



ELSEVIER

Journal of Chromatography A, 791 (1997) 350–356

JOURNAL OF
CHROMATOGRAPHY A

Short communication

Liquid chromatography of polyisoprenes: determination of critical composition using non protic binary and acidic ternary solvent mixtures

Gunther Czichocki^a, Robert Heger^a, Werner A. Goedel^{a,*}, Helmut Much^b

^aMax-Planck-Institut für Kolloid- und Grenzflächenforschung, Rudower Chaussee 5, 12489 Berlin, Germany

^bInstitut für Angewandte Chemie Adlershof, Rudower Chaussee 5, 12489 Berlin, Germany

Received 2 April 1997; received in revised form 9 July 1997; accepted 9 July 1997

Abstract

Mixed solvent systems for the critical liquid chromatography of polyisoprenes are investigated. Under critical solvent conditions, the energetic and entropic interactions between the polymer and the stationary phase cancel each other, the free energy of interaction approaches zero and the retention time is independent of the polymer chain length. Critical conditions for polyisoprene–silica gel have been established for a non-protic binary solvent mixture [*i*-octane–tetrahydrofuran (THF); critical composition at 0.88% (v/v) of THF] and a ternary system containing an acidic component [*i*-octane–[0.5% (v/v) acetic acid in methyl-*tert*.butylether (MTBE)]; critical composition at 0.65% (v/v) of the acetic acid–MTBE mixture]. © 1997 Elsevier Science B.V.

Keywords: Mobile phase composition; Polyisoprenes

1. Introduction

Liquid chromatography is a powerful tool for analysing and purifying polydisperse and/or chemically heterogeneous polymer samples. Synthetic polymers usually do not have uniform chain lengths. In addition, the chains of copolymers may be heterogeneous in composition or the reactions carried out to generate special functional end-groups of telechelics/macromonomers might be incomplete.

In the well-established characterisation of molecular mass distribution via size-exclusion chromatography (SEC), the chromatographic separation is caused by entropic effects. The polymer does not

adsorb to the stationary phase, but partitions between the bulk of a good solvent and solvent-filled small pores. The Gibbs free energy of interaction between polymer and the stationary phase is positive and increases with chain length. Large polymers are eluted earlier than low-molecular-mass substances.

Liquid adsorption chromatography (LAC) has been established for the determination of the composition of copolymers and the determination of end-group functionality [1]. LAC is based on adsorption of the polymer due to the fact that the Gibbs free energy of interaction between the polymer and the stationary phase is lower than the Gibbs free energy of interaction between the polymer and the liquid phase. This free energy of interaction decreases monotonically with increasing chain length

*Corresponding author.

or increasing content of adsorbing groups. Therefore, the longer the chain or the higher the amount of adsorbing groups that are incorporated in the chain, the longer it takes for the polymer to be eluted from the column.

Problems often arise if both processes take place simultaneously: (i) non-polar polymers with strongly polar end groups tend to adsorb to the stationary phase in standard SEC; (ii) identification of end groups via LAC is hindered by a broadening of the peaks due to size exclusion of the polymer chains of non-uniform chain length [2].

The recently established method of liquid adsorption chromatography under critical solvent conditions (CSC) solves the latter problem. It is based on the observation that, under certain conditions, the decrease in entropy of the chain due to the interaction with the stationary phase is compensated by an equivalent adsorption energy. In this case, the Gibbs free energy of interaction between the polymer chain and the stationary phase is zero and the polymer chain length has no influence on the retention. However, the retention is affected by e.g. strongly adsorbing heterogeneous groups. Thus, chromatography under CSC can give valuable information on the chemical heterogeneity of polydisperse polymer samples and can be used for the preparative purification of polymers with defined end groups. In addition, a polymer can be completely characterised using 2D chromatography: the polymer is first separated by functionality using the CSC mode and then the polydispersity of each fraction is evaluated by SEC [3].

Preliminary results have shown that critical conditions can be realised using solvent mixtures as a mobile phase [1,4–8]. For example, chromatography

at CSC of OH-functionalised oligo-isoprenes (M_n 900 g mol⁻¹) has been performed in the chromatographic system CCl₄–CHCl₃–C₂H₅OH on silica columns [9,10]. Because any change in solvent composition leads to a deviation from critical conditions, it is usually difficult to optimise the CSC mode with respect to the adsorbing functional groups. However, it should, in principle, be feasible to establish a set of different critical solvent mixtures and then to select the mixture that is most suitable for the discrimination between the adsorbing groups.

