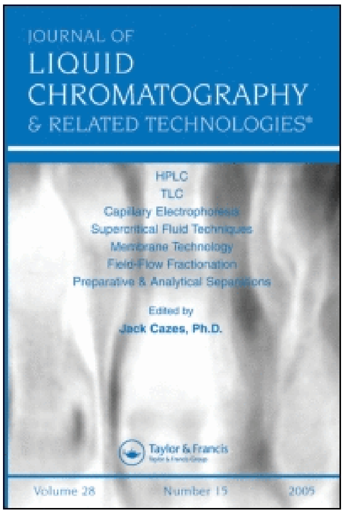

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CHARACTERIZATION OF POLYBUTYLACRYLATE-B-POLYVINYLPYRIDINE BLOCK COPOLYMERS BY SIZE-EXCLUSION CHROMATOGRAPHY AND DUAL REFRACTIVE INDEX/UV-DETECTION

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CHARACTERIZATION OF POLYBUTYLACRYLATE-B-POLYVINYLPYRIDINE BLOCK COPOLYMERS BY SIZE-EXCLUSION CHROMATOGRAPHY AND DUAL REFRACTIVE INDEX/UV-DETECTION

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☐ *An efficient and reliable size exclusion chromatographic (SEC) method has been developed allowing satisfactory insight into distribution of both molecular weight of polybutylacrylate-b-polyvinylpyridine (PBA-b-PVP) block copolymers as well as the relative amount of polyvinylpyridine (PVP) versus polybutylacrylate (PBA) along the molecular weight axis. This aim was accomplished by SEC using a rather new polyester-based GRAM stationary phase support of intermediate polarity and a polar eluent system of N,N-dimethylacetamide containing small amounts of lithium bromide and acetic acid for extensive suppression of adsorptive solute-stationary phase interactions.*

INTRODUCTION

As shown in Figure 1 polybutylacrylate-b-polyvinylpyridine (PBA-b-PVP) block copolymers belong to the family of polymers with intermediate polarity possessing both hydrophobic polybutylacrylate (PBA) as well as relatively hydrophilic polyvinylpyridine (PVP) moieties. Depending on the specific application, the block length of the PBA block needs to be substantially longer than the PVP block. Nevertheless, in order to obtain tailor-made PBA-b-PVP block copolymers for specific applications, the relative block length of PBA vs. PVP had to be more or less varied in order to achieve a fine-tuned balance between either hydrophobic or more polar electrosteric interactions.

Due to the fact that the copolymers of interest contain basic PVP blocks, strong interactions with the commonly available stationary phases for size

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This paper is dedicated to Professor Gottfried Schill on the occasion of his 80th birthday.

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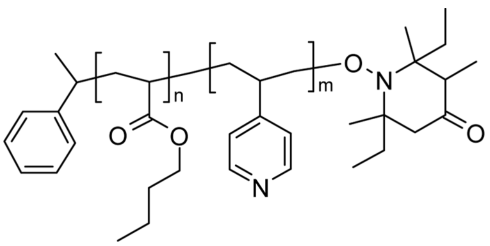

FIGURE 1 Structural formula of PBA-b-PVP block copolymer.

exclusion chromatography (SEC) had to be taken into account. Mainly in polar organic and aqueous SEC, these comprise intermolecular electrostatic interactions (ion-exchange, ion exclusion) between macromolecules and the stationary phase, as well as intra-molecular interactions and adsorption between polymer and stationary phase caused by hydrogen bonding and, furthermore, also by hydrophobic interactions.[1–6] For this reason, it seems obvious that separation according to molecular size can only be achieved by more or less complete suppression of these unwanted secondary effects as described by Mori in the case of SEC of sodium polystyrene sulfonate compounds.[6] This can be done by, e.g., variation of the mobile phase, addition of an electrolyte, and/or an organic modifier to the mobile phase. As a logical consequence, SEC of both cationic and anionic polymers still offers a challenge to the chemist involved in characterization of polymer species of this kind.

