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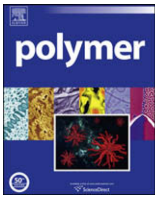

A theory of topological separation of linear and star-shaped polymers by two-dimensional chromatography

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ABSTRACT

Topology influences the size of macromolecules, but polymers are usually distributed with respect to molar mass, which also results in the size distribution within a polymeric sample. Due to this fact size-exclusion chromatography (SEC) is not able to separate even moderately polydisperse polymers by topology; the same is also true for the adsorption chromatography (AC). The full separation by molar mass and topology is not possible by any single mode of chromatography. These problems can be solved by means of two-dimensional chromatography which combines SEC and AC mechanisms. A theory of interactive chromatography of linear and star-shaped ideal-chain polymers is used to analyze two-dimensional chromatographic separation of polydisperse linear and star polymers. Basing on this theory, we simulate 2D-chromatograms for model mixtures of polydisperse linear and star-shaped polymers of equal average molar mass, and demonstrate that 2D-separation of such polymers by topology is possible. A possibility to separate symmetric and very asymmetric stars by 2D-chromatography is predicted. The influence of the molar-mass heterogeneity, pore size and adsorption interaction parameter on the 2D chromatographic pattern is analysed, and the conditions for a good separation of linear and star polymers are formulated. The theoretical results are in a qualitative agreement with the experimental data, which have been reported previously by Gerber and Radke.

1. Introduction

Separation of polymers by topology is still a challenge in the analytical chemistry of polymers. Since topology influences the size of macromolecules, size-sensitive methods like SEC seem to be appropriate. It is the well-known fact that SEC separates macromolecules by their effective molecular radii; since stars have smaller average sizes than linear chains, separation of such polymers by topology by means of SEC is possible, in principle [1,2]. The problem is however in the fact that polymers are usually polydisperse, and the molar-mass heterogeneity may cause difficulties for separation by topology.

Many reports have appeared, describing successful chromatographic analysis of branched, and in particular, of star polymers [3–7]. Mostly the systems were studied with strong correlations between topology and other molecular characteristics of polymers.

Quite often a correlation exists between topology and molar mass. A typical experimental problem is for example to separate stars with different number of equal arms from each other and from a linear precursor of an arm. In this situation topologically different polymers – stars with different number of arms – have quite different molar mass (and different molecular radii), and therefore can be separated by means of either SEC or adsorption chromatography. Such successful separations have been really achieved and reported [3–5].

Another correlation can be used when specific groups differing in their adsorption properties from regular chain units are present in branched polymers (such groups can be situated on the chain ends or/and in the branch points). In such cases the chromatography at the critical conditions (CCC) has turned to be very efficient. CCC corresponds to the critical interaction point which separates the SEC and AC regimes of chromatography. In CCC, the decrease of the entropy of a linear polymer chain due to the presence of the pore walls is compensated by enthalpic adsorption effects. As a consequence, the separation of linear homopolymers with respect to molar mass is absent. The retention is solely governed by small differences in the structure of polymer chains, such as the presence of specific groups, tacticity, or topology. By using CCC, big progress has been made towards solving the functionality type distribution, determining the heterogeneity in the number and the type of the specific end groups. Good reviews on practical achievements in this field are available [8–10]; the theoretical proof has been given in Refs. [11,12]. According to the theory, CCC also turns useful, when specific groups differing in their adsorption properties from regular chain units are present in branched polymers. In particular, it has been shown theoretically [13]

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that a comb-like polymer with adsorbing branch-point groups behaves in CCC as a linear polymer (identical to a comb backbone) with specific groups corresponding to branch-point units; therefore such comb-like polymers can be separated by the number of branch points by means of CCC. A theory of CCC for stars with adsorbing terminal and branch-point groups has been developed as well, and this theory was used to describe and to predict the behavior of such stars in the CCC mode [14]. There are several successful attempts to use CCC for the characterization of star-shaped and branched polymers [4–7]. In fact, CCC separates these polymers not by topology itself, but by the number of specific groups, that is by chemical composition of macromolecules (which is correlated with the chain topology).

We would like to study the problem of topological separation of linear and star polymers in its pure form, and therefore in this paper we are considering a situation where there is no difference in molar mass and in the adsorption interaction of regular, branch-point and terminal chain units of star polymers. By using a molecular-statistical theory of chromatography of linear [12] and star [15] polymers, we analyze possibilities to separate such polymers by means of chromatography. The problem is interesting in a theoretical aspect, since the considered polymers differ solely in their topology. According to the theory [13,14], CCC cannot help in this situation, and the problem seems to be quite difficult. A theoretical analysis can help in finding a solution and in determining favourable chromatographic regimes and conditions.

This problem was not discussed previously, but some relevant experimental results have been reported by Gerber and Radke [16], who found that linear polymers can be separated from stars of the same molar mass by means of two-dimensional chromatography. However the star sample in this study was composed of stars with different number of equal arms, and these stars were separated from each other mostly due to the difference in molar mass.

By using a theory, which is based on the ideal-chain model of macromolecules, we shall study the 2D-chromatography of linear and star polymers in more detail. We shall show theoretically that the separation of linear and star polymers as well as the topological separation of stars with different number of arms can be possible by using two-dimensional chromatography with AC as the first and SEC as the second dimension. The influence of the molar-mass heterogeneity, pore size and adsorption interaction parameter on the 2D chromatographic pattern will be analysed, and the conditions for a good separation of linear and star polymers by topology will be formulated.

To investigate the separation of polydisperse linear and star-shaped polymers and to visualize theoretical predictions, we apply a virtual chromatograph [17] which is a simulation tool for studying chromatographic separation of polymers. The virtual chromatography technique, which is based on the molecular-statistical theory of chromatography of polymers, has been already used to predict one-dimensional separations of polydisperse polymers of various types [13,15,17–20]; in those studies simulated chromatograms have proven to be very similar to the real ones. We extend this technique to simulate 2D-chromatograms for the mixtures of polydisperse linear and star polymers.

The theoretical results will be compared with the available experimental data obtained by Gerber and Radke for the mixture of linear and star-shaped polydisperse polystyrenes.

2. Theory

A theory of size-exclusion chromatography of linear and star polymers has been developed by Casassa [1]. This theory describes the case of the absence of adsorption interaction between chain units and pore walls.

A theory that accounts for adsorption interaction and therefore is capable to describe a behavior of linear polymers in all modes of chromatography has been built by Gorbunov and Skvortsov [21,12]. Kosmas et al. [22] have developed a theory describing the chromatographic behavior of star polymers in wide pores. A more general theory, which covers both wide- and narrow-pore situations and all modes of chromatography of star-shaped polymers, has been developed by Gorbunov and Vakhrushev [15].

All these theories are based on the thermodynamic approach [1], according to which the chromatographic behavior of macromolecules is determined by the equilibrium partitioning of the chains between the mobile and the stationary phases, and therefore can be described by laws of statistical thermodynamics. Although an ideal-chain model used by the theories [1,2,11–15,19,21,22] does not account for excluded volume interactions, these theories explain many features of the behavior of real polymers in chromatography. The ideal-chain model is very advantageous because exact mathematical solutions for polymers of complex architectures are obtainable by using this model.