The goal of this contribution is to establish more than one critical solvent composition for the chromatography of polyisoprenes and to supplement and/or substitute the above-mentioned critical solvent composition. Bearing in mind the optimisation of the discrimination between functional groups, these mixtures should differ in character. We consider, as an example, the separation of protic and/or acidic adsorbing groups. Therefore, we develop two critical solvent mixtures, one is aprotic while the second one has a protic and acidic component.

2. Experimental

Polyisoprene (stabilised with 0.1% 2,6-di-*tert.*-butyl-4-methyl phenol) and polystyrene standards were obtained from PSS (Mainz, Germany) and used as received. The characterisation according to the suppliers' data sheets is given in Table 1.

Tetrahydrofuran (THF) and *i*-octane (=2,2,4-trimethylpentane) (Sigma–Aldrich, HPLC grade), methyl-*tert.*butyl ether (MTBE; Lab-Scan) and glacial acetic acid (Merck) were used as received.

Table 1
Characterization of standard polyisoprene-1,4 (PI) and polystyrene (PS).

Abbreviated name	M_n /kg mol ⁻¹	M_w /kg mol ⁻¹	M_p /kg mol ⁻¹	M_w/M_n
PI-0.5	0.56	0.615	0.60	1.10
PI-4.5	4.5	4.7	4.7	1.04
PI-10.5	10.5	10.8	10.9	1.02
PI-15	15.3	15.6	16.0	1.02
PI-39	39.0	40.5	42.0	1.03
PI-60	60.0	62.5	64.0	1.04
PI-98	98.2	99.8	102.0	1.02
PS	–	–	1400	1.05

The water content of the THF (<0.004%) was checked via Karl–Fischer titration.

Investigations were carried out on a JASCO liquid chromatograph equipped with two PU-987 preparative HPLC pumps (50 ml), an AS-950 autosampler, an RH-7125 injecting valve (Rheodyne), a CO-200 Peltier column thermostat, a Sedex 55 evaporative light scattering detector (ERC), an 830-RI refractive index detector, an SF 2120 fraction collector (Advantec) and a NINA data system II (Nuclear interface).

The column (250×4.6 mm I.D.) was packed with silica (YMC-Pack SIL AP, 200 Å mean pore diameter, 5 µm mean particle diameter, YMC Europe).

Solutions of the samples (1–50 µl) were injected into the chromatographic system with an automatic sampler. The elution time of toluene was determined by injecting 10 µl of pure toluene. Chromatographic separations were carried out at 308 K. The flow-rate was 0.5 or 1 ml/min.

A refractive index detector and an evaporative light scattering detector (temperature, 323 K; inert gas, N₂; pressure, 2 bar) were used.

3. Results and discussion

3.1. Characterisation of the column

The mean pore diameter of the silica packing material was selected in order not to completely exclude the polyisoprene molecules considered in Table 1. The interparticle or exclusion volume of the silica column, v_i , was determined by elution of a polystyrene sample of 1400 kg mol⁻¹, which is completely excluded from the pores of the packing material. The dead volume, v_o , was determined by elution of a toluene sample. In both cases, THF was used as the eluent. The pore volume (difference between dead volume and interparticle volume) was evaluated according to Eq. (1):

$$v_p = v_o - v_i. \quad (1)$$

Fig. 1 shows the liquid chromatography of 1.4 · 10⁶ g mol⁻¹ polystyrene (Fig. 1a) and of toluene (Fig. 1b). The evaporative light scattering detector is rather insensitive to toluene (under optimised conditions, it is able to detect toluene just above the noise level).

