to the so-called thermal diffusion effect, the temperature gradient forces the polymer molecules to concentrate near the cold wall. This selective migration is opposed by molecular diffusion and subsequently retention is determined by the ratio of the thermal and molecular diffusion coefficients. This ratio is also expressed as α/T, where α is the Soret coefficient and T is the temperature.

Although it has been studied for a long time [5], thermal diffusion in liquids is still a largely uncomprehended phenomenon [6]. From ThFFF retention data α/T values can be obtained. Therefore, thermal diffusion coefficients can be determined with ThFFF, provided that molecular diffusion coefficients are measured independently. In this way thermal diffusion coefficients of various polymer–solvent systems have been obtained and with this data the understanding of

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the thermal diffusion phenomenon has increased significantly. For all polymer–solvent systems studied so far it has been found that the magnitude of the thermal diffusion effect is independent of the molecular mass of the polymer [6]. Therefore, the ThFFF fractionation of a given polymer species is based solely on differences in molecular diffusion or mass. Also the branching configuration of the polymer has no influence on the thermal diffusion coefficient [7]. However, the extent of thermal diffusion is strongly dependent on the chemical nature of both the polymer and the solvent [6,8]. Furthermore, it has been found that the thermal diffusion coefficient is temperature dependent [9] and that the thermal diffusion phenomenon is usually not very strong in aqueous solutions [10].

The fact that ThFFF is a powerful tool for the fractionation of synthetic polymers has often been demonstrated [8,11–15]. Recently it has been shown that ThFFF can also be used for particle analysis [16,17]. Due to the fact that adsorption and degradation phenomena are minimal inside the ThFFF channel, polymers of ultra high molecular mass can effectively be analyzed with ThFFF [18]. Furthermore, samples of a broad molecular weight range can be fractionated with sufficient resolution in an acceptable analysis time using temperature programming [19,20]. As retention in ThFFF is also determined by the thermal diffusion coefficient, fractionation occurs not only according to molecular mass but also according to chemical nature [8,21]. Therefore, ThFFF can be used to obtain chemical and structural information of polymer samples [22].

As was stated earlier the extent of thermal diffusion strongly depends on the chemical nature of both the polymer and the solvent. Because a high thermal diffusion coefficient is beneficial for the separation speed, the choice of the solvent is extremely important for the ThFFF analysis of a particular polymer species [8,23]. Giddings et al. [11] demonstrated that PTHF standards could successfully be retained in both tetrahydrofuran (THF) and ethyl acetate. In the work presented here, α/T values for four PTHF standards of different molecular mass have been determined in eight organic solvents, including the two mentioned above. Diffusion measurements are currently being done in order to determine thermal diffusion coefficients for PTHF in the various organic solvents. With the separation of two PTHF standards in a constant run time, the best solvent for the ThFFF analysis of PTHF samples has been found. The effects of polymer concentration and temperature drop on retention have also been investigated.

2. Theory

2.1. Determination of α/T values

The theoretical description of retention in FFF is given in several textbooks and papers [24–26]. The retention ratio, R, defined as the ratio of the void time of the system and the retention time, can be expressed as:

$$R \approx 6\lambda\left[\coth\left(\frac{1}{2\lambda}\right) - 2\lambda\right] \quad (1)$$

where λ is the dimensionless zone thickness, which is equal to the ratio of the mean layer thickness of the concentrated polymer zone and the channel thickness.

The mean layer thickness is defined as the ratio of the molecular diffusion coefficient and the migration velocity towards the accumulation wall caused by the external field or gradient. In ThFFF the migration velocity towards the cold wall, u, is approximated by [27]:

$$u \approx D_{\mathrm{T}} \frac{\Delta T}{w} \quad (2)$$

where D_{T} is the thermal diffusion coefficient, w is the channel thickness and ΔT is the temperature drop over the channel.

From Eq. (2) it follows that the parameter λ in ThFFF can be approximated by:

$$\lambda \approx \frac{D}{D_{\mathrm{T}}\,\Delta T} = \frac{1}{\frac{\alpha}{T}\Delta T} \quad (3)$$

where D is the molecular diffusion coefficient, T is the temperature and α is the Soret coefficient.

The retention ratio can easily be measured from the fractogram and with Eqs. (1) and (3) the R value can be converted into a corresponding α/T value. If the molecular diffusion coefficient of the polymer sample in solution is known, the thermal diffusion coefficient can be determined from the α/T value.

However, the use of a temperature gradient in ThFFF leads to theoretical complications. Because both the solvent viscosity and thermal conductivity of an organic solvent are temperature dependent, the ideal flow profile of the carrier liquid and concentration profile of the compressed solute will be disturbed. If empirical relations are used to express the temperature dependence of the solvent viscosity and thermal conductivity, these effects can be theoretically accounted for [28,29]. The temperature dependence of the solvent viscosity, η, can best be expressed as:

$$\frac{1}{\eta} = a_0 + a_1 T + a_2 T^2 + a_3 T^3 \tag{4}$$

where the coefficients a_i are empirical constants which can be found when viscosity data at various temperatures are fitted according to Eq. (4).

The temperature dependence of the thermal conductivity, κ, is usually described as:

$$\kappa = b_0 + b_1 (T - T_c) \tag{5}$$

where b_0 and T_c are the thermal conductivity and temperature at the cold wall, respectively.

The parameter b_1, which is frequently denoted as $d\kappa/dT$, is considered to be constant in the working range of the temperature.