We use the continuum formalism [23] which is analogous to describing the motion of a Brownian particle in the presence of spatial constraints. It is this technique that was used to build the theory of chromatography of linear and ring homopolymers [1,12,21] and its extensions for more complex polymers [1,2,13–15,19,20,22]. The model parameters in the continuum description are related only to large-scale spatial properties, such as the average radius of an unconfined polymer molecule and the average thickness of an adsorbed macromolecule. The continuum models are largely universal and independent of the microscopic structure of a surface layer and the details of the adsorption interaction potential. The continuum approach gives qualitatively the same results as another popular description based on lattice random walks [24,25]; and there is a relationship between the interaction parameters in both models [26].

In the continuum approach all possible conformations of an ideal chain are described by the diffusion equation, while de Gennes' boundary condition [27] is applied on the pore walls to account for the adsorption interaction. A parameter c entering into the boundary condition, serves in the theory as the adsorption interaction parameter. Negative values of the interaction parameter c correspond to effective repulsive forces between the chain units and pore walls ($c \to -\infty$ corresponds to the conditions of SEC). The point $c = 0$ is usually referred to as an adsorption threshold point or as a critical point of adsorption [12]. Positive values of c correspond to adsorption. At $c > 0$ a sufficiently long macromolecule forms an adsorption layer on the surface; the average thickness of this layer is equal to c^{-1}.

In chromatography adsorption interaction is controlled by selection of adsorbent and solvent; by variation of temperature and composition of a mixed mobile phase.

The theory [15] results in the exact equation expressing the partition coefficient K of an ideal star with f arms as a function of dimensionless parameters $\lambda = -cd$ and $g_i = R_i/d$ $(i = 1,2,\ldots f)$.

$$K_{\text{star}} = \sum_{k_1,k_2,\ldots,k_f=1}^{\infty} I_{k_1,k_2,\ldots,k_f} \cdot \prod_{i=1}^{f} \frac{2\sec(\alpha_{k_i})}{1+\lambda+\lambda^{-1}\alpha_{k_i}^2} \cdot \exp\left(-\alpha_{k_i}^2 g_i^2\right), \tag{1}$$

where

$$I_{k_1,k_2,\ldots,k_f} = \int_0^1 \cos(\alpha_{k_1}\varphi)\cos(\alpha_{k_2}\varphi)\ldots\cos\left(\alpha_{k_f}\varphi\right)\mathrm{d}\varphi.$$

The parameter R_i has a meaning of the radius of gyration of an ideal linear chain which is identical to the i-th arm of the star, $2d$ is the characteristic size of a slit-like pore (the pore is modeled as a space between two parallel planes separated by the distance of $2d$; the two other dimensions of the slit are assumed much greater than the size of a macromolecule), while c is an adsorption interaction parameter. Eigenvalues α_k $(k = 1,2,\ldots\infty)$ depend on λ, being the roots of the equation $\alpha\cdot tg(\alpha) = \lambda$; the properties of these eigenvalues are well known [21].

For a symmetric f-arm star K is expressible in terms of three parameters: f, λ, and $g = R/d$ (R has a meaning of the radius of gyration of a linear chain which has the same molar mass as the whole star macromolecule; for an ideal-chain f-arm star $R^2 = \sum_{i=1}^{f} R_i^2$).

Fig. 1 shows the theoretical dependences of the partition coefficient K of a linear polymer chain and of symmetric stars with $f = 3$, 4 and 5 on the parameter $-\lambda = cd$ at two values of the molecule-to-pore size ratio $g = R/d$; these dependences are calculated by using the Equation (1). If the pore radius d is assumed constant, these functions represent the dependences of K on the adsorption interaction parameter c for linear polymers and different stars at fixed values of R (that is, at fixed molar mass). According to this theory, at $c = 0$, $K = 1$ for both linear and star molecules. There is practically no difference in K of linear and star polymers in the vicinity of the critical point. However, as it can be seen in Fig. 1, at both large negative and positive values of c some difference appears between the partition coefficients of linear polymers and stars.

The theory [15] also results in a number of useful approximate formulae for the partition coefficient of star polymers in various regimes of chromatography. For the purposes of this study asymptotes describing SEC and AC regimes are of interest.

The limit of $c \to -\infty$ corresponds to the condition of SEC. In narrow slit-like pores (at $d \ll R$) the SEC partition coefficient of an ideal star macromolecule with f equal arms can be approximated as:

$$K_{\rm star} \approx A_f\,\frac{8}{\pi^2}\exp\left(-\frac{\pi^2R^2}{4d^2}\right) \approx A_f K_{\rm lin}, \tag{2}$$

where

$$A_f = \left(\frac{4}{\pi}\right)^{f-1}\int\limits_0^{\pi/2}\cos^f(\varphi)\mathrm{d}\varphi = \left(\frac{4}{\pi}\right)^{f-1}\cdot\begin{cases}\dfrac{1\cdot3\cdot5\cdot\ldots\cdot(f-1)}{2\cdot4\cdot6\cdot\ldots\cdot f}\cdot\dfrac{\pi}{2} & f\ \text{even}\\[2ex] \dfrac{2\cdot4\cdot\ldots\cdot(f-1)}{1\cdot3\cdot5\cdot\ldots\cdot f} & f\ \text{odd}\end{cases}.$$

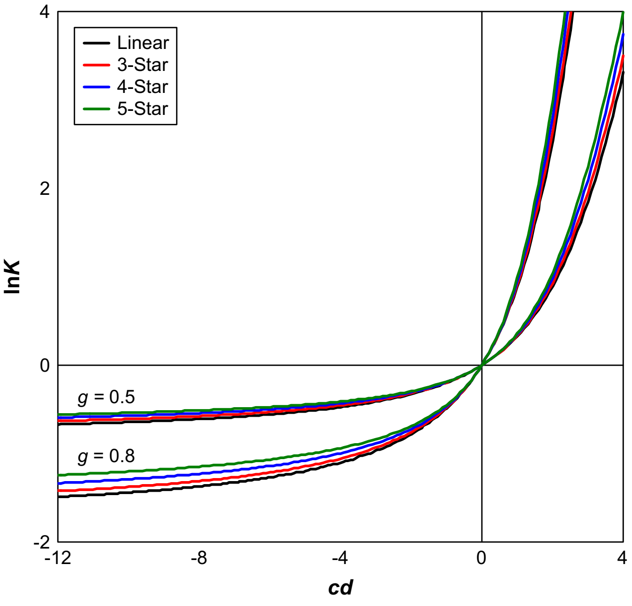

Fig. 1. Dependences of the partition coefficient of linear and star-shaped macromolecules on the scaled interaction parameter cd at two values of $g = R/d$.

In fact, the asymptote of the Equation (2) also holds good at $d < R$, and even at $d \approx R$.

For wide pores (at $d > R$) the well-known result [1] is recovered:

$$K_{\rm star} \approx 1 - \frac{2B_f}{\sqrt{\pi}}\frac{R}{d} \approx K_{\rm lin} + \left(1 - B_f\right)\frac{2}{\sqrt{\pi}}\frac{R}{d}, \tag{3}$$

wherein

$$B_f = \sqrt{\frac{\pi}{f}}\cdot\int\limits_0^{\infty}\left[1 - (\mathrm{erf}(x))^f\right]\mathrm{d}x.$$

The values of A_f and B_f for the first five values of f are given in Table 1. Both $f = 1$ and $f = 2$ correspond to a linear macromolecule.