Relatively few works has been published on SEC of polyacrylate-based polymers bearing both cationic and anionic functionalities. In 2003, Wittgren et al.[7] published a paper dealing with molecular mass characterization of cationic methyl methacrylate-ethylacrylate copolymers containing up to 10% quaternary ammonium groups by SEC coupled to multi-angle light scattering (MALS) and refractometric detection using mobile phases of ethanol (95%) containing various amounts of lithium chloride as the electrolyte. However, when increasing the salt concentration too much, a concomitant increase of hydrophobic, i.e., adsorptive interactions were observed. Adler et al.[8,9] reported on molar mass characterization of neutral, anionic and cationic poly(meth)acrylate copolymers on a novel polyester-based GRAM polymer-based packing specially designed for polymer solutes with a wide range of polarity. *N,N*-dimethylacetamide containing electrolytes such as lithium bromide and TRIS, as well as acetic acid, was used as the mobile phase. Ringena et al.[10] used mobile phases consisting of either *N,N*-dimethylsulfoxide – water or *N,N*-dimethylacetamide, to which either lithium bromide or lithium chloride as the electrolytes was added and either PFG-PRO columns bearing free hydroxyl groups or GRAM stationary phases without free hydroxyl groups for SEC of other anionic polymers, such as technical lignin sulfonate samples.

The aim of the present study was to characterize PBA-b-PVP block copolymers by use of a robust and reproducible SEC method, which is able to provide insight into distribution of molecular weight as well as the relative amount of PVP versus PBA along the molecular weight axis. In order to achieve this aim, a SEC technique employing RI/UV-dual detection was chosen.

EXPERIMENTAL

Materials

Lithium bromide, acetic acid, and N,N-dimethylacetamide (all analytical-reagent grade) for mobile phase preparation was from Fluka (Buchs, Switzerland). For conventional calibration PMMA calibration standards were obtained from Polymer Laboratories (Church Stretton, UK) and a set of six molecular weight calibrators was used comprising PMMA of $M_p = 1140$ Da ($M_w/M_n = 1.16$), $M_p = 2680$ Da ($M_w/M_n = 1.09$), $M_p = 5200$ Da ($M_w/M_n = 1.06$), $M_p = 10260$ Da ($M_w/M_n = 1.04$), $M_p = 30650$ Da ($M_w/M_n = 1.02$), and $M_p = 60150$ Da ($M_w/M_n = 1.03$). Polystyrene 100'000 Da ($M_w/M_n = 1{,}02$, dn/dc = 0,185 mL/g, dA/dc = 1 mL/g, intrinsic viscosity (IV) = 0,3693 dL/g), also obtained from Polymer Laboratories (Church Stretton, UK) was used for calibration of the RI-, UV-, LALS/RALS (low angle light scattering/right angle light scattering) and DP (differential pressure = measure of viscosity) detectors. The PBA-b-PVP block copolymer samples to be investigated as well as the PBA and PVP starting materials WS 05023 and NCDA 05011, respectively, were synthesized in the polymer chemistry department of Ciba Inc. (Basel, Switzerland). The GRAM stationary phase system of intermediate polarity composed of a novel polyester copolymer specially designed for SEC of polymers with intermediate polarity including charged species obtained from Polymer Standard Services (Mainz, Germany) consisted of two GRAM 100 columns (300 × 8 mm I. D., 10 µm particle size, 100 Å pore size) covering a separation range between 600 and 60000 Da and a corresponding GRAM pre-column (50 × 8 mm I. D., 10 µm particle size).

Analytical Equipment

The chromatographic system consisted of a model GPC_{max} apparatus comprising isocratic pump, autosampler, and degassing unit in a common housing; a model TDA 302 triple detector array instrument comprising refractive index (RI), differential pressure (DP), low angle (7°) light scattering (LALS), and right angle (90°) light scattering (RALS) detection with an integrated column oven all housed together; and a model 2501 variable wavelength UV-detector, all obtained from Viscotek (Houston, TX, USA). The sample loop permitted injection of fixed sample amounts. For UV-detection a wavelength of 270 nm was chosen, whereas the LASER wavelength for LALS- and RALS-detection was 670 nm. The volumes of the RI-, and LALS/RALS measuring cells were 12 μL and 10 μL, respectively.