The theoretical model describing retention with the use of Eqs. (4) and (5) becomes so complex that a numerical integration routine is necessary to calculate accurate α/T values from the measured retention ratios [29]. In this work the temperature dependence of the solvent viscosity and thermal conductivity has been accounted for. However, the temperature dependence of α/T itself [29] has been ignored and the α/T values have been directly assigned to the temperature in the centre of gravity of the corresponding solute zones [9]. Viscosity and thermal conductivity data were taken from literature to obtain the values of the empirical constants given in Table 1 [9,28,30–32].

2.2. Solvent choice in ThFFF

A time optimization scheme as used in chromatography can also be employed for polymer separation methods [33]. The analysis time, t_a, defined as the time needed to separate two polymer fractions of absolute molecular mass with a given resolution R_s, is determined by the separation requirements and the efficiency and mass selectivity of the fractionation method [8,33]. If the temperature dependence of the solvent viscosity and thermal conductivity is neglected, the analysis time in ThFFF can be expressed as:

$$t_a = \frac{16}{b^2} R_s^2 \left[\frac{M}{\Delta M} \right]^2 \left[\frac{\chi}{R\,(S/b)^2} \right] \frac{w^2}{D} \tag{6}$$

Table 1
Values of the empirical constants describing the temperature dependence of the solvent viscosity and thermal conductivity

Solvent	a_0	a_1	a_2	$a_3 \times 10^5$	$\kappa(293)$ (W/sK)	$b_1 \times 10^5$ (W/sK^2)
Benzene	6445.30	−80.060	0.2936	−26.477	0.1477	−35.00
Cyclohexane	4081.22	−40.278	0.1094	−2.5481	0.1209	−25.19
Ethyl acetate	3828.96	−49.428	0.1914	−14.130	0.1519	−50.21
Ethylbenzene	2892.92	−35.176	0.1284	−8.3949	0.1322	−24.39
MEK	−227.871	2.8762	−0.01531	12.076	0.1464	−22.68
THF	7622.73	−88.933	0.3344	−32.587	0.1398	−19.89
Toluene	3109.76	−45.318	0.1818	−15.078	0.1320	−27.24

where S is the mass selectivity [11], χ is the λ dependent function in the plate height expression [34], b is a constant approximately equal to 0.5 [6] and M and ΔM are the mean molecular mass and the difference in molecular mass of the two polymer fractions, respectively.

The term $[\chi/(R\,(S/b)^2)]$, which is only a function of the parameter λ, decreases with decreasing λ value. When λ approaches zero, χ can be approximated by $24\lambda^3$ [34], R is equal to 6λ [24] and (S/b) approaches 1 [8]. Under these circumstances Eq. [6] can be simplified to:

$$\begin{aligned} t_\mathrm{a} &= \frac{64}{b^2} R_\mathrm{s}^2 \left[\frac{M}{\Delta M}\right]^2 \frac{\lambda^2 w^2}{D} \\ &= \frac{64}{b^2} R_\mathrm{s}^2 \left[\frac{M}{\Delta M}\right]^2 \frac{D}{D_\mathrm{T}^2} \left[\frac{w}{\Delta T}\right]^2 \end{aligned} \tag{7}$$

The latter expression in Eq. (7) is obtained with the use of Eq. (2). It can be noted that the analysis time is independent of the flow rate of the carrier liquid. The choice of the solvent will influence the thermal and molecular diffusion coefficients [6] and, thereby, the analysis time. For the ThFFF analysis of a particular polymer species the solvent for which the shortest analysis time (i.e. highest separation speed) is obtained (for given separation requirements, temperature gradient and molecular mass range) should be used.

To find the optimum solvent for the ThFFF analysis of PTHF samples, two PTHF standards have been separated in a constant analysis time (or rather a constant run time [23]) in the various organic solvents. The fractograms are given in the Results and Discussion section. By adjusting the flow rate the analysis time has been kept the same for each solvent. In this way differences in separation speed are indicated by differences in resolution. The solvent for which highest resolution is found should be used for the ThFFF analysis of PTHF samples. Mathematically this approach can be visualized by rewriting Eq. (7) to:

$$\begin{aligned} R_\mathrm{s} &= \frac{b}{8}\sqrt{t_\mathrm{a}}\,\frac{\Delta M}{M}\,\frac{\sqrt{D}}{\lambda w} \\ &= \frac{b}{8}\sqrt{t_\mathrm{a}}\,\frac{\Delta M}{M}\,\frac{D_\mathrm{T}}{\sqrt{D}}\,\frac{\Delta T}{w} \end{aligned} \tag{8}$$

In earlier work we concluded that using the solvent in which lowest λ values were found for a given molecular mass and temperature drop, would lead to an optimum ThFFF separation performance [8]. Although this conclusion will usually be valid, some extra considerations are in order. Eq. (8) demonstrates that the resolution at constant analysis time is not only influenced by λ but also by D. A high value for the latter parameter can therefore compensate for an unfavourable λ value, and in this way a high separation speed still can be obtained. This is demonstrated in Fig. 1, where the resolution is plotted as a function of the molecular diffusion coefficient for various λ values. Fig. 1 is generally valid because it is based on Eq. (6) rather than on Eq. (8). As can be seen from this figure, a higher λ value can still lead to the same or even better resolution in a constant analysis time, when the molecular diffusion coefficient and, therefore, also the thermal diffusion coefficient are very high.