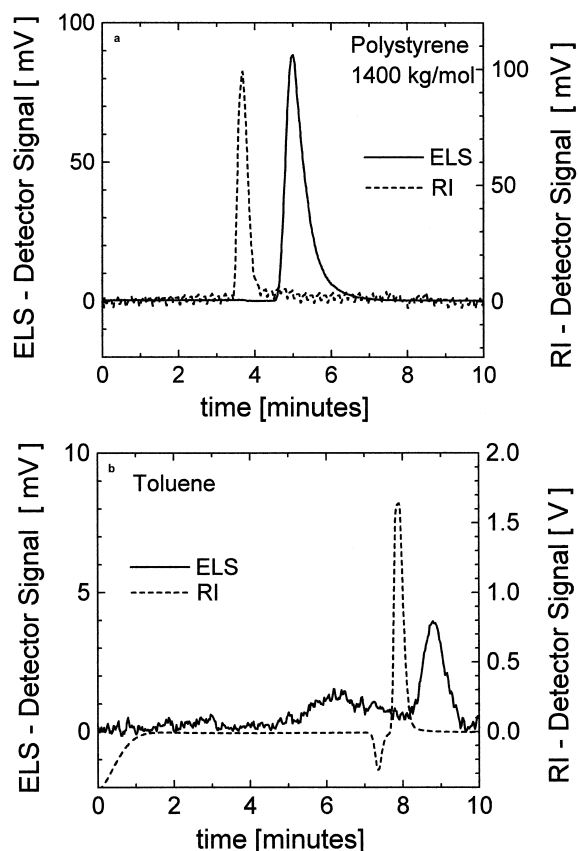


Fig. 1. Experimental determination of (a) the interparticle volume, v_i , via elution of polystyrene ($M_n = 1.4 \cdot 10^6$ g mol⁻¹) and (b) the dead volume, v_o , via elution of toluene. RI=differential refractive index detection; ELS=evaporative and light scattering detection. Solvent: tetrahydrofuran. For other conditions, see Fig. 2.

Therefore, the characterisation was made using two detectors; a refractive index (RI) detector and an evaporative light scattering detector (ELSD). The two detectors are mounted in-line. As a result, a time lag occurs between the two detectors. Negative peaks detected by the differential refractometer in the chromatogram of toluene most likely are due to preferential solvation effects [2].

Characterisation of the column is summarised in Table 2. The interparticle volume and the exclusion volume as given in Table 2 include the volume of the tubing to the detectors and, therefore, are larger than the actual characteristics of the column. In the calculation of v_p and K_D , these effects should cancel, because these values are based on differences be-

Table 2

Experimental values of v_i , v_o and v_p at a flow-rate of 0.5 ml/min

	Differential refractive index detection (RI)	Evaporative light scattering detection (ELS)
v_i (Polystyrene) (ml)	1.84	2.49
v_o (Toluene) (ml)	3.93	4.42
$v_p = v_o - v_i$ (ml)	2.09	1.93

tween two elution volumes. Because of the difference between the two values of v_p calculated from the two detector signals, we estimate that the K_D values given below have an error of approximately 8%. In the experiments below, the refractive index detector was removed and the evaporative light scattering detector was mounted in its place. The additional error introduced by this procedure is usually less than the estimated uncertainty in the K_D of 8%.

3.2. Determination of critical compositions

In order to affect the retention time, the energetic interactions between the stationary phase and the functional groups should be negative and dominate the enthalpy of interaction. Since the functional groups attached to polyisoprenes are usually more polar than the polymer backbone [11], polar non-derivatised silica was chosen as a stationary phase.

Critical solvent conditions for the chromatography of polyisoprenes have already been reported using halogenated solvents [9,10]. Out of environmental concern, we evaluate only solvent mixtures that are composed of non-halogenated solvents.

3.3. THF-*i*-octane system

As a first solvent system, we used a mixture of *i*-octane (as the non-polar component) and THF (as the polar component). *i*-Octane was used because it is less hygroscopic than *n*-alkanes and, hence, it is easier to obtain reproducible conditions.

In Fig. 2, the retention times of polyisoprene samples using a variety of solvent compositions are plotted as a function of molar mass. In order to avoid irreversible adsorption of the polymer in pure *i*-octane, the measurements were started with THF-rich mixtures and the THF content was successively

diminished from run to run. For binary mixtures of *i*-octane and THF, containing more than 0.88% THF, the retention time decreases with increasing molecular mass; in this concentration regime, separation takes place in the SEC mode. If the THF content in the mobile phase is reduced, differences in the elution volumes of samples of different molar mass