Sample Preparation, Chromatographic Separation, and Detection

A chromatographic system similar to that successfully used by Adler et al.[8,9] for SEC of neutral, anionic, and cationic poly(meth)acrylate copolymers was chosen. In brief, the mobile phase for chromatographic separation consisted of N,N-dimethylacetamide containing 3 g/L lithium bromide and 6 g/L of acetic acid. A refractive index value of $n = 1.437$ was used for N,N-dimethylacetamide by assuming that its value is not substantially changed by addition of the small amounts of salt and acid necessary to prepare the final mobile phase. The PMMA calibration standards used for conventional calibration were dissolved in THF (approx. 5 mg/mL) and samples (approx. 10 mg/mL) prepared in mobile phase. Volumes of 100 μL of both PMMA molecular weight calibration standards and PBA-b-PVP block copolymer samples were injected onto the two SEC columns. Chromatographic separation of PBA-b-PVP block copolymer samples was done at a column temperature of 50°C and a flow-rate of 0.5 mL/min. Measurement of the RI-, UV-, as well as the LALS- and RALS-responses, was performed at 50°C. Rinsing of the sample syringe before and after injection of every sample in order to avoid carryover effects was done with 300 μL of mobile phase.

Determination of Copolymer Composition and Molecular Weight

Copolymer composition, i.e., determination of the weight fractions W_{PBA} and W_{PVP} of PBA and PVP, respectively; in PBA-b-PVP block copolymer samples was calculated according to the following three approaches:

In approach 1 the four detectors are calibrated with polystyrene 100,000 Da. Calculation of the signal responses of the different detectors is shown in the equations. 1–4:

$$\text{Area of UV signal} = K_{UV} \cdot dA/dc \cdot c \tag{1}$$

$$\text{Area of RI signal} = K_{RI} \cdot dn/dc \cdot c \tag{2}$$

$$\text{Area of DP signal} = K_{DP} \cdot [\eta] \cdot c \tag{3}$$

$$\text{Area of LS signal} = K_{LS} \cdot M_W \cdot (dn/dc)^2 \cdot c \tag{4}$$

K_{UV}, K_{RI}, K_{DP}, and K_{LS} are the calibration constants of the UV-, RI-, DP-, (differential pressure = measure of viscosity) and light scattering detectors. dA/dc and dn/dc are the absorption and refractive index increments, respectively, c is the sample concentration and $[\eta]$ is the sample's intrinsic viscosity. M_w is the weight average molecular weight expressed in Da.

In order to determine the different calibration factors the following parameters of the calibration standard must be known: c, M_w, dn/dc in the used solvent, $[\eta]$ and dA/dc.

Presumed all calibration constants are known, the homopolymers constituting the final copolymer are measured at defined concentration and the dn/dc and the dA/dc values from the RI- and the UV-detector can be determined according to equations. 5 and 6:

$$\mathrm{dA/dc = area\ of\ UV\ signal}/(K_{UV} \cdot c) \tag{5}$$

$$\mathrm{dn/dc = area\ of\ RI\ signal}/(K_{RI} \cdot c) \tag{6}$$

Once the dn/dc and the dA/dc values for the two homopolymers are determined, the method for calculation of the copolymer composition can be established according to equations 7 and 8:

$$RI = c_A \cdot A_{RI} + c_B \cdot B_{RI} \tag{7}$$

$$UV = c_A \cdot A_{UV} + c_B \cdot B_{UV} \tag{8}$$

RI and UV are the areas of the RI and UV signal. C_A and C_B are the concentrations of homopolymer A and homopolymer B. A_{RI} and B_{RI} are the dn/dc response factors of homopolymer A and homopolymer B. A_{UV} and B_{UV} are the dA/dc response factors of homopolymer A and homopolymer B.

The method allows determination of the percentage of homopolymer A and homopolymer B at every slice across the polymer peak. As a consequence of the known composition an exact dn/dc value can now be calculated at every point of the polymer peak by assuming a linear dependence of the dn/dc value on the composition.

Furthermore, due to the known dn/dc value at every point of the molecular weight distribution curve, it is now possible to calculate the exact molecular weight by light scattering (eq. 4) at every point of the molecular weight distribution curve.

Approach 2 relies on calculation of so-called "response factors" according to Trathnigg et al.[11] using the following set of equations:

$$X_{RI} = m_i \cdot (W_{PBA} \cdot f_{RI(PBA)} + W_{PVP} \cdot f_{RI(PVP)}) \tag{9}$$

$$X_{UV} = m_i \cdot (W_{PBA} \cdot f_{UV(PBA)} + W_{PVP} \cdot f_{UV(PVP)}) \tag{10}$$