3. Experimental

3.1. Instrumentation

The T100 Thermal FFF system used in this work was obtained from FFFractionation (Salt Lake City, UT, USA). The experimental set-up for the ThFFF measurements has been described in detail previously [8,23]. A channel thickness of 127 μm was used. The channel length and breadth were 46 cm and 1.6 cm, respectively. The void volume of the channel was equal to 0.75 ml. The eluting polymer zones were detected by means of an Evaporative Light Scattering Detector (Model 2A, Varex, Burtonsville, MD, USA). The outlet of the channel was connected to the detector with the use of a fused-silica capillary with an internal diameter of 100 μm, an external diameter of 360 μm and a length of 1 m (Polymicro Technologies, Phoenix, AZ, USA). In this way a sufficiently large back pressure was created to avoid boiling of the organic solvents inside the ThFFF channel.

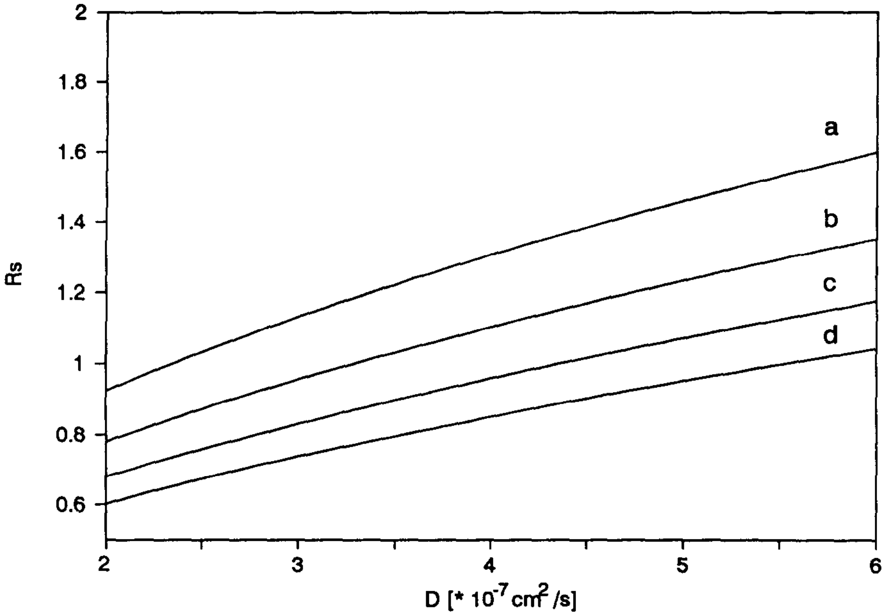

Fig. 1. Resolution at constant analysis time for the separation of two polymer fractions with a mean molecular mass of 175 000 and a difference in molecular mass of 100 000, with $w = 127$ μm, $t_a = 20$ min, and $b = 0.5$. (a) $\lambda = 0.05$; (b) $\lambda = 0.06$; (c) $\lambda = 0.07$; (d) $\lambda = 0.08$

Furthermore, this set-up ensured that the dead volume was negligibly small (0.008 ml).

The α/T measurements of the PTHF standards in the various organic solvents were performed with a temperature drop of 40 K and a cold wall temperature of 298 K. Due to the low retention of the PTHF standards in cyclohexane, a temperature drop of 80 K ($T_c = 302$ K) was used in this case. For all retention measurements the flow rate was set at 0.2 ml/min and a stop flow period of 5 min was employed after injection to allow relaxation. The concentration of the injected solutions of the PTHF standards was usually equal to 0.25 mg/ml. However, for the PTHF standard with a molecular mass of 547 000 the polydispersity was quite high and as a result a higher concentration (0.5 or 0.75 mg/ml) had to be used for this standard. The void time was determined by adding a polystyrene standard with a molecular mass of 580 (0.05 mg/ml) to the samples. For some of the experiments displayed in the figures different experimental conditions have been used, as specified in the corresponding legends.

For the plate height measurements the linear solvent velocities were determined from the void times and the channel length. The channel length was corrected for the distance traversed prior to the relaxation process. For the PTHF standard with a molecular mass of 282 300 a sample concentration of 0.5 mg/ml was used to ensure a sufficiently high signal-to-noise ratio for an accurate determination of the peak width. Sigma values were determined by fitting the signal to a Gaussian curve using the FFFractionation analysis software (version 2.0). In this way also the peak symmetry could be checked.

3.2. Materials

All solvents were of analytical-reagent grade and were filtered (type FH, 0.5 μm, Millipore, Bedford, MA, USA) prior to use. Cyclohexane, dioxane, ethylbenzene and methylethylketone (MEK) were obtained from Merck (Darmstadt, Germany), tetrahydrofuran (THF) and toluene from Janssen Chimica (Geel, Belgium), benzene from Baker (Deventer, The Netherlands) and ethyl acetate from Lamers and Pleuger ('s Hertogenbosch, The Netherlands). The polystyrene standard (PS) used in this work ($M_r = 580$, $\mu < 1.18$) was supplied by Merck, the PTHF standards ($M_r = 67\,000$, $\mu = 1.08$; $M_r = 99\,000$, $\mu = 1.08$; $M_r = 282\,300$, $\mu = 1.08$; $M_r = 547\,000$, $\mu = 1.35$) by Polymer Laboratories (Church Stretton, Shropshire, UK).

