

A Comparison of the Thermal Gradient and Elution Methods of Polymer Fractionation

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INTRODUCTION

The effectiveness of the thermal gradient (or chromatographic) method for the fractionation of polymers developed by Baker and Williams¹ depends on the efficiency of the precipitation stages of the column which result from the thermal gradient produced by heating the top and cooling the bottom of the column. It is this factor which differentiates the thermal gradient method as a multistage process from the elution method, which is essentially a single-stage extraction. An indication of the degree of refractionation produced by the thermal gradient can be obtained from curves of the solubility of polystyrene in methyl ethyl ketone-ethyl alcohol mixtures given in Baker and Williams' paper. From these curves it may be estimated that 30% or more of each fraction precipitates for a 10°C. drop in temperature. Thus, with the temperature interval of 50°C. which has been used for this polymer-solvent system, at least 80% of each fraction should undergo reprecipitation.

In view of these considerations it is not surprising that the thermal gradient method is generally regarded to be superior to the elution method and that no detailed comparison of the two methods has been made. However, various factors, such as the following, suggest that it would be desirable to assess the relative effectiveness of the thermal gradient under actual fractionation conditions. Since the polymer coacervate is a viscous liquid, it may filter through the 0.1-mm. diameter beads customarily used as a column packing instead of being limited to the temperature zone dictated by solvent composition and molecular weight. The latent period which precedes polymer flocculation following a decrease in the solvent temperature would also contribute to the spreading of the precipitation zone. Krigbaum and Kurz² have shown that the solubility of the polymer is influenced by

adsorption on the sand which they used as a column packing and suggested that such adsorption might be responsible for the molecular weight reversals frequently observed in column fractionations. Since the ratio of beads to polymer is far greater in the thermal gradient method than in the elution method, any adverse effect of adsorption would be more pronounced. Finally, the elution method is simpler experimentally and more readily scaled up to handle large samples, advantages which may outweigh some loss in resolution if the fractionation results do not differ greatly from those on the thermal gradient column.

A comparison is made here of the fractionation by the thermal gradient and elution methods under nearly identical conditions. Since the results obtained from the elution column should represent the separation achieved in the initial extraction step of the thermal gradient column, this comparison affords an estimate of the effectiveness of the thermal gradient. High molecular weight polystyrene samples were chosen for fractionation since the efficient fractionation of high molecular weight polymers is difficult to achieve by any conventional single-stage process and, therefore, the contribution of the successive stages of refractionation introduced by the thermal gradient should show up most clearly. Results are also presented on the important problem of controlling the concentration of polymer in the fractions and on the effect on fractionation of trace amounts of chemical heterogeneity in the polymers.

EXPERIMENTAL

Polymer Samples

All of the fractionations described in this paper were carried out with a commercial-grade polystyrene of moderately high molecular weight (Styron) and two very high molecular weight auto-

polymerized polystyrene samples. The Styron sample, product of the Dow Chemical Company, had a viscosity-average molecular weight of 5×10^6 . The autopolymerized samples were obtained from a quantity of inhibited styrene that had polymerized during prolonged storage on the shelf. The first portion of this polymer, removed when the sample was still a loose gel, had a viscosity-average molecular weight of 3.8×10^6 . A second, larger portion was removed about 1½ years later, at which time the gel was more rigid and the viscosity-average molecular weight had increased to 5×10^6 . These two samples are designated A and B, respectively.

Solvents

Reagent-grade methyl ethyl ketone and ethyl alcohol were used in the fractionations without further purification. Although peroxides known to be present might be expected to degrade the polymer at the temperature of the top of the column (50–60°C.), the fractionation of the Styron sample under a variety of conditions which resulted in different times of contact with the methyl ethyl ketone gave reproducible results, indicating that degradation was not significant. An experiment on the Styron sample in which all the polymer from the column was collected in a single vessel and the molecular weight measured on an aliquot showed that the change in molecular weight was less than 5%. With one portion of the autopolymerized sample, however, degradation on the column was extensive, and a commonly used inhibitor, *tert*-butyl catechol, was added to the solvents. The addition of the inhibitor appeared to eliminate degradation of the polymer. However, as will be discussed later, it was found that the effect of the inhibitor on the fractionation was not limited to the prevention of degradation.

The Columns

The thermal gradient system, modified in certain respects from that used by Baker and Williams,¹ has already been described.³ The glass column used in this work was 4 cm. in diameter and had a usable section 16 in. long which was surrounded by the temperature jacket. The column was operated with the top temperature at 58–60°C. and the bottom at 10–12°C. In any single experiment the temperature variation was less than 0.5°C., and short-time fluctuations were less than 0.1°C. No experiments were conducted

to prove that such precise temperature control was necessary.