X_{RI} and X_{UV} are the measured signal responses (expressed in area counts) in the SEC chromatogram; m_i is the sample weight; and the two pairs $f_{RI(PBA)}/f_{RI(PVP)}$ and $f_{UV(PBA)}/f_{UV(PVP)}$, respectively, are the factors of the RI-responses of PBA and PVP and the UV-responses of PBA and PVP, respectively, to be determined. Presuming known data for the weight fractions W_{PBA} and W_{PVP} and further due to the fact that either equation 9 or 10 contains two unknown response factors, at least two different samples are required for calculation of the response factors. The weight fractions W_{PBA} and W_{PVP} can be easily determined by either NMR-spectroscopy or by SEC using a calibration curve constructed from a blend of both PBA and PVP of different composition and monitoring of the RI- vs. UV-signal responses. It should be stated that equations 9 and 10 are only valid for the two blocks and do not take into account the contributions of end-groups to either UV- or RI-responses, which however are considered in equation 11:

$$f_i = f_{i,\omega} + K_{mol}/M_i \tag{11}$$

Here, $f_{i\omega}$ is the response factor of a copolymer of very high mass, M_i is the mass of copolymer i and K_{mol} represents the influence of the end-groups. From equation 11, it is obvious that the term K_{mol}/M_i will be negligible at high molecular masses. Assuming negligible end-group effects, equations 9 and 10 can be rearranged yielding equation 12 from which the weight fraction of the second copolymer component results by simple subtraction yielding equation 13:

$$W_{PBA} = \frac{1}{[1 - (f_{UV(PBA)} \cdot X_{RI/XUV} - f_{RI(PBA)})/(f_{UV(PVP)} \cdot X_{RI/XUV} - f_{RI(PVP)})]} \tag{12}$$

and

$$W_{PVP} = 1 - W_{PBA} \tag{13}$$

Thus, once the response factors determined, every unknown PBA-b-PVP block copolymer composition can be easily calculated from equations 12 and 13.

Approach 3 uses a set of five PBA-b-PVP block copolymers of known PBA vs. PVP ratio obtained by NMR-spectroscopy instead of only two as used in approach 2. The two response factor pairs $f_{RI(PBA)}/f_{RI(PVP)}$ and $f_{UV(PBA)}/f_{UV(PVP)}$, respectively, were then varied until the best correspondence between the results from NMR-spectroscopy and SEC with dual RI/UV-detection was accomplished. To do this, the RI- and UV-peak areas of the five PBA-b-PVP block copolymers are measured and the four response factors determined by an iterative procedure by use of the Solver routine program of Microsoft Excel. This function allowed the resolution of a non-linear equation, where in the present case, a set of ten equations is obtained resulting from five copolymer samples measured with two detectors (RI and UV). The unknowns (i.e., the four response factors $f_{RI(PBA)}$, $f_{RI(PVP)}$ and $f_{UV(PBA)}$, $f_{UV(PVP)}$, respectively) were then calculated by the Excel program as long as the deviation from the calculated value and the exact one from NMR-measurements approximates a minimum. In this way, the following values for the four response factors have been determined:

$f_{RI(PBA)} = 4.18\,\mathrm{mV \cdot mL/mg}$
$f_{RI(PVP)} = 17.13\,\mathrm{mV \cdot /mL/mg}$
$f_{UV(PBA)} = 5.06\,\mathrm{mV \cdot mL/mg}$
$f_{UV(PVP)} = 252.15\,\mathrm{mV \cdot mL/mg}$

The response factors obtained so far are a measure of the sensitivity of the two detectors with respect to the signal intensities of the two homopolymer building blocks constituting the PBA-b-PVP block copolymer samples. Using these response factors, it will now be possible to determine the PBA vs. PVP ratio within the PBA-b-PVP block copolymer chain as a function of molecular weight.

RESULTS AND DISCUSSION

A total number of about 70 PVP-b-PBA block copolymer samples were measured with the chromatographic system described in the section Experimental. From this data set a collective of nine PBA-b-PVP block copolymer samples, for which the NMR-data were also available for comparative purpose was used for determination of copolymer composition according to approaches 1–3 as depicted in Table 1. In Figures 2a–d the RI-traces of the SEC-chromatograms of the PBA-b-PVP block copolymer sample WSRK 04044 and those of the corresponding building blocks PBA (sample WS 05023) and PVP (sample NDCA 05011) as well as the overlay of the three chromatograms are shown. In order to verify that substantial UV-absorbance is observed with the PBA-b-PVP block copolymer, the overlay of the RI- and UV-traces of the PVP NCDA 05011 homopolymer sample and the PBA-b-PVP block copolymer sample WSRK 04044 is depicted in Figures 3a,b.