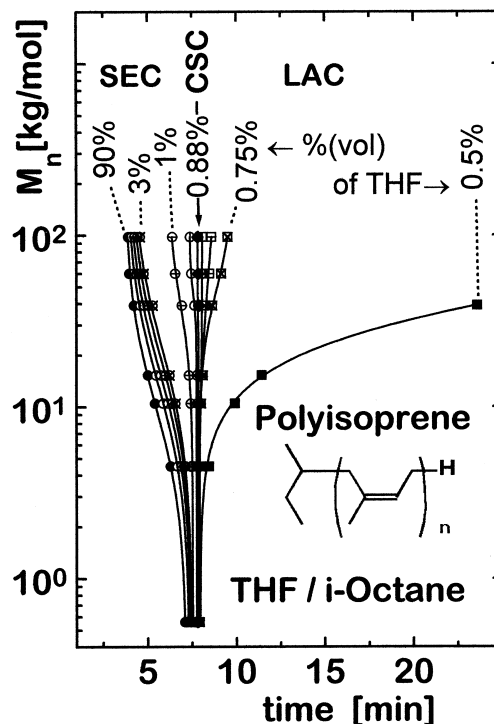


Fig. 2. Correlation between the chromatographic retention time and the molar mass (M_n) of polyisoprene-1,4 using mixtures of THF and *i*-octane. Composition [% (vol.) of THF]: 90, 20, 10, 5, 3, 1, 0.9, 0.88, 0.85, 0.80, 0.75 and 0.5%. Conditions: column, YMC-Pack SIL AP, 200 Å, 5 μm, 250×4.6 mm I.D.; flow-rate, 0.5 ml/min; column temperature, 308 K; detector temperature, 323 K; N_2 pressure, 2 bar; RI detector, removed; ELS detector, mounted at the position that was occupied by the RI-detector during characterisation of the column in Fig. 1.

become smaller. At a composition of 0.88% THF, all polyisoprene samples were eluted at the same retention time (Fig. 2). Thus, this composition can be identified as the CSC. For compositions containing less than 0.88% THF, we observe increasing retention times with increasing molecular mass; in this concentration regime, separation takes place in the LAC mode.

The determination of the critical composition is illustrated in a slightly different representation in Fig. 3. The retention in chromatography is defined by the distribution coefficient, K_D , associated with a change in the Gibbs' free energy of interaction of the polymer, ΔG [5]:

$$K_D = \frac{v_e - v_i}{v_p} = \exp\left(-\frac{\Delta G}{k_B T}\right) \quad (2)$$

with v_e =elution volume of the polymer, v_i =interparticle volume, v_p =pore volume, k_B =Boltzmann constant and T =temperature.

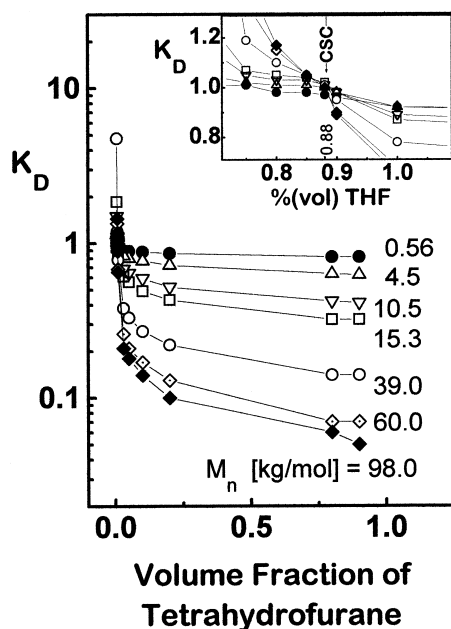


Fig. 3. Determination of the critical solvent composition from the dependence of K_D on the volume fraction of the mixed mobile phase. For conditions, see Fig. 2. The inset represents the part of the diagram that is close to the critical conditions. As expected, the data for different molecular mass polymers intersect at the critical conditions at a K_D value of one.

The free energy can be represented by an energetic and an entropic contribution:

$$\Delta G = \Delta H - T\Delta S \quad (3)$$

In a good solvent (e.g. THF), the energetic contribution is either positive or negligible; the negative entropic contribution (ΔS) dominates; the net free energy is positive and therefore $K_D < 1$. In the adsorption mode, a large negative energetic contribution dominates the free energy of interaction, ΔG is negative and, hence, $K_D > 1$. In Fig. 3, the K_D values of the polyisoprene samples are plotted as a function of solvent composition. As expected, all lines intersect at a K_D value that is close to unity ($\Delta G = 0$). This representation reduces the time needed for the determination of the CSC, because CSC can be estimated by interpolation of the data obtained under non-critical conditions.