The column used for the elution runs had the same diameter as the thermal gradient column and a usable section 20 in. long. A uniform temperature, 15°C. for most runs, was maintained over this portion of the column by circulating water kept thermostatically at $\pm 0.05^\circ\text{C}$., through a water jacket. In working with the column it was found convenient to use the special mixing vessel and reservoir system designed for the thermal gradient column. In a special run at 50°C. removing air from the solvents, a feature of this special system, proved a necessity, since spaces appeared in the column packing, especially just above and below the area containing the polymer-coated beads. As the air-free solvent flowed through the column, the spaces in the packing disappeared. Both columns were packed with glass beads approximately 0.1 mm. in diameter.

Drying of the Polymer Sample on the Beads

The sample was deposited on the beads by evaporating the solvent, toluene, in a casserole over a water bath at 50°C. The mixture was stirred during drying to minimize the formation of clumps. To break up those clumps which formed, the sample was tamped until most of it passed through a No. 30 sieve. This processing was found to be especially important in the fractionation of the autopolymerized samples.

Even after being passed through a No. 30 sieve, 50% or more of the sample was in aggregates of five or more beads. Examination under a low-power microscope suggested that most of the polymer occurred in the interspace between the beads where distinct particles of polymer could be seen. It was also noted that drying from methyl ethyl ketone resulted in a more fibrous product than drying from toluene, which yielded glassy polymer.

Analysis of the Fractions

Fractions from the Styron sample were dried directly to constant weight in disposable aluminum dishes on a large glass heating plate at 60°C. in a strong draft of air. The fractions of the autopolymerized polystyrene were precipitated with a fivefold volume of methanol and collected by centrifugation to avoid possible degradation in drying directly from methyl ethyl ketone and to remove the inhibitor which was added to the solvents in certain runs.

Viscosities were measured at a single concentration, usually 0.3%, in toluene at 30°C., in a specially designed viscometer.⁴ The intrinsic viscosity was calculated from Hart's equation⁵ and the molecular weight from the equation obtained by Alfrey and co-workers⁶ for polystyrene polymerized at 60°C.

RESULTS

A. Thermal Gradient Fractionation of the Sample

In trial fractionations on polystyrenes of low to moderate molecular weight, Pepper and Rutherford⁷ found that the most important factor controlling the success of fractionation was the concentration of the fraction of maximum size eluted from the column. In the application of this principle to higher molecular weight polymers⁸ it was suggested that the maximum permissible concentration C (weight per cent) varied with the molecular weight of the fraction as $M^{1/2}$ and that the product $CM^{1/2}$ should be less than about 400 for successful fractionation. Since there are several ways of altering the concentration of the fractions, it is of interest to see whether these are equivalent and predictable in their effect. Therefore, we first present data obtained on the thermal gradient fractionation of the Styron sample with various ratios of eluting volume to sample size.

The conditions of fractionation for this set of runs are summarized in Table I, along with the molecular weight of the highest fraction and the largest value of $CM^{1/2}$. The integral distribution curves are shown in Figure 1. The points for each run are plotted against a reference curve, and the

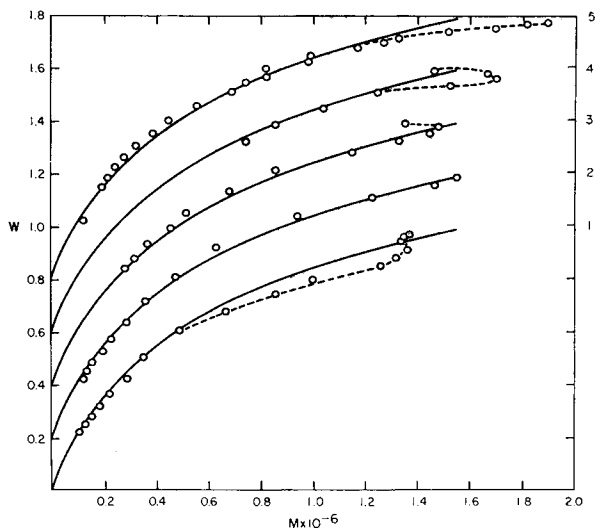


Fig. 1. Thermal gradient fractionation of Styron sample with different ratios of eluting volume to sample size. See Table I for conditions.

individual reference curves are displaced by 0.2 of unit along the ordinate. Any deviation of points from the reference curve at high molecular weights is shown by a dotted line. Curve 2 was chosen as the reference curve, since this represents a successful fractionation, the results of which have been reproduced in several other runs where the sample and mixing vessel size were the same but other conditions were changed. This method of plotting against a reference curve is also used in the other figures where a comparison of results is to be made.