As can be easily concluded from the profiles of the chromatograms in Figures 2a–d, the used procedure proves to be satisfactory in order to solve the target problem. Both symmetrical and monomodal peaks were obtained in almost all cases, which may be considered as indicative for

TABLE 1 Weight % Data of PVP of Different Samples Calculated by Means of the Different Approaches 1–3 vs. Data obtained from NMR-Spectroscopy

	Weight % PVP			
	Method of Determination			
	NMR-data	Approach 1	Approach 2	Approach 3
WS RK 04044*	16.0	13.7	13.9	15.3
WS RK 04044-2*	12.7	10.9	11.5	13.0
WS RK 04057*	12.3	11.5	10.7	12.2
WS RK 04079 RG*	32.6	23.8	32.6	32.8
WS RK 04084*	12.1	11.3	11.2	12.6
WS 05008	13.9	11.7	12.2	13.7
WS RK 04044-1	7.3	5.9	5.6	7.1
WS RK 04065	12.9	12.6	13.0	14.5
WS RK 04071	8.3	9.1	8.3	9.8

Approach 1: Multi detector copolymer calibration using OmniSEC software with dn/dc = 0.1315 for PVP homopolymer, dn/dc = 0.02852 for PBA homopolymer, dA/dc = 7.596 for PVP homopolymer, and dA/dc = 0.0604 for PBA homopolymer. Individual response factors are determined by measuring dn/dc and dA/dc of the two homopolymers PBA and PVP. Polystyrene 100'000 Da ($M_w/M_n = 1{,}02$, dn/dc = 0,185 mL/g, dA/dc = 1 mL/g, intrinsic viscosity (IV) = 0,3693 dL/g, sample concentration = 1,01 mg/mL, injection volume = 100 μL) was used as the standard for detector calibration.

Approach 2: Calibration with the two copolymer samples WS RK 04071 and WS RK 04079 RG yielding weight fractions of PVP of 8.3% and 32.6%, respectively (data obtained from NMR-measurements); response factors $f_{(RI)PBA} = 4.14$, $f_{(RI)PVP} = 17.21$, $f_{(UV)PBA} = 8.39$, $f_{(UV)PVP} = 245.80$.

Approach 3: Calculation using an iteration procedure, which includes the 5 copolymer samples WS RK 04044, WS RK 04044-2, WS RK 04057, WS RK 04079 RG, and WS RK 04084, i.e., comprising all samples marked with an asterisk in Table 1; $f_{(RI)PBA} = 4.18$, $f_{(RI)PVP} = 17.13$, $f_{(UV)PBA} = 5.06$, $f_{(UV)PVP} = 252.15$.

the presence of sufficiently homogenous copolymer samples. Furthermore, as expected, it is also well-recognizable that molecular weight of the PBA-b-PVP block copolymer indeed increased relative to the corresponding data of the building block and, thus, provides additional proof for separation according to a true SEC mechanism.

Molecular weight parameters M_n, M_w, and M_w/M_n for the copolymer samples were determined by SEC with light scattering following the copolymer analysis described in subsection 4 of "Experimental." As shown in Table 1, satisfactory correspondence of the data depicting the PVP percentage of nine PBA-co-PVP block copolymer samples was obtained between the three calculation approaches. This fact underlines the suitability of approach 1 not only with respect to determination of true molecular weight exploiting calculation of the PVP vs. PBA ratio for each slice of the SEC chromatogram but also with respect to the distribution of PVP within the PBA-b-PVP block copolymer across the molecular weight axis.

For example, the following molecular weight parameters were measured for sample WSRK 04044: $M_n = 15248$ Da, $M_w = 18170$ Da, $M_w/M_n = 1.19$ (see