In the strong adsorption regime, that is at high values of the adsorption interaction parameter $c \gg 1/\min(R_f^{-0.5}, d)$, the theory results in an asymptotic equation, which coincides with the previous result obtained by Kosmas et al. [22]:

$$K_{\rm star} \approx \frac{2^f\exp\left(c^2R^2\right)}{fcd} \approx \frac{2^f}{2f}K_{\rm lin} \tag{4}$$

As it follows from Equations (2)–(4) and can be seen in Fig. 1, at both large negative and positive values of the interaction parameter stars have higher values of the partition coefficient K than linear polymers of the same molar mass; K increases with the number of arms f.

Thus, according to the theory, it is not possible to separate linear macromolecules and stars by topology by means of chromatography at the critical adsorption point or in the vicinity of this point, but there is a principle possibility to do this by using either SEC or adsorption chromatography.

Fig. 1 and Equations (2)–(4) show that in both SEC and AC the effect of topology on the chromatographic retention is rather delicate, while the effect of molar mass is quite pronounced. If polymers are polydisperse, both molar mass and topology influences the chromatographic retention. As it will be shown later, the effects of molar mass and topology can be separated by combining SEC and AC mechanisms in a two-dimensional chromatographic process.

In order to investigate the problem of chromatographic separation of polydisperse polymers by topology, we simulate one- and two-dimensional chromatograms for model mixtures of linear and star-shaped polymers; simulation procedures are based on the equations of the corresponding theories [12,15].

A polydisperse polymer represents an ensemble of individual macromolecules of different chain length (molar mass). The chromatographic elution volume $V = V_0 + V_pK$ is determined by the partition coefficient K (the interstitial volume V_0 and the pore volume V_p are the parameters of the chromatographic system; these parameters can be assumed as constants in a given experiment). Therefore, by calculating partition coefficients for the members of a given ensemble of macromolecules one can construct a theoretical chromatogram for a polydisperse polymer [17–19]. Peak broadening of individual species in the column is accounted for by a parameter characterizing the plate count of the chromatographic system [17].

A similar procedure, modified in a straightforward way, is used here for the simulation of a two-dimensional chromatogram. This procedure includes calculation of pairs of values K_1 and K_2 for the macromolecules of an ensemble. In order to account for different

Table 1
The coefficients A_f and B_f in Equations (2) and (3).

Number of arms, f	1, 2	3	4	5
A_f	1	1.0808	1.2159	1.4016
B_f	1	0.9598	0.9178	0.8798

chromatographic conditions in two dimensions, K_1 and K_2 are calculated at different values of pore size d and interaction parameter c, which correspond to the first and the second dimensions.

A polydisperse star polymer represents a more complex ensemble in which not only the total molar mass but also molar masses of all arms can be different. In simulations all arms are assumed to have logarithmically normal molar mass distributions; the parameter $(M_w/M_n)_{arm}$ is used as a measure of molar-mass heterogeneity of arms.

3. Simulation results and discussion

3.1. A chromatographic separation of polydisperse linear and star-shaped polymers by topology

Real polymers are usually polydisperse. Since in SEC and in AC both molar mass and topology influences the chromatographic retention, the question arises whether these modes of chromatography can provide for a good separation of polydisperse linear and star polymers by topology.

Fig. 2a and b shows one-dimensional chromatograms simulated at SEC and AC conditions for 3-, 4-, and 5-arm star-shaped polystyrenes (of equal average molar mass, $M_w = 30\,000$, and of the same molar-mass heterogeneity of arms, $M_w/M_n = 1.03$) in the mixture with a corresponding linear polystyrene. As can be seen, in both cases peaks corresponding to species of different topology are somewhat shifted, but not resolved because of the considerable peak broadening, which results from the molar mass distribution.

Thus, neither single SEC nor single AC technique can separate linear and star polymers of equal average molar mass by topology even if these polymers have narrow molar mass distributions. However, such separation may become possible, if one will combine SEC and AC mechanisms in a two-dimensional chromatographic process.

Two-dimensional chromatography is a very powerful tool for the analysis of complex polymers, which are heterogeneous in more than one dimension [8–10,28,29]. A fully automated two-dimensional chromatographic system was developed and successfully applied for the analysis of heterogeneous polymers by Kilz et al. [29–32]. A general benefit of two-dimensional chromatography is that it provides more space for a separation: although sample components co-elute in any single dimension, the component zone can be well resolved in two dimensions. Evidently, selected separation mechanisms should not be identical – otherwise separation will converge back to a one-dimensional separation. Since the mechanisms of separation of linear macromolecules and stars in SEC and AC are different, one can expect a considerable reduction in band overlap when coupling these two mechanisms in a 2D technique.

Fig. 2c shows a two-dimensional SEC-AC chromatogram, which is simulated at the SEC and AC conditions of Fig. 2a and b. Usually experimental 2D-chromatograms are given as contour plots [9], and we also use contour plots to present simulated 2D-chromatograms. The ordinate in Fig. 2c represents the retention in the AC dimension, while the abscissa indicates the retention of the fractions in the SEC dimension. As it was expected, Fig. 2c shows a substantial separation of all topologically different components of the mixture.

The chromatogram of Fig. 2c indirectly represents the distributions on both molar mass and topology in the considered model mixture of linear and star polymers. Therefore SEC-AC 2D-chromatography can be considered as a possible method for the complete (at least qualitative) characterization of such polymers with complex heterogeneity. This conclusion is made basing on the results for a model sample of a particular molar mass and molar-mass heterogeneity, and for particular conditions (pore size, adsorption interaction) in the first and in the second dimension. In the subsequent discussion we analyze theoretically how the above-mentioned parameters can influence the separation.

3.2. Effect of molar mass and molar-mass heterogeneity

Similar two-dimensional separations by topology can be achieved for polymers of different molar mass. According to the theory, only the