4. Results and discussion

In Table 2 the α/T values measured for the PTHF standards in the various organic solvents are given. The temperature dependence of the solvent viscosity and thermal conductivity was accounted for. The α/T values have been assigned to the temperature in the centre of gravity of the concentrated polymer zones. As the molecular diffusion coefficient decreases with increasing molecular mass, the zone thickness of the concentrated polymer zone is smaller for the standards of a high molecular mass. Because a decrease in zone thickness corresponds to a decrease in T_{cg}, the temperature in the centre of gravity of the solute zone decreases with increasing molecular mass of the PTHF standard. Since in the different solvents the PTHF standards possess different molecular and thermal diffusion coefficients, the observed T_{cg} was also dependent on the solvent used. A high α/T value corresponds to a low λ value and, therefore, to a temperature in the centre of gravity of the solute zone which is almost equal to the cold wall temperature. These considerations do not, of course, apply for the retention measurements of the PTHF standards in cyclohexane, which were performed at a higher temperature drop due to the fact that only very low retention was found in this solvent. No α/T values are given for the PTHF standards in either dioxane or MEK. The reason for this is that insufficient retention was found for the standards in these two solvents, even at a temperature drop of 80 K. The fact that for polystyrene in MEK a very high thermal diffusion coefficient is found [6], is in sharp contrast with these findings and demonstrates the strong influence of the chemical nature of the polymer on the thermal diffusion phenomenon.

For the determination of the α/T values the concentration of the injected samples of the PTHF standards was usually equal to 0.25 mg/ml. Only for the PTHF standard with a molecular mass of 547 000 a higher sample concentration of 0.5 or 0.75 mg/ml was used because of the higher polydispersity of this standard. Because inside the ThFFF channel the polymer zones are compressed into thin layers at the cold wall, in which the concentration is much higher than the concentration of the injected sample,

Table 2
α/T values for PTHF standards in the various organic solvents

Solvent	Molecular mass of the PTHF standards							
	67 000		99 000		282 300		547 000	
	α/T (K^{-1})	T_{cg} (K)	α/T (K^{-1})	T_{cg} (K)	α/T (K^{-1})	T_{cg} (K)	α/T (K^{-1})	T_{cg} (K)
Benzene	0.116	306	0.151	305	0.272	302	0.436	300
Cyclohexane	0.061	318	0.071	316	0.132	310	0.213	307
Ethyl acetate	0.121	307	0.151	306	0.263	303	0.443	301
Ethylbenzene	0.147	306	0.183	305	0.321	302	0.604	301
THF	0.074	311	0.093	310	0.175	306	0.321	303
Toluene	0.136	306	0.179	305	0.316	302	0.537	301

$w = 127$ μm, $\Delta T = 40$ K or 80 K (cyclohexane), $T_c = 299$ K (ethyl acetate, ethylbenzene and toluene), $T_c = 298$ K (benzene), $T_c = 300$ K (THF), $T_c = 302$ K (cyclohexane). Each α/T value is the average of three different measurements, the relative standard deviation is in the order of 2–4%

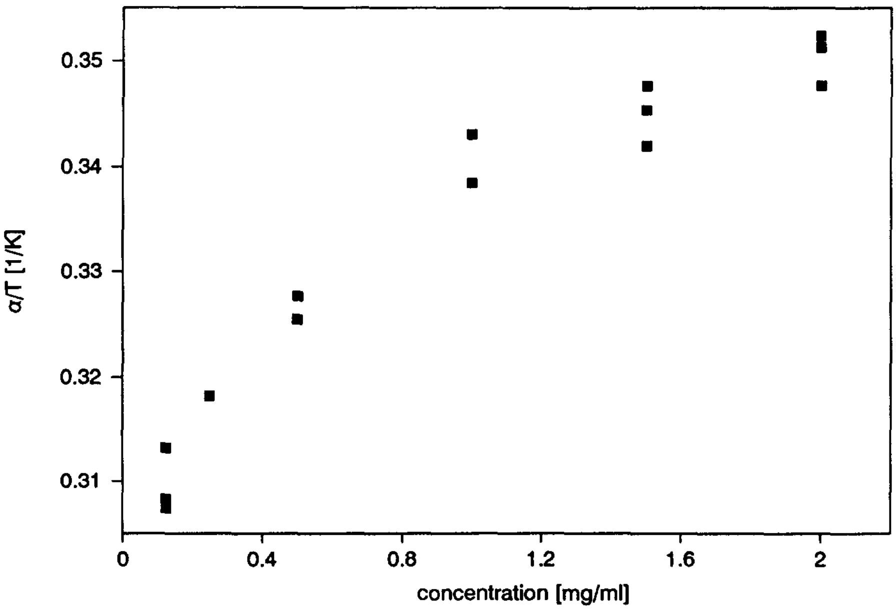

Fig. 2. The influence of the polymer concentration on the α/T value of PTHF 282 300 in toluene, with $w = 127$ μm, $\Delta T = 40$ K, $T_c = 298$ K, t_0 = PS 580 (0.05 mg/ml), flow = 0.3 ml/min, and stop-flow time = 3 min.

concentration overloading can be a severe problem in ThFFF [35,36]. In Fig. 2 the effect of the sample concentration on the measured α/T value is demonstrated. For this system (PTHF M_r = 282 300 in toluene with $\Delta T = 40$ K) the λ value is approximately equal to 0.08. This corresponds initially to a polymer concentration at the cold wall which is more than 12 times higher than the concentration of the injected sample displayed on the x-axis of Fig. 2 [36]. As was also found in previous work [8], retention and thus α/T increased with increasing polymer concentration. This effect cannot be explained by the concentration dependence of the molecular diffusion coefficient because molecular diffusion tends to increase with increasing polymer concentration [37]. The increase in retention with increasing polymer concentration is probably caused by the increasing viscosity in the compressed polymer zone [36]. The concentration profile will lead to a viscosity gradient which will skew the velocity profile of the carrier liquid near the cold wall. As a result the migration velocity will be lower than expected and an apparent increase in retention is observed. Fig. 2 demonstrates that concentration effects are visible even when the polymer concentration is well below 1 mg/ml. Even for the lowest sample concentrations a concentration dependence of α/T was found. For polymer concentrations higher than 1 mg/ml the concentration effect appears to level off and for these solutions a deviation of approximately 12% in the α/T values was found compared to the values obtained at the lowest concentration (0.125 mg/ml).