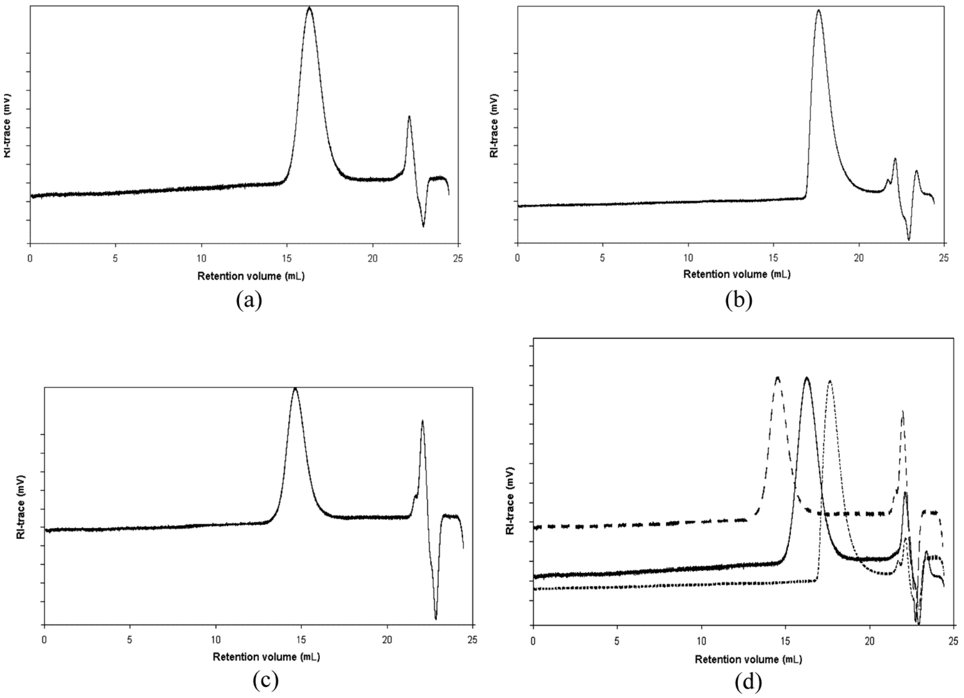

FIGURE 2 SEC-RI chromatograms of (a) PBA homopolymer WS 05023; (b) PVP homopolymer NCDA 05011; (c) PBA-b-PVP block copolymer WSRK 04044; and (d) the overlay of the three samples: WSRK 04044 (straight line), WS 05008 (dotted line), NCDA 0511 (dashed line).

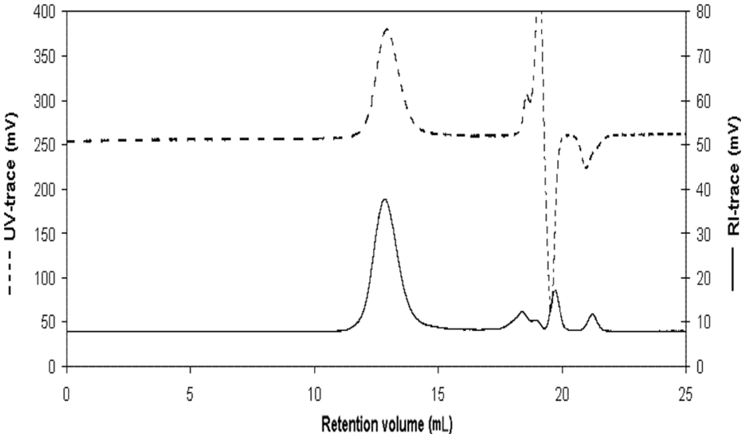

FIGURE 3 Overlay of the SEC-RI (straight line) and SEC-UV (dotted line) traces of the PVP NCDA 05011 homopolymer (3a) and PBA-b-PVP block copolymer WSRK 04044 (3b).

also columns 2–4 in Table 2) and Figure 4 depicts its PVP vs. PBA weight percent ratio as a function of molecular weight calculated by means of approach 1 (see Experimental). As demonstrated in Figure 4, the PVP content slightly increases with decreasing molecular weight in this special case. For comparative purpose, conventional calibration using narrow range PMMA calibration standards was also applied (see columns 5–7 in Table 2) and, as expected, substantial differences are observed compared with absolute calibration. These can reasonably be attributed to different dn/dc ratios across the polymer peak depending on the PBA vs. PVP content (for this, see also the tremendous differences between the dn/dc values of PBA and PVP) and, thus, will cause more or less pronounced differences in molecular weight calculation. In addition, different hydrodynamic volumes, which also may markedly depend on the PBA vs. PVP ratio, had to be considered as well.

Furthermore, polydispersity indices M_w/M_n, in the case of both conventional and multi-detector polymer calibration lying at about 1.2 and even below (except sample WS RK 0479 RG), show that the polymerization reaction provides block copolymer samples with relatively narrow polymer distribution.