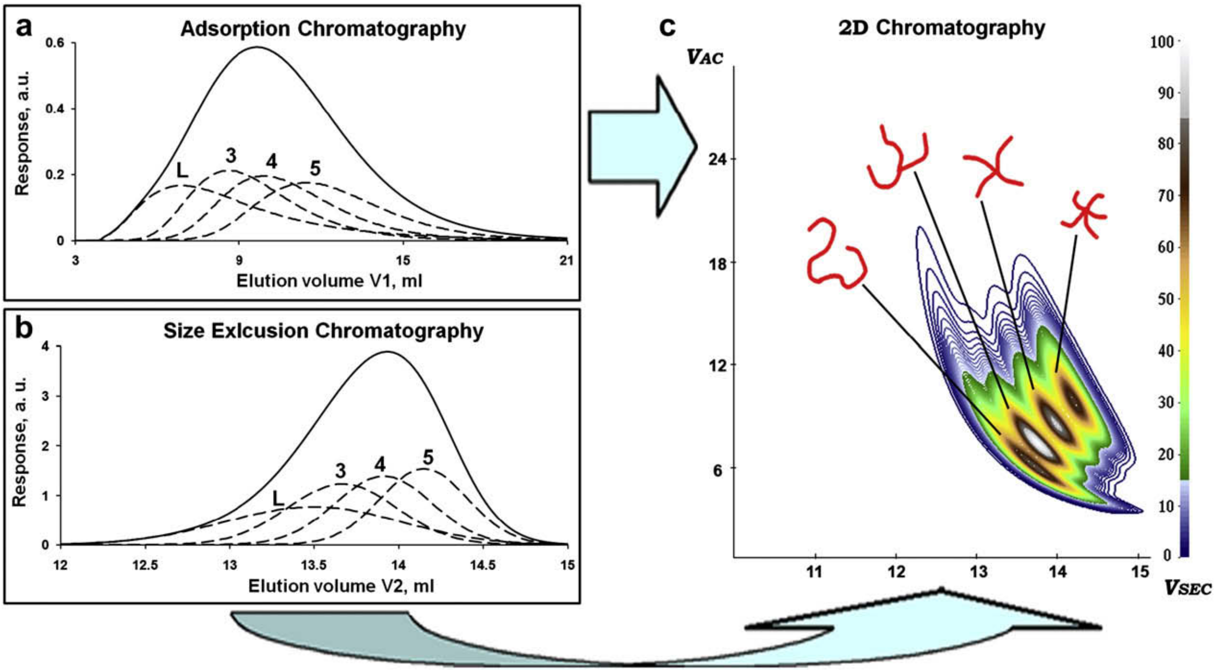

Fig. 2. One-dimensional (a) SEC, (b) AC chromatograms, and (c) two-dimensional SEC-AC chromatogram, simulated for the mixture of linear (L) polystyrenes and PS stars with three (3), four (4) and five (5) arms. Average molar mass $M_w = 30\,000$; $(M_w/M_n)_{arm} = 1.03$. Simulation parameters: pore diameter of SEC and AC columns 16 nm; plate count 3000; (a): void volume $V_0 + V_p = 20$ ml, pore volume $V_p = 10$ ml; (b): adsorption interaction parameter $c = 0.32$ nm^{-1}, $V_0 + V_p = 2$ ml, $V_p = 1$ ml; (c): parameters for SEC and AC dimensions are those of (a) and (b) respectively. Dashed lines in (a) and (b) show chromatograms for the components of the mixture; solid lines represent resulting chromatograms.

scaled parameters like R/d and cd enter Equations (2)–(4), therefore by proper choice of d and c it is always possible to get the same values of the parameters R/d and cd for polymers of different molar mass, and thus to obtain similar separation patterns for these polymers.

In order to illustrate this idea let us consider mixtures which are similar to the above considered one, but composed of either low or high molar mass linear and star components (for example, polymers of the average molar mass $M_w = 3000$ and $M_w = 300\,000$). By choosing the parameters c and d in a way to keep constant values of $M^{0.5}/d$ and cd, we can expect similar separation patterns for these low and high molar mass mixtures. Two-dimensional chromatograms which are simulated for low and high molar mass model samples at such properly scaled parameters c and d are shown in Fig. 3a and b. These chromatograms are really very similar. There is one difference: in the chromatogram for the low molar mass sample (Fig. 3a) oligomeric homologues are completely or partially resolved, and this chromatogram is of a discrete nature, but this is not the case for the high molar mass sample (Fig. 3b). Apart from this difference, the chromatograms of Fig. 3a and b are very similar even quantitatively.

The molar mass distribution is an important factor, which considerably influences the separation pattern: the increase in the polydispersity parameter makes separation poorer. Simulation showed that linear and all different star components can be reasonably separated from each other by 2D-chromatography, if the heterogeneity factor for the arms is less then $(M_w/M_n)_{arm} \approx 1.1$.

Separation of linear polymers and stars having more than three arms is however possible for much higher values of the heterogeneity factor. Fig. 4a–c shows the effect of the molar-mass heterogeneity on the separation of linear polymers and 4-arm stars. As can be seen, a substantial separation of these polymers is still possible at $(M_w/M_n)_{arm} = 1.5$.

In order to learn at which experimental conditions the two-dimensional separation by topology could be possible, we analyze the influence of the main system parameters on the separation pattern.

3.3. *Influence of the pore sizes of SEC and AC columns and of the adsorption interaction on the 2D-separation by topology*

As it follows from the theory and simulations, the pore size of the SEC column has a significant effect on the 2D-separation pattern. While the separation is poor in both narrow (Fig. 5a) and wide pores (Fig. 5c), it becomes practically perfect at some intermediate values of the pore diameter (Fig. 5b). Therefore for a good topological separation it is very important to optimize the SEC column pore size. Simulations show that the best results can be expected when the average pore radius of the SEC column is of the same order as the average radius of gyration of the macromolecules of the mixture.

The pore size of the AC column does not much influence the separation pattern. As it follows from Equation (4), mostly a scale of the retention in the AC dimension is affected by the pore size of the AC column. Since the AC retention of both linear polymers and stars decreases with pore size, the use of the wide-pore AC columns may be helpful to reduce the time of the 2D-separation process.

In order to control the retention it is more convenient to use the adsorption interaction, which can be changed in practice by varying mobile phase composition or temperature. Figs. 6a, b and 5b show the effect of the adsorption interaction in AC on the separation by topology. Generally the strengthening of the adsorption interaction improves the topological separation. According to the theory, there is no separation of linear macromolecules and stars when the adsorption interaction is weak (when c is close to the critical point of adsorption, $c = 0$). It can be seen that the separation is still poor at $c = 0.05\ \mathrm{nm}^{-1}$ (Fig. 6a), but becomes quite good at $c = 0.12\ \mathrm{nm}^{-1}$ (Fig. 5b). However, the further increase of the adsorption interaction parameter does not much improve the separation pattern, mostly influencing a scale of the retention in the AC dimension (this can be seen by comparing Fig. 5b, $c = 0.12\ \mathrm{nm}^{-1}$, and Fig. 6c, $c = 0.2\ \mathrm{nm}^{-1}$). So, in the adsorption regime the effect of the adsorption interaction on the separation pattern is of little significance; therefore in practice in order to reduce the time of a separation process one can use gradient adsorption chromatography instead of AC at fixed interaction conditions. It can be expected that with gradient AC in the AC dimension one can obtain quite good results, provided that the interaction conditions within the gradient protocol are not too close to the critical point of adsorption.