The above considerations depend on the assumption that the entropic and the energetic contribution depend in the same way on the chain length (e.g. linear). If this is not the case, the critical conditions can only be approximately valid for a limited range of polymer chain lengths. Hence, the CSC should be verified with a range of standards, especially of higher molar mass.

To illustrate the different separation modes, liquid chromatography has been performed using a mixture of the polymers PI-0.5, PI-10.5 and PI-39. Under critical conditions, the mixture of polyisoprene standards is eluted as one narrow peak (Fig. 4b). In the SEC mode (Fig. 4a) and in the LAC mode (Fig. 4c), the polymers are separated by molecular mass. The molecular mass distribution, in principle, can be obtained in the SEC as well as in the LAC mode. However, the order of elution of the polymers is inverted, when switching from one mode to the other.

3.4. MTBE–acetic acid–i-octane system

In the preceding section, it was shown that critical conditions can be established by mixing an alkane with an ether. However, critical conditions occurred at a comparatively low content of the polar ether component. The aim of this study was to establish critical conditions using an acidic component, which

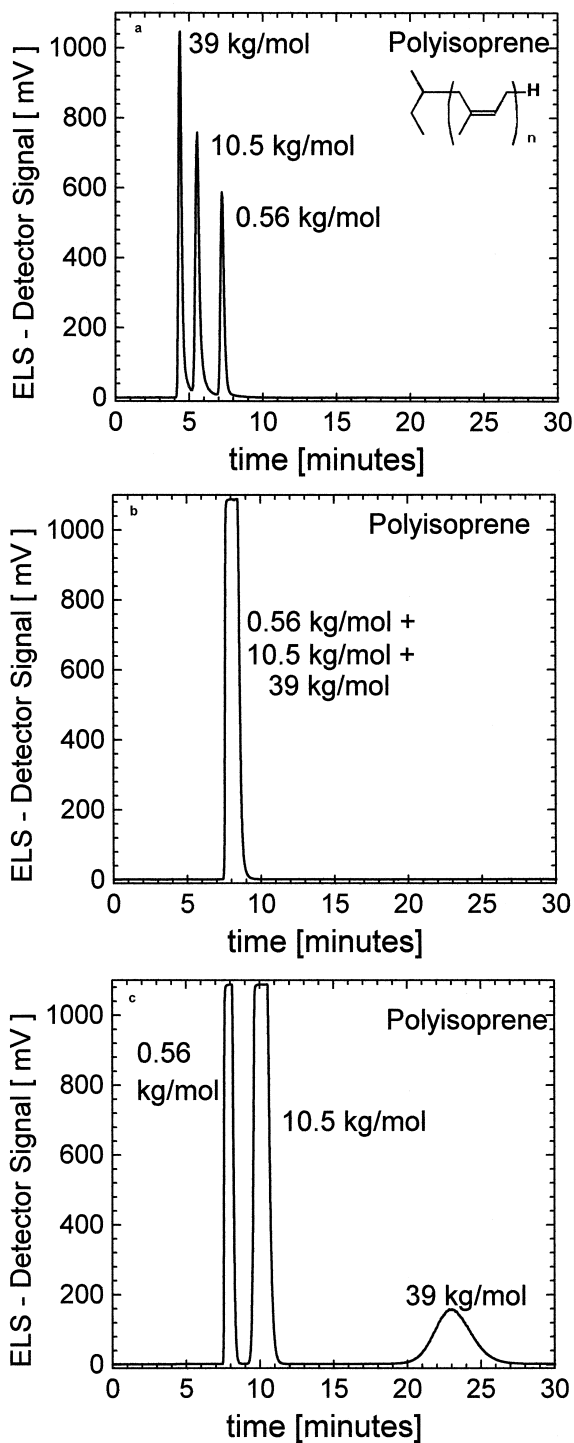


Fig. 4. Separation of a mixture of three polyisoprene-1,4 standards ($M_n = 0.56, 10.5$ and 39 kg/mol) using the following mixing ratios of THF-*i*-octane: (a) SEC mode (90:10, v/v), (b) CSC mode (0.88:99.12, v/v) and (c) LAC mode (0.50:99.50, v/v).

should depress the association and adsorption of polar and acidic end groups. This acidic component is more polar than THF. One might expect that the addition of an acid to the polar component would drastically shift the critical composition towards an even lower ether content. Therefore, we replaced the THF by the less polar MTBE and chose a ternary system composed of *i*-octane-acetic acid-MTBE. A further advantage of this system is given by the better UV cut-off of MTBE compared to that of THF. Acetic acid may strongly adsorb to the stationary phase and change the adsorption characteristics by competing with the polar groups of the polymers. In order to have stable conditions, the column was washed and equilibrated with the corresponding solvent mixture for at least 10 min before and after each separation. Reproducibility was checked with several consecutive runs.