In the past NMR-spectroscopy developed into a well-established and mature technique for rapid and reliable determination of the relative molar amounts of the individual blocks of a copolymer sample. Considering the molecular weights of the individual monomeric units, the results from NMR measurements can be easily exploited for calculation of both weight % and molecular weight data. However, determination of molecular weight will be either difficult or even impossible as the molecular weight of the copolymer using this technique becomes higher. Furthermore, it only yields the average PVP vs. PBA ratio, but no statement can be given with

TABLE 2 Determination of Molecular Weight Parameters M_n, M_w, and M_w/M_n by both Multi-Detector Polymer Calibration using the OmniSec Software Program "Copolymers" and Conventional Calibration using PMMA Standards

	Multi-detector Polymer Calibration			Conventional PMMA Calibration		
Sample	M_n	M_w	M_w/M_n	M_n	M_w	M_w/M_n
WS RK 04044	15 248	18 170	1.19	12 482	14 531	1.16
WS RK 04044-2	13 913	16 277	1.17	12 372	14 918	1.21
WS RK 04057	15 107	17 423	1.15	11 111	12 929	1.16
WS RK 04079 RG	6 579	9 822	1.49	4 657	5 306	1.14
WS RK 04084	14 389	16 136	1.12	13 226	15 880	1.20
WS 05008	14 399	16 285	1.13	12 754	14 408	1.13
WS RK 04044-1	11 295	13 282	1.18	11 760	13 874	1.18
WS RK 04065	13 514	16 639	1.23	11 053	13 374	1.21
WS RK 04071	15 605	16 890	1.08	10 066	11 937	1.19

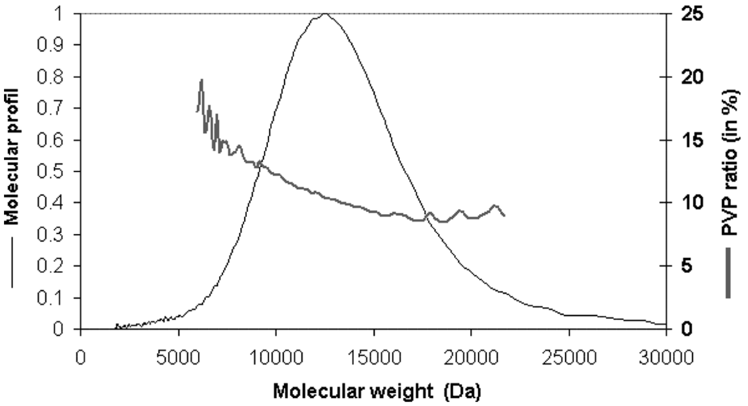

FIGURE 4 Molecular weight distribution of PBA-b-PVP block copolymer WSRK 04044 (dotted line) showing the PVP vs. PBA percentage (right y-axis) along the molecular weight axis (straight line).

respect to the molecular weight distribution of the two homopolymer blocks along the molecular weight axis. However, although being a relatively tedious and time-consuming procedure, the latter information is only accessible by SEC and dual detection exploiting the signal responses from both RI- and UV-detection. Furthermore, besides molecular weight calculation and copolymer distribution along the molecular weight axis, it is conspicuous that SEC proves to be well-suited to also detect distinct sample heterogeneities deviating from "normal" behaviors, the recognition of which may often be mandatory for efficient improvement of the synthetic route. This aspect has been considered in Figure 5 showing the overlay of the RI- and UV-traces, where a significant heterogeneity is observable

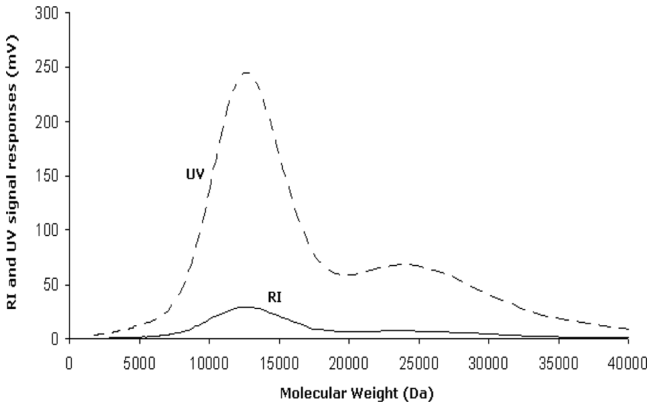

FIGURE 5 Molecular weight distribution obtained from SEC-RI (straight line) and SEC-UV (dashed line) traces of block copolymer WS RK 04079 RG showing a substantial heterogeneity in the "high molecular weight" region.

in the "high molecular weight" edge of the chromatogram. Due to the fact that UV-absorption is much more sensitive than RI measurement, the former shows a much more pronounced signal response compared with RI.