In Fig. 3 the λ values of the PTHF standards are plotted as function of $M^{-\frac{1}{2}}$ for three solvents. The correlation between the molecular diffusion coefficient and the molecular mass is often expressed according to the empirical relationship $D = A \cdot M^{-b}$, where A and b are constants [6]. The constant b is approximately equal to 0.5 for most polymer–solvent systems. With the use of Eq. (3) it can be seen that the linear relationship (which was found in all solvents) displayed in Fig. 3, indicates that also for PTHF in the various solvents the thermal diffusion coefficient appears to be independent of the molecular mass of the polymer. However, molecular diffusion coefficients should be determined independently

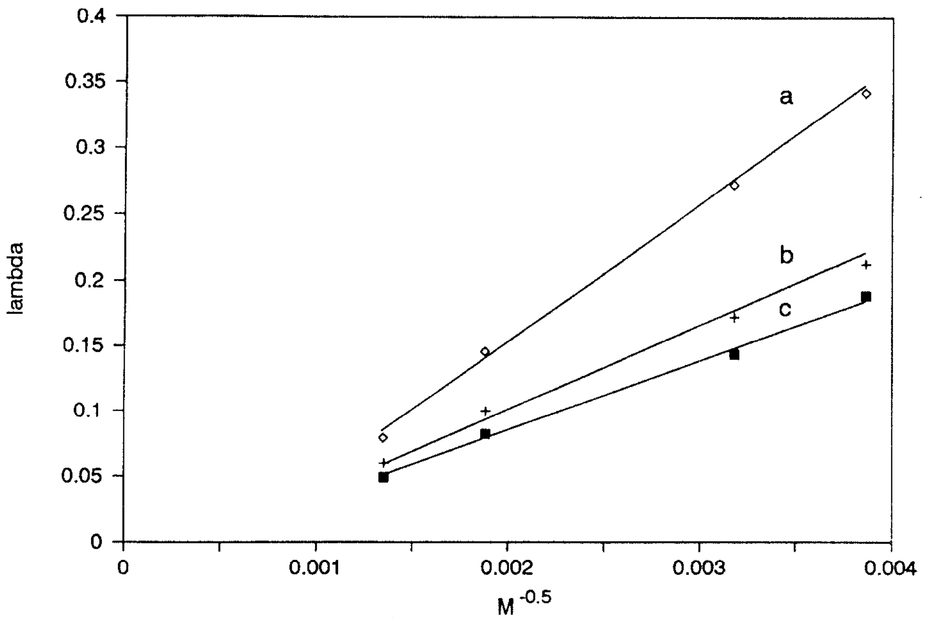

Fig. 3. λ Values of the PTHF standards as function of $M^{-\frac{1}{2}}$. Conditions as given in Table 2. (a) THF; (b) ethyl acetate; (c) toluene.

to verify this conclusion. Just as was found for polybutadiene [8], relatively high λ values (low retention) of the PTHF standards were obtained in THF, whereas highest retention was found in toluene and ethylbenzene.

For the PTHF standard with a molecular mass of 282 300 also the effect of the temperature drop on the λ value in toluene was investigated. The results, given in Fig. 4, are in good agreement with Eq. (3). As was expected a linear relationship was found between the λ value and the reciprocal value of the temperature drop. These results indicate that the measurements are free of systematic errors caused by the set-up. This was also confirmed by the fact that the retention ratio was independent of the flow rate of the carrier liquid.

Next, two PTHF standards with molecular masses of 67 000 and 282 300 were separated in the various organic solvents at a constant run time [23] and temperature gradient. The results are displayed in Fig. 5. The fractionation of the two standards in cyclohexane is not given in Fig. 5 due to the very poor resolution found in this solvent. Going from fractogram a to e, the λ value for a given molecular mass and temperature drop decreases. As a lower λ value corresponds to higher retention, the flow rate had to be increased going from fractogram a to e in order to keep the run time constant. To a first approximation the separation speed is expected to increase with decreasing λ value [see Eq. (8)]. From this point of view the resolution at constant run time is expected to increase going from THF to ethylbenzene and toluene. Although in general this trend can be observed in Fig. 5, two striking features attract attention.

Firstly, it can be seen that, although λ values for a given molecular mass and temperature drop are comparable for PTHF in ethylbenzene and toluene, a higher resolution and thus separation speed was obtained using the latter solvent. The same effect was also found for the ThFFF fractionation of polybutadiene [8]. As the λ values are comparable also the α/T values are identical, and therefore it can be stated that the ratio of D and D_T for a given PTHF standard is the same in toluene and ethylbenzene. However, the individual values of the molecular and thermal diffusion coefficients can still be different in the two solvents. Without the loss of retention, high molecular diffusion (and thus high thermal