The critical conditions for polyisoprene samples using mixtures of *i*-octane and a 0.5% solution of

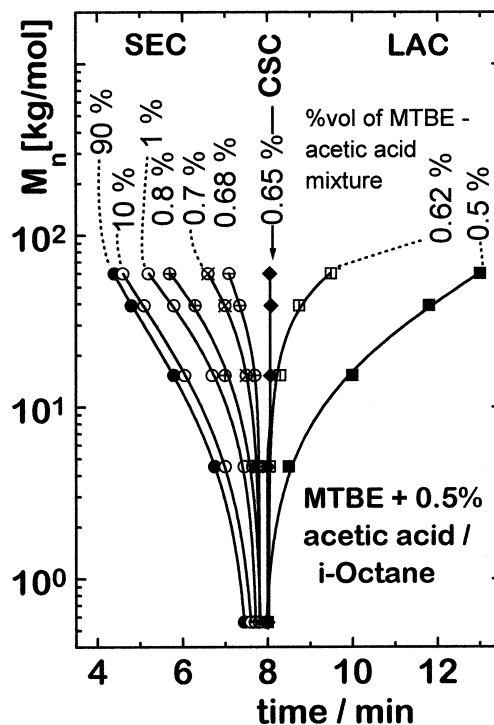


Fig. 5. Correlation between the chromatographic retention time and the molar mass (M_n) of polyisoprene-1,4 using mixtures of *i*-octane and a 0.5% (v/v) solution of acetic acid in methyl-*tert*-butyl ether. For conditions, see Fig. 2.

acetic acid in MTBE are evaluated the same way as in Section 3.3. As can be seen from Fig. 5, the critical composition is at 99.35% *i*-octane and 0.65% of a 0.5% (v/v) solution of acetic acid in MTBE.

4. Conclusions

Critical conditions for chromatographic investigations of polyisoprenes can be established for mixtures of non-halogenated alkanes and ethers. Two different critical solvent compositions have been found (non-protic/acidic). Both systems give the same retention time for non-functionalised polyisoprenes, independent of molecular mass. Because of the different natures of the systems (non-protic vs. acidic) we expect, however, that the elution characteristics of polyisoprenes with polar groups will be different. Hence, these two systems may be utilised to optimise the chromatographic analysis of polyisoprenes with polar groups.

Investigations on functionalised polyisoprenes are under way.

Acknowledgements

The authors wish to thank P. Mueller and A. Martins for their helpful cooperation.

References

- [1] H. Pasch, H. Much, G. Schulz, Trends Polymer Sci. 3 (1993) 643.
- [2] B. Trathnigg, Prog. Polym. Sci. 20 (1995) 615.
- [3] R.-P. Krüger, H. Much, G. Schulz, J. Liq. Chromatogr. 17 (1994) 3069.
- [4] B.G. Belenki, E.S. Gankina, M.B. Tennikov, L.Z. Vilenchik, J. Chromatogr. 123 (1990) 91.
- [5] S.G. Entelis, V.V. Evreinov, A.V. Gorshkov, Adv. Polym. Sci. 76 (1986) 129.
- [6] A.V. Gorshkov, H. Much, H. Becker, H. Pasch, V.V. Envirov, S.G. Entelis, J. Chromatogr. 523 (1990) 91.
- [7] G. Schulz, H. Much, H. Krüger, C. Wehrstedt, J. Liq. Chromatogr. 13 (1990) 1745.
- [8] H. Pasch, H. Much, G. Schulz, A.V. Gorshkov, LC·GC Int. 5 (1992) 38.
- [9] I.V. Valuev, Metody analiza i kontrolja Kach. Prod. Khim. Prom-sh., Moscow NIITEK h IM, 1978, p. 24.
- [10] S. Pokorny, J. Liq. Chromatogr. 4 (1981) 1.
- [11] R. Heger, W.A. Goedel, Macromolecules 29 (1996) 8912.