Polycations, as well as polyanions, are critical compound classes for chromatographic separation exhibiting more or less strong adsorption onto most of the commercially available stationary phases, which needs to be eliminated in order to maintain separation according to molecular size only. The problems and pitfalls in SEC of polycations and polyanions are highlighted in two excellent papers by Barth and Regnier[1] and Mori,[6] in which strategies are proposed to efficiently overcome unwanted "secondary" effects.

For SEC of polar polyanions,[2–5] sodium chloride,[2,4] and sodium hydroxide[3] aqueous mobile phases were used, and an ammonium acetate eluent system was chosen for neutral and cationic polymers.[12] In another study targeted on molecular mass characterization of cationic methacrylate-ethylacrylate copolymers, a 95% ethanolic mobile phase containing different amounts of lithium chloride was used.[7] Although the authors observed continuous reduction of the "polyelectrolyte" effect with increasing salt concentration and separation according to a SEC mechanism, unwanted adsorption occurred due to hydrophobic solute-stationary phase interactions when the salt concentration was further increased above its critical limit.

Mixed aqueous-organic mobile phases of water and dimethylsulfoxide containing sodium hydroxide and lithium bromide[10,13] and also completely organic solvents of high polarity, such as, e.g., 1,3-dimethyl-2-imidazolidinone containing lithium chloride[14] and N,N-dimethylacetamide with lithium chloride[10,14,15] were applied for SEC of lignin and polysaccharide derivatives, which often contained both cationic and anionic structural moieties in their macromolecular chains.

Some years ago two papers from the same authors were published dealing with SEC of neutral, anionic, and cationic poly(meth)acrylate copolymers with a N,N-dimethylacetamide mobile phase containing small amounts of both lithium bromide or TRIS and acetic acid on a rather novel GRAM stationary phase specially developed for SEC of polymers with intermediate polarity.[8,9] A short time later, a mobile phase comparable to that used in some research[8,9] and also using GRAM columns has been successfully exploited for characterization of technical lignins.[10]

In the present investigation, a chromatographic system similar to that described in previous studies,[8,9] successfully afforded separation according to molecular size yielding rather than symmetrical peaks for the PBA and PVP starting materials as well, as of a representative PBA-b-PVP block copolymer sample, as shown by Figures 2a–d. This view is also supported by the observation that, as expected from their differences in the molecular weights, there is an increase in retention volume in the range PBA-b-PVP $<$ PBA $<$ PVP and thus being indicative for a preponderant separation according to molecular size.

Although only "one-dimensional" separation has been applied, the result can be considered at least partly as "two-dimensional" (2D) because the used procedure provides insight into both molecular weight distribution (MWD) determined by SEC and chemical composition distribution (CCD) by exploiting the responses from either RI- or UV-detection and calculation of the PVP vs. PBA ratio. Nevertheless, "true" 2D chromatography[16–22] could yet result in more comprehensive structural information, in particular with respect to CCD or functionality type distribution (FTD) as well as the block length of the individual building blocks. However, this goal will be rather tedious and time-consuming and in the most convenient way LCCC[23–27] or AC in the first dimension should preferentially be coupled to SEC in the second one. However, it should be considered that in most cases solvent exchange may be required in order to achieve the appropriate conditions of "true" SEC in the second dimension, which is expected to be a difficult task.

CONCLUSIONS

As can be concluded from the section "Results and Discussion," sufficient proof is provided that PBA-b-PVP copolymers can be separated by means of true SEC using a GRAM stationary phase of intermediate polarity and a rather polar eluent system containing both inorganic salt and organic acid for extensive suppression of the "electrolyte" effect, which is mainly responsible for adsorptive solute-stationary phase interactions. The developed procedure gives satisfactory insight into both MWD and CCD of PVP along the molecular weight axis. Due to existing methodology as described previously, calculation of molecular weight, as well as copolymer composition of large sample numbers, can be accomplished within a relatively short time frame. Satisfactory correspondence of the weight % data of PVP calculated on the basis of calculation approaches 1–3 with those obtained from NMR measurements was observed. In addition, the analytical procedure described in this paper can also successfully be applied without modification to completely water-soluble polymer samples containing, for example, low molecular weight polyethylene glycol side chains in the acrylate moieties or even free carboxylic groups resulting from partial hydrolysis of the corresponding esters.

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