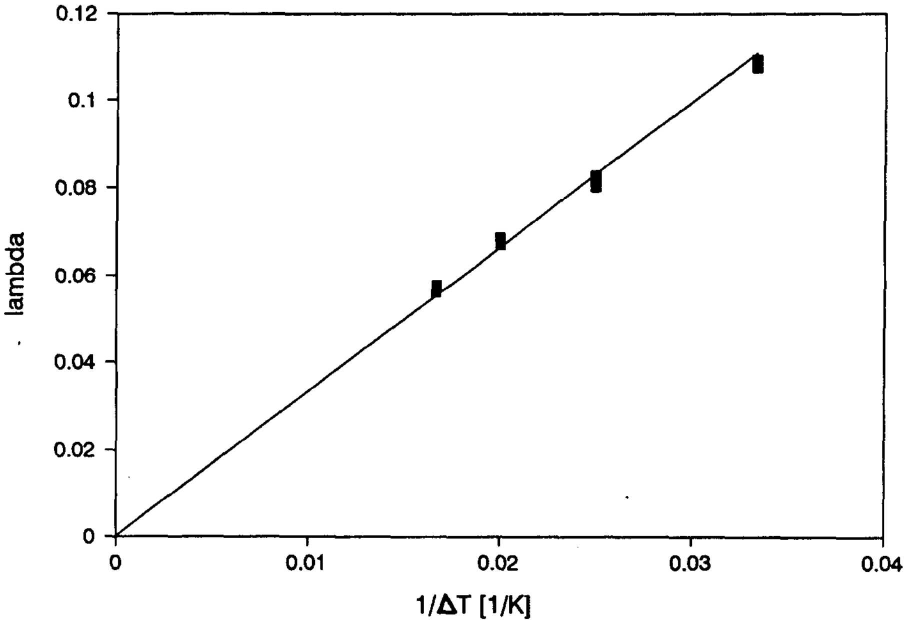

Fig. 4. The effect of ΔT on the λ value for PTHF 282 300 (0.25 mg/ml) in toluene, with $w = 127$ μm, t_0 = PS 580 (0.05 mg/ml), flow = 0.2 ml/min, stop-flow time = 5 min, and $\Delta T = 60$ K ($T_{cg} = 300$ K); $\Delta T = 50$ K ($T_{cg} = 301$ K); $\Delta T = 40$ K ($T_{cg} = 299$ K); $\Delta T = 30$ K ($T_{cg} = 299$ K).

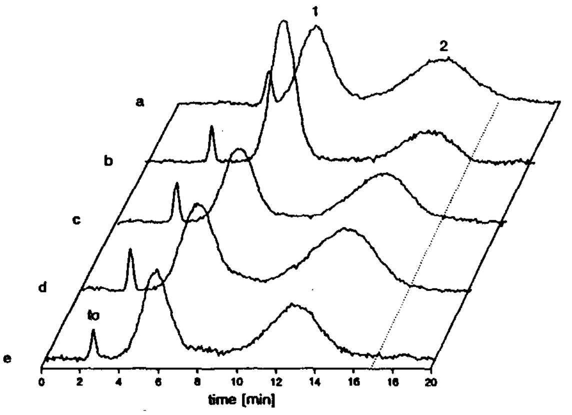

Fig. 5. Separation of a PTHF standard with M = 67 000 (0.33 mg/ml) (1) and a PTHF standard with M = 282 300 (0.5 mg/ml) (2) in various solvents performed at a constant analysis time of 17 min, with $w = 127$ μm, $\Delta T = 80$ K, $T_c = 302$ K, stop-flow time = 2 min, and t_0 = PS 580 (0.03 mg/ml). (a) THF, flow = 0.16 ml/min; (b) ethyl acetate, flow = 0.22 ml/min; (c) benzene, flow = 0.25 ml/min; (d) ethylbenzene, flow = 0.3 ml/min; (e) toluene, flow = 0.3 ml/min.

diffusion) is beneficial for the separation speed in ThFFF [8]. From fractograms d and e in Fig. 5 it can therefore be concluded that both D and D_T for a PTHF standard of a given molecular mass must be lower in ethylbenzene than in toluene. Consequently, it is better to use the latter of the two solvents for the ThFFF analysis of this polymer species.

Secondly, Fig. 5 demonstrates that highest separation speed for the ThFFF fractionation of PTHF samples is obtained when ethyl acetate is used. Although relatively low retention was found for the PTHF standards in this solvent, the highest resolution was obtained for fractogram b. The results displayed in Fig. 5 indicate that for an optimal ThFFF analysis of PTHF samples ethyl acetate is the best solvent to use. As was already explained in the theoretical part, this must be caused by the fact that PTHF standards posses a high thermal diffusion coefficient combined with an even higher molecular diffusion coefficient when they are dissolved in ethyl acetate.

Under normal working conditions the plate height H, in ThFFF, can be approximated by [13]:

$$H = H_{\mathrm{pd}} + \frac{\chi w^2}{D} v \qquad (9)$$

where v is the mean linear velocity of the carrier liquid and H_{pd} is the apparent contribution to the plate height caused by the molecular mass distribution of the polymer sample.

Therefore, measuring the plate height at various fluid velocities in principle provides a means to determining both the molecular diffusion coefficient and the polydispersity [13]. In combination with retention measurements also the magnitude of the thermal diffusion coefficient can be estimated.

For the PTHF standard with a molecular mass of 282 300, plate height measurements were performed at various flow rates in THF, benzene, toluene and ethyl acetate. The results are given in Fig. 6. In good agreement with Eq. (9), a linear increase in system dispersion was found with increasing linear solvent velocity. Fig. 6 also demonstrates that with the use of ethyl acetate the lowest plate heights were obtained. For PTHF in ethyl acetate a relatively high λ value was found, which corresponds to a high χ value. Therefore, these low plate heights can only be caused by a high molecular diffusion coefficient.