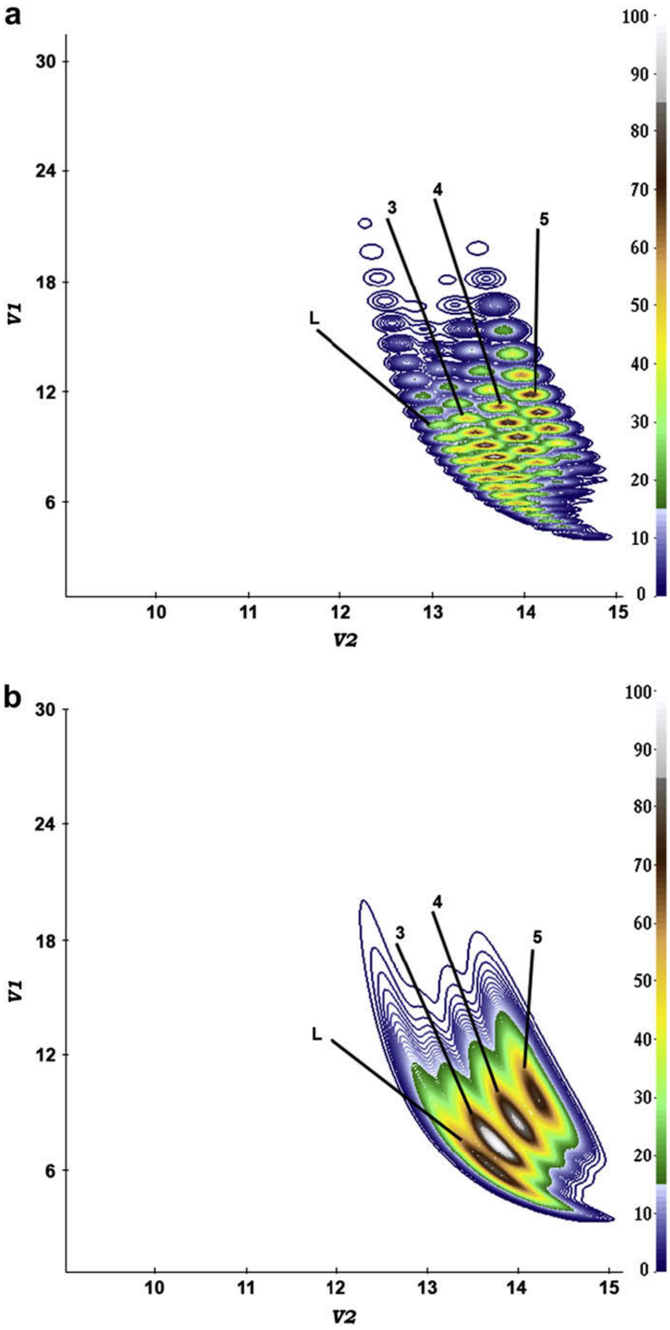

Fig. 3. Simulated two-dimensional AC-SEC chromatograms for the mixture of linear (L) and star (3, 4, 5 arms) polystyrenes of equal average molar mass (a) $M_w = 3000$, and (b) $M_w = 300\,000$. (a) Pore diameter of SEC and AC columns $2d = 5$ nm; interaction parameter in AC $c = 1\ \mathrm{nm}^{-1}$; (b) $2d = 50$ nm, $c = 0.1\ \mathrm{nm}^{-1}$. The other parameters are the same as in Fig. 2.

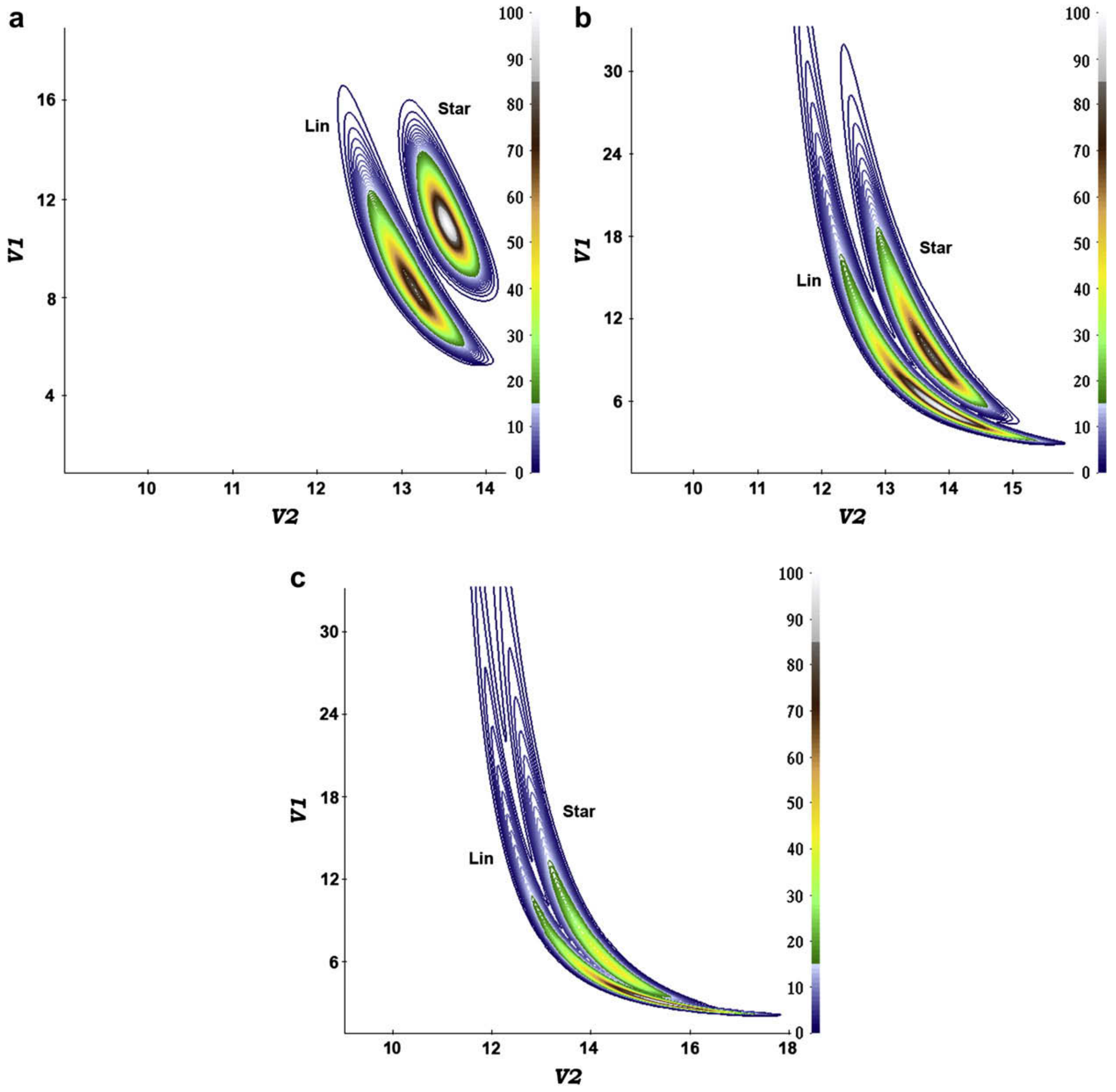

Fig. 4. (a–c) The effect of the molar-mass heterogeneity on the 2D-separation of linear polystyrenes and 4-arm stars of equal average molar mass 120 000. Pore diameter of SEC and AC columns 30 nm. Interaction parameter $c = 0.16\ \text{nm}^{-1}$. $(M_w/M_n)_{\text{arm}}$: (a) 1.01, (b) 1.1, and (c) 1.5. The other parameters are the same as in Fig. 2.

3.4. Asymmetric stars

Generally, the partition coefficient of a star in SEC and in AC regimes depends also on the asymmetry of a star macromolecule.