As was previously mentioned, molecular and thermal diffusion coefficients can be determined from the results in Fig. 6 and Table 2. In this way for PTHF $M_r = 282\,300$ in ethyl acetate a D value of $6.1 \cdot 10^{-7}$ cm^2 s^{-1} and a D_T value of $1.7 \cdot 10^{-7}$ cm^2 s^{-1} K^{-1} was found, whereas for the same standard dissolved in toluene a D value of $2.7 \cdot 10^{-7}$ cm^2 s^{-1} and a D_T value of $0.9 \cdot 10^{-7}$ cm^2 s^{-1} K^{-1} was obtained. This clearly illustrates how for PTHF in ethyl acetate the highest separation speed is obtained, despite the relatively low retention.

The values for D and D_T determined in this way are not very accurate. Firstly, it is known that with the use of an Evaporative Light Scattering Detector a non-linear calibration curve is obtained. With an increase in polymer concentration a more than proportional increase in signal is usually observed [38]. As a result the

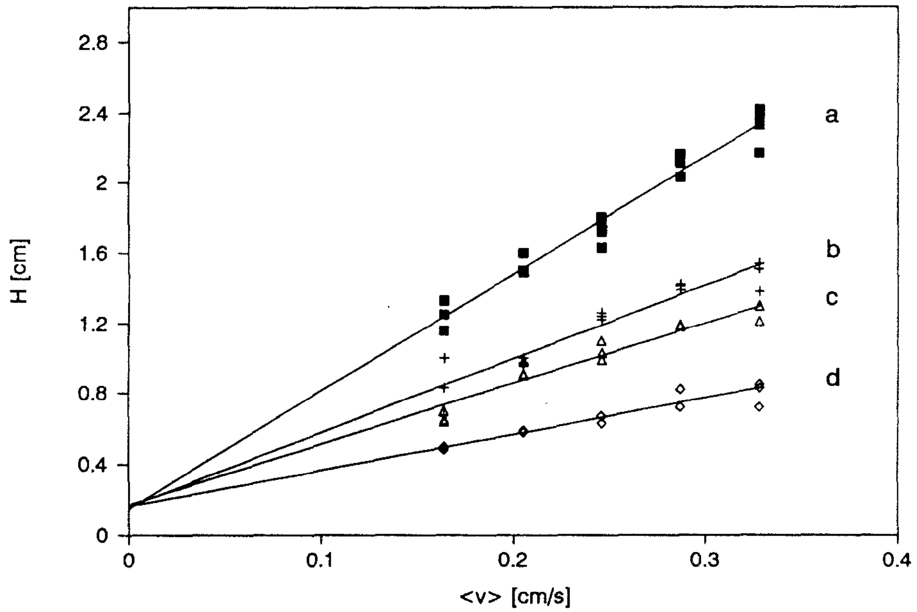

Fig. 6. Plate height as function of the mean linear velocity of the carrier liquid for a PTHF standard with $M_r = 282\,300$ (0.5 mg/ml) in various solvents, with $w = 127$ μm, $\Delta T = 40$ K, $T_c = 298$ K, stop-flow time = 2 min, and t_0 = PS 580 (0.1 mg/ml). (a) THF; (b) benzene; (c) toluene; (d) ethyl acetate.

measured peak width and, therefore, the plate height will be smaller than the actual dispersion of the polymer zone. Secondly, the χ value was determined from the λ value using the relationship given by Giddings et al. [34]. Therefore, no correction was made for the temperature dependence of the solvent viscosity and thermal conductivity and as a result systematically lower χ values have been used [24]. Although the two effects mentioned above counteract each other when the molecular diffusion coefficient is determined from the plate height data, the results must be considered with great care. The large difference in the values found for the diffusion coefficient of the PTHF standard in the two solvents should be verified by an independent technique (e.g. Light Scattering). The results displayed in Figs. 5 and 6 and Table 2 show only qualitatively that the thermal and molecular diffusion coefficients of the PTHF standard are highest in ethyl acetate.

Figs. 5 and 6 clearly demonstrate that for the ThFFF analysis of PTHF samples ethyl acetate is the best solvent to use. Therefore, the maximum separation speed for a given molecular mass range and temperature gradient is fully determined. Even in the most appropriate solvent, separation speeds can differ strongly for different polymer species. For the ThFFF analysis of polybutadiene it is best to use toluene as solvent (n.b. polybutadiene samples do not dissolve in ethyl acetate). However, even with this solvent the separation speed is fairly low and as a result a long analysis time is necessary to obtain a good fractionation [8]. Because of the high molecular and thermal diffusion of PTHF samples in ethyl acetate, a very high maximum separation speed is obtained in this case. This is demonstrated in Fig. 7 with the baseline separation in 8 min of two PTHF standards ($M_r = 67\,000$ and 282 300) in ethyl acetate performed under a reasonable temperature gradient.

One of the objectives of a polymer fractionation method is the accurate determination of the molecular mass distribution of polydisperse materials. With adequate calibration standards and a deconvolution procedure to remove system dispersion from the observed signal, ThFFF can be used effectively to obtain such information [39]. When ThFFF is used in combination with a concentration dependent detector and a viscosity detector, molecular mass distributions can even be measured without the need for calibration [15]. However, as was demonstrated by Kirkland and Rementer [15], concentration overloading can be very important in this application. As a result of the polydispersity often a very broad signal is observed for polydisperse materials. This leads to a low signal-to-noise ratio in the fractogram and for an accurate determination of the molecular mass distribution high sample concentrations have to be injected. As was mentioned previously, retention tends to increase with increasing polymer concentration and as a result a systematic error in the determination of the molecular mass distribution can be made. The polydispersity of the PTHF standard with a molecular mass of 547 000 is quite high ($\mu = 1.35$) and as a result broad signals were obtained for this standard. In Fig. 8 the effect of the sample concentration for polydisperse materials is demonstrated with the fractionation of PTHF $M_r = 547\,000$ in toluene ($\lambda \approx 0.05$). Injection of higher concentrations resulted in a shift of the signal to higher retention, which will lead to a determination of a mean molecular mass which will be systematically too high. Toluene was used as the solvent because lowest

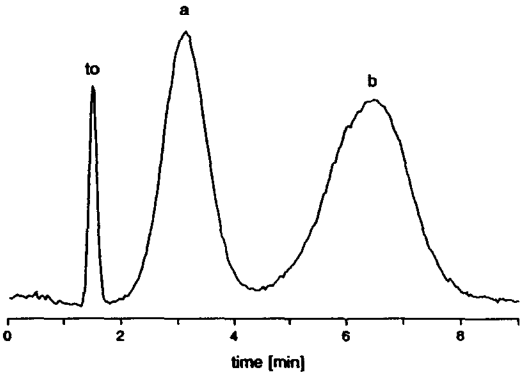

Fig. 7. High speed ThFFF separation of two PTHF standards with (a) $M_r = 67\,000$ (0.25 mg/ml) and (b) $M_r = 282\,300$ (0.5 mg/ml) in ethyl acetate, with $w = 127$ μm, $\Delta T = 80$ K, $T_c = 303$ K, flow = 0.5 ml/min, stop-flow time = 2 min, and t_0 = PS 580 (0.05 mg/ml).

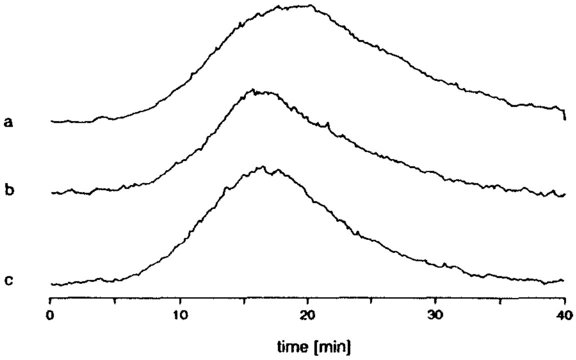

Fig. 8. Effect of polymer concentration on the ThFFF fractionation of a broad PTHF standard with $M_r = 547\,000$ ($\mu = 1.35$) in toluene, with $w = 127$ μm, $\Delta T = 40$ K, $T_c = 299$ K, flow = 0.2 ml/min, and stop-flow time = 5 min. Concentration of injected sample is 5 mg/ml for (a), 2 mg/ml for (b) and 1 mg/ml for (c).

λ values for PTHF were obtained for this combination. Note that the use of ethyl acetate, the optimum solvent for the analysis of PTHF samples in respect to separation speed, also has the advantage that concentration overloading will be less because the compression of the polymer zones at the cold wall is not that strong due to the relatively low retention found for PTHF samples in this solvent. No accurate molecular mass distribution could be obtained from Fig. 8 in combination with the retention data of the other PTHF standards. This was caused by the fact that the other three standards were all of lower molecular mass and calibration with these standards resulted in a large error due to extrapolation.

5. Conclusions

ThFFF can be used effectively for the fractionation and analysis of PTHF samples. For a given temperature gradient and molecular mass, highest retention for the PTHF standards was found when ethylbenzene or toluene were used as the solvents. No retention was observed when the standards were dissolved in dioxane or MEK. The retention measurements of the PTHF standards in the various organic solvents indicated that the thermal diffusion coefficient is independent of the molecular mass of the polymer, as has been observed for other polymer species.

Although only moderate retention for the PTHF standards was measured when ethyl acetate was used as the carrier liquid, highest separation speed was observed when the standards were dissolved in this solvent. Therefore, for the ThFFF analysis of PTHF samples ethyl acetate is the best solvent to use. These results show that low retention for a given molecular mass and temperature gradient does not always correspond to a low separation speed. If low retention is caused by a very high molecular diffusion coefficient, still a very efficient ThFFF fractionation is possible.

Even for polymer concentrations well below 1 mg/ml an increase in retention was observed with increasing sample concentration. Especially when molecular mass distributions of polydisperse materials have to be determined with ThFFF, care should be taken to avoid systematic errors due to concentration effects. The use of ethyl acetate as the carrier liquid for the ThFFF analysis of PTHF samples has the additional advantage that concentration overloading is not very profound due to the relatively low retention found in this solvent.

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Symbols

a_i	empirical constants relating solvent viscosity to temperature
b_i	empirical constants relating solvent thermal conductivity to temperature
A,b	empirical constants relating diffusion to molecular mass
D	diffusion coefficient (m^2 s^{-1})

D_T	thermal diffusion coefficient (m^2 s^{-1} K^{-1})
H	plate height (m)
H_{pd}	polydispersity plate height contribution (m)
M	(mean) molecular mass (g mol^{-1})
R	retention ratio
R_s	resolution
S	selectivity
T	temperature (K)
t_0	unretained time (s)
t_a	analysis time (s)
T_c	cold wall temperature (K)
u	field induced velocity of the polymer molecules in the direction of the accumulation wall (m s^{-1})
v	mean linear velocity of the carrier liquid (m s^{-1})
w	channel thickness (m)
α	Soret coefficient
ΔT	temperature drop (K)
η	solvent viscosity (Pa s)
κ	solvent thermal conductivity (J s^{-1} m^{-1} K^{-1})
λ	dimensionless zone thickness
μ	polydispersity
χ	λ dependent function in the non-equilibrium term of the plate height equation

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