In particular, it can be drawn from the full theory that the coefficient B in Equation (3) for a three-arm star of total molar mass M (with arms of M_1, M_2 and M_3) in the wide-pore SEC regime has a form:

$$B = \frac{2}{\pi}\sum_{i=1}^{3}\sqrt{1-w_i}\,\mathrm{arctg}\left(\frac{\sqrt{w_1 w_2 w_3}}{w_i\sqrt{1-w_i}}\right) \qquad (5)$$

where $w_i = M_i/M$, $i = 1, 2, 3$. For very asymmetric stars $B \to 1$; at $w_1 = w_2 = w_3$ Equation (5) recovers the result for a symmetric 3-star, $B = 0.9598$. For stars with higher number of arms the effect is similar, and even more pronounced.

According to the theory, in the AC regime there is also some difference in the partition coefficient K of symmetric and asymmetric stars; in both SEC and AC $K_{\text{sym}} > K_{\text{asym}}$. Therefore the separation of such polymers by means of AC-SEC 2D-chromatography can be possible, in principle.

The above considered model star polymers (simulation results for which are given in Figs. 2–7) had arms of equal average molar mass, but each arm was distributed with respect to molar mass. This means that those star polymers were actually the mixtures of symmetric and asymmetric stars. Hence, the effect of asymmetry was also present in those chromatograms, manifesting itself in the form of broadening of the corresponding chromatographic zones.

In order to focus on the asymmetry effect, let us consider mixtures of symmetric and asymmetric star polymers having equal average molar mass and the same number of arms, but differing in the average asymmetry of arms. (Now we apply the terms 'symmetric' and 'asymmetric' to polydisperse star polymers in the sense that the average molar masses of their arms are the same or different). Since the effect of asymmetry of stars on their chromatographic behavior is quite delicate, a substantial separation of symmetric and asymmetric star polymers can only be expected when the molar-mass distributions of arms are narrow.

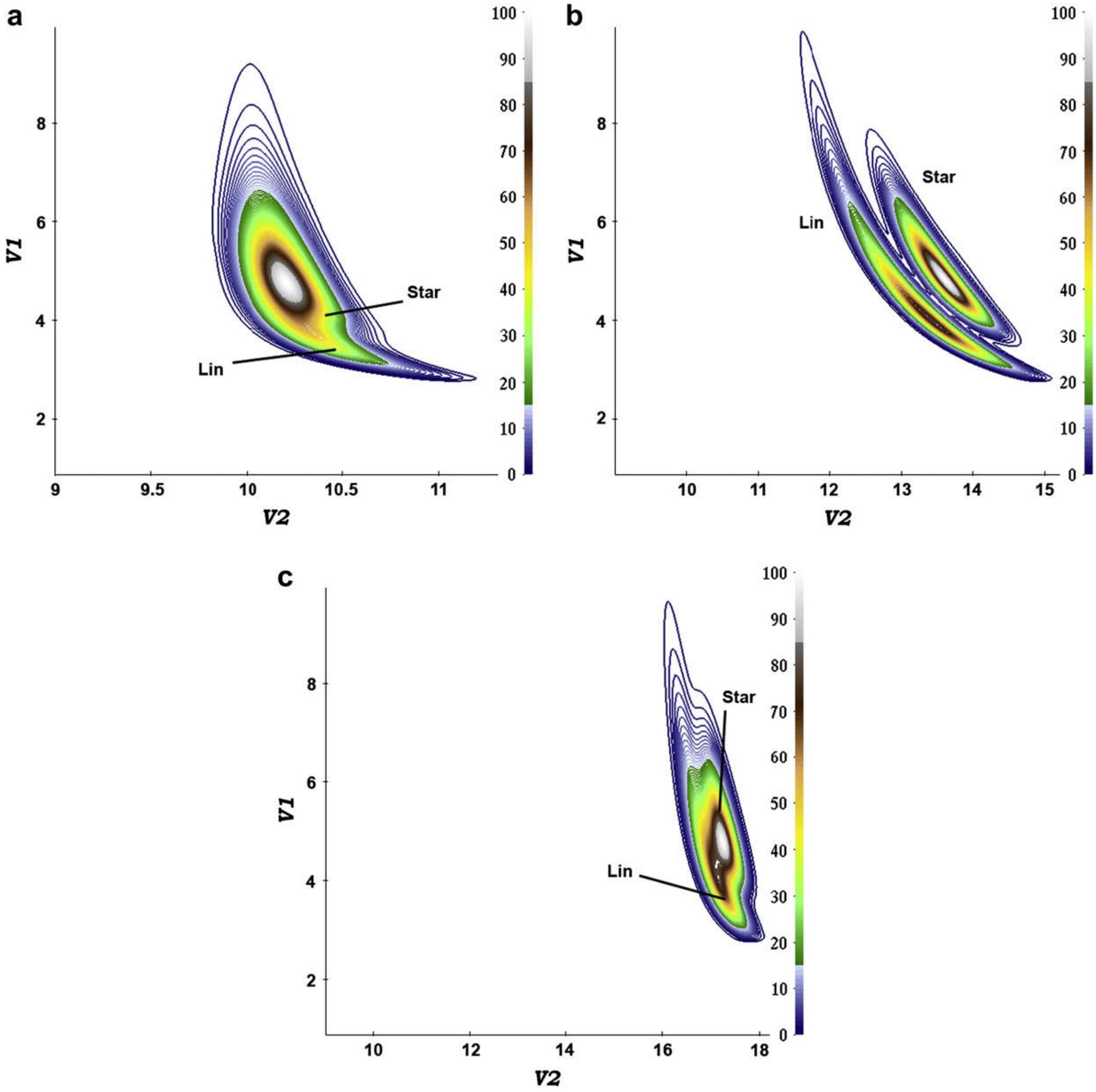

Fig. 5. Simulated 2D-chromatograms for the mixture of linear polystyrenes and 4-arm stars of average molar mass 120 000 and $(M_w/M_n)_{arm} = 1.05$ at various values of the SEC column pore diameter: (a) 15 nm, (b) 30 nm, and (c) 70 nm. $c = 0.12\ \text{nm}^{-1}$. The other parameters are as in Fig. 4.

Examples of theoretical 2D-chromatograms for mixtures of symmetric and asymmetric star polymers are presented in Fig. 7. Simulation shows that a considerable separation of star-shaped polymers by asymmetry occurs only if at least one arm in an asymmetric star polymer is very short. As it can be seen in Fig. 7a and b, the effect of separation by asymmetry in AC-SEC 2D-chromatography is more pronounced for stars with higher number of arms.

4. Concluding remarks

By using a theory of interactive chromatography of polymers, we have analysed the separation of polydisperse linear and star-shaped polymers by topology by means of the AC-SEC two-dimensional chromatography.

The molar mass distribution is an important factor that influences the separation. Simulation shows that linear and all different star components can be reasonably separated by 2D-chromatography, only if the heterogeneity index for the arms is less then $(M_w/M_n)_{arm} \approx 1.1$. Separation of linear polymers and stars having more than three arms is possible for much higher values of the heterogeneity index.

The pore size of the SEC column has a significant effect on the 2D-separation pattern. We conclude that the best results can be expected when the average pore radius of the SEC column is of the same order as the average radius of gyration of the macromolecules in the mixture. To the other hand, the pore size of the AC column does not much influence the separation pattern.

According to the theory and simulations, the effect of the adsorption interaction in the AC dimension on the separation pattern is of little significance; therefore in order to reduce the time of a separation process one can use gradient adsorption chromatography instead of AC at fixed interaction conditions. With gradient AC in the AC dimension one can expect reasonable separation results, only if the interaction conditions within the gradient protocol are not too close to the critical point of adsorption.

It is shown theoretically that the separation of symmetric and very asymmetric stars by AC-SEC 2D-chromatography can be also

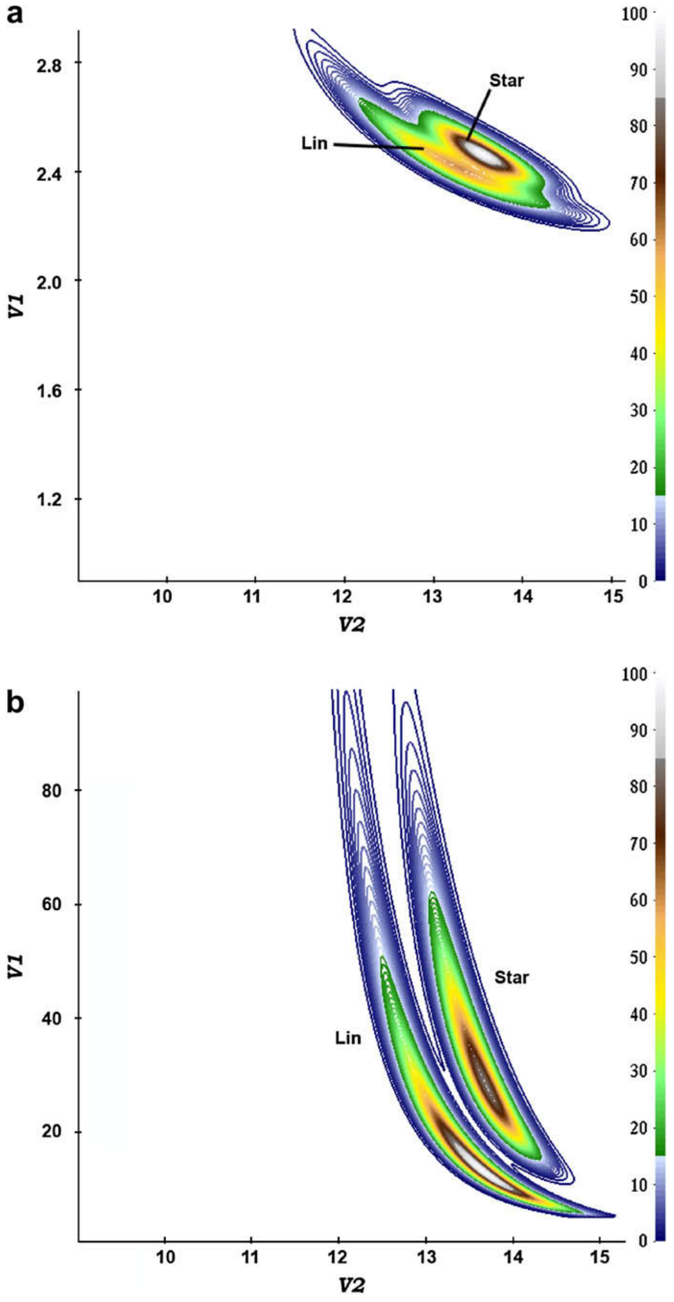

Fig. 6. The influence of adsorption interaction on the simulated 2D-chromatograms for the mixture of linear polystyrenes and 4-arm stars of average molar mass 120 000 and $(M_w/M_n)_{arm} = 1.05$. Interaction parameter: (a) $c = 0.05\ nm^{-1}$; (b) $c = 0.2\ nm^{-1}$ (see also Fig. 5b, $c = 0.12\ nm^{-1}$). The other parameters are the same as in Fig. 5.

possible. For stars with higher number of arms the effect of separation by asymmetry is expected to be more pronounced.

It should be noted, that the theory is based on the ideal-chain model which does not account for the excluded volume effects, and therefore it is applicable rather to the theta-solvent than to the good-solvent systems. In the systems where the excluded volume effects are expected to be important the chromatographic behavior of real polymers may be more complicated. In particular, in good solvents the deviations from the simplified ideal-chain theory can be expected in narrow-pore systems, and especially for stars with many arms which are strongly stretched due to the excluded volume effects. A complicated chromatographic behavior, which was attributed, in part, to the excluded volume effects, has been observed recently by Im et al. [5], who studied the retention of star-shaped polystyrenes near the chromatographic critical condition. The use of more sophisticated theoretical and simulation approaches [33–35], which take into the account the excluded volume effects, may bring the further insights into the chromatographic behavior of star polymers.

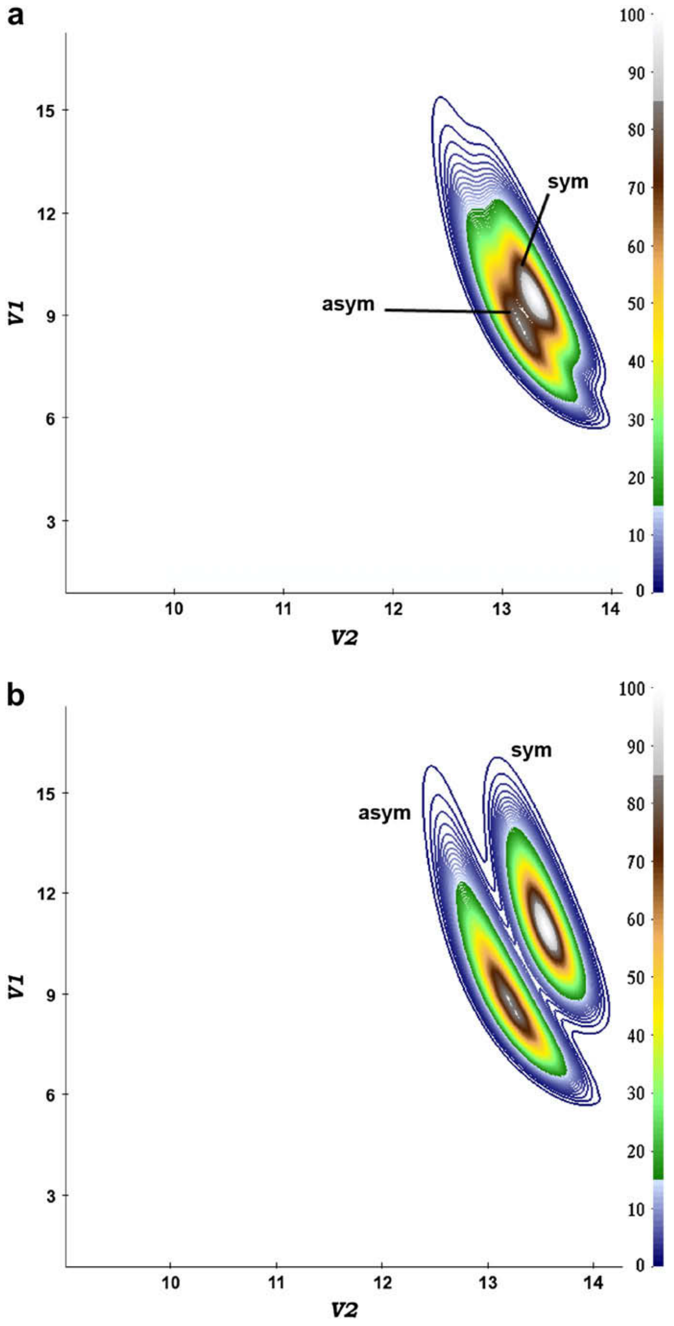

Fig. 7. Theoretical 2D-chromatograms for the mixture of symmetric and asymmetric three-armed (a) and four-armed (b) star polystyrenes. The average molar mass 120 000; $(M_w/M_n)_{arm} = 1.01$. M_w of arms of asymmetric stars: (a) 2000, 18 000 and 100 000; (b) 1000, 4000, 15 000 and 100 000. The other parameters are the same as in Fig. 4.

In this paper we consider mixtures of polydisperse linear polymers and different stars of the same average molar mass. As far as we know, such systems were not studied experimentally. However, the relevant experimental data on the off-line 2D-chromatography (with the temperature gradient adsorption chromatography in the first dimension, and SEC in the second one) of linear and star-shaped polystyrenes have been reported by Gerber and Radke [16]. In the experiments [16] the star sample was a mixture of stars with different number of equal arms; stars in this mixture were of different molar mass. Since our theoretical system and that of Ref. [16] are different, the simulated and real 2D-chromatograms cannot be directly compared, however the qualitative comparison of these experiments and the theory is possible. The experiments [16] resulted in clear 2D-separations of the linear and star-shaped structures, and it was shown that separation between the linear and star polymers of the same molar mass improves with increasing arm number. These experimental findings are in a good qualitative agreement with the present theory.

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References

[1] (a) Casassa EF. J Polym Sci Part B 1967;5:773;
(b) Casassa EF, Tagami Y. Macromolecules 1969;2:14.
[2] Teraoka I. Macromolecules 2004;37:6632.
[3] (a) Lee HC, Chang T, Harville S, Mays JW. Macromolecules 1998;31:690;
(b) Lee HC, Lee W, Chang T, Yoon JS, Frater DJ, Mays JW. Macromolecules 1998;31:4114;
(c) Ryu J, Im K, Yu W, Park J, Chang T, Lee K, et al. Macromolecules 2004;37:8805;
(d) Im K, Park S, Cho D, Chang T. Anal Chem 2004;76:2638.
[4] Chang T. J Polym Sci Part B Polym Phys 2005;43:1591.
[5] Im K, Park H-W, Kim Y, Ahn S, Chang T, Lee K, et al. Macromolecules 2008;41:3375.
[6] (a) Biela T, Duda A, Penczek S, Rode K, Pasch H. J Polym Sci Part A Polym Chem 2002;40:2884;
(b) Biela T, Duda A, Rode K, Pasch H. Polymer 2003;44:1851.
[7] Im K, Kim Y, Chang T, Lee K, Choi N. J Chromatogr A 2006;1103:235.
[8] Pasch H, Trathnigg B. HPLC of polymers. Berlin, Heidelberg, NY: Springer-Verlag; 1997.
[9] Pasch H. Macromol Symp 2002;178:25.
[10] Jiang XL, van der Horst A, Lima V, Schoenmakers PJ. J Chromatogr A 2005;1076:51.
[11] (a) Skvortsov AM, Gorbunov AA. Vysokomol Soedin Ser A 1980;22:2641;
(b) Skvortsov AM, Gorbunov AA. J Chromatogr A 1990;507:487;
(c) Entelis SG, Evreinov VV, Gorshkov AV. Adv Polym Sci 1986;76:129;
(d) Gorbunov A, Trathnigg B. J Chromatogr A 2002;955:9;
(e) Skvortsov AM, Fleer GJ. Macromolecules 2002;35:8609.
[12] Gorbunov AA, Skvortsov AM. Adv Colloids Interface Sci 1995;62:31.
[13] Gorbunov AA, Vakhrushev AV. Polymer 2004;45:7303.
[14] Radke W, Rode K, Gorshkov AV, Biela T. Polymer 2005;46:5456.
[15] Gorbunov AA, Vakhrushev AV. J Chromatogr A 2005;1064:169.
[16] Gerber J, Radke W. Polymer 2005;46:9224.
[17] Trathnigg B, Gorbunov A, Skvortsov A. J Chromatogr A 2000;890:195.
[18] (a) Trathnigg B, Gorbunov A. J Chromatogr A 2001;910:207;
(b) Trathnigg B, Rappel C, Raml R, Gorbunov A. J Chromatogr A 2002;953:89;
(c) Rappel C, Trathnigg B, Gorbunov A. J Chromatogr A 2003;984:29.
[19] Gorbunov AA, Vakhrushev AV. Polymer 2004;45:6761.
[20] Vakhrushev AV, Gorbunov AA, Tezuka Y, Tsuchitani A, Oike H. Anal Chem 2008;80:8153.
[21] (a) Gorbunov AA, Skvortsov AM. Vysokomol Soedin Ser A 1986;28:2170;
(b) Gorbunov AA, Skvortsov AM. Vysokomol Soedin Ser A 1986;28:2453.
[22] Kosmas M, Kokkinos I, Bokaris EP. Macromolecules 2001;34:7537.
[23] (a) Eisenriegler E, Kremer K, Binder K. J Chem Phys 1982;77:6296;
(b) Lepine Y, Caille A. Can J Phys 1978;56:403.
[24] (a) DiMarzio EA, Rubin R. J Chem Phys 1971;55:4318;
(b) DiMarzio EA, Guttman CM, Mah A. Macromolecules 1995;28:2930;
(c) Guttman CM, DiMarzio EA, Douglas JF. Macromolecules 1996;29:5723.
[25] Fleer GJ, Scheutjens JMHM, Cohen Stuart MA, Cosgrove T, Vincent B. Polymers at interfaces. London: Chapman & Hall; 1993.
[26] Gorbunov AA, Skvortsov AM, van Male J, Fleer GJ. J Chem Phys 2001;114:5366.
[27] de Gennes PG. Rep Prog Phys 1969;32:187.
[28] Pasch H, Brinkmann C, Much H, Just U. J Chromatogr 1992;623:315.
[29] Kilz P, Krüger R-P, Much H, Schulz G. In: Provder T, Barth HG, Urban MW, editors. Chromatographic characterization of polymers: hyphenated and multidimensional techniques. Advances in chemistry series, vol. 247. Washington, DC: American Chemical Society; 1995. p. 223–42.
[30] Kilz P. Labor Praxis 1992;6:628.
[31] Kilz P, Krüger R-P, Much H, Schulz G. Polym Mater Sci Eng 1993;69:114.
[32] van der Horst A, Schoenmakers PJ. J Chromatogr A 2003;1000:693.
[33] Joanny J-F, Johner A. J Phys II France 1996;6:511.
[34] Cifra P, Bleha T. Polymer 2000;41:1003.
[35] Chen Z, Escobedo FA. Macromolecules 2001;34:8802.