The effect on the fractionation of a change in sample size is shown by curves 1 and 2. With a 0.4-g. sample (curve 2), complete fractionation is obtained, the terminal molecular weight being 1.55×10^6 ; with an 0.8-g. sample, the fractions deviate from the reference curve at about 60% of the sample. Although the fractions continue to increase in molecular weight, resolution is not complete, and the four fractions above 88% of the sample all have a molecular weight close to 1.35×10^6 . Since these two runs represent a particularly clear example of the effect of charge size on fractionation, the results for the individual fractions are given in Table II. The most striking feature of these results is that below 60% cumulative weight of the sample, corresponding fractions in the two runs have the same molecular weight and each of the fractions from the 0.8-g. run is twice that from the 0.4-g. run. Beyond this point, the correspondence in weight and molecular weight breaks down, and fractionation is obviously superior with

TABLE I

Conditions Used for Fractionation of Styron Sample^a

Curve in Fig. 1	Sample wt., g.	Mixing vessel volume, cc.	MEK concn. in reservoir, %	$M \times 10^{-6}$ for highest fraction	$CM^{1/2} \times 10^{-2}$ ^b
1	0.8	300	100	1.36	4.4
2	0.4	300	100	1.55	3.7
3	0.8	550	100	1.48	4.5
4	0.8	300	90 ^c	1.70	4.1
5	0.8	300	90 ^d	1.90	1.7

^a General conditions: 0.4-g. samples deposited on 30 g. of beads, 0.8-g. sample on 60 g. of beads; maximum flow rate 30–40 cc./hr; 13 cc. fractions.

^b Largest value for the percentage concentration of a fraction, C , and the square root of its molecular weight, M .

^c 10% of absolute ethyl alcohol.

^d 10% of 95% ethyl alcohol.

the 0.4-g. sample. While no definitive explanation can be given for the details of this behavior, it has been found, in tests which are not discussed here, that the increased concentration in fractions from the 0.8-g. run is not a result of unsaturation, at least below 60% cumulative weight of the sample. However, in the 0.8-g. run there is a shift of one tube to higher solvent concentration for fractions below 60% of the sample. If this shift is reliable, it could be responsible for the greater concentration of these fractions. On this basis, the break in the fractionation pattern which occurs at 60% of the sample is probably due to the fact that the size of the fractions is increasing more rapidly than can be accommodated by the shift in solvent concentration. Such an explanation implies that the value for $CM^{1/2}$ would depend somewhat on fractionation conditions.

TABLE II
Comparison of Fractionation Results for 0.8-g. and 0.4-g. Styron Samples^a

0.8-g. sample (Fig. 1, curve 1)			0.4-g. sample (Fig. 1, curve 2)		
Cumulative weight, %	Wt. of fraction, mg.	$M \times 10^{-5}$	Cumulative weight, %	Wt. of fraction, mg.	$M \times 10^{-5}$
21.8	20.2	1.08	21.9	9.7	1.20
24.6	23.8	1.30	25.0	12.4	1.35
28.0	28.3	1.55	28.4	14.4	1.58
32.1	33.4	1.83	32.6	16.8	1.98
36.9	40.1	2.25	37.6	20.6	2.30
42.8	48.5	2.95	43.5	24.8	2.90
50.4	62.1	3.60	51.1	30.7	3.60
60.2	80.8	4.90	60.3	38.6	4.72
68.0	63.8	6.70	72.1	49.2	6.35
74.3	51.8	8.60	84.0	48.8	9.40
80.0	47.3	10.0	91.0	28.4	12.3
84.9	39.8	12.6	95.6	20.0	14.7
88.8	31.7	13.2	98.8	11.8	15.5
91.9	25.5	13.6	99.5	5.5	
94.4	20.8	13.4	99.8	1.3	
96.0	13.3	13.5			
97.1	9.3	13.6			

^a Conditions as given in Table I.

In addition to reduction of sample size for fixed fractionation conditions, two methods are available for increasing the eluting volume as a means of decreasing the concentration of the fractions to the level required for successful fractionation. The differential mixing equation for the closed mixing vessel system is:

$$dc/dv = (c_0 - c)/V$$

Here c is the instantaneous solvent concentration in the mixing vessel, v is the volume which has left the mixing vessel, V is the mixing vessel volume (constant), and c_0 is the concentration of solvent in the reservoir (constant). According to this equation, doubling the mixing vessel size doubles the volume required to traverse each increment of solvent concentration. On the other hand, reducing the concentration of solvent in the reservoir produces a nonlinear increase in eluting volume. For example, reducing the concentration in the reservoir from 100% methyl ethyl ketone to a 90-10 mixture of methyl ethyl ketone-ethyl alcohol causes a progressive increase in the eluting volume varying from a factor of 1.1 at the start to 2.0, 2.25, and 2.7 at 80, 82 and 84% methyl ethyl ketone, respectively. By comparison with a doubling of the size of the mixing vessel, the elution volume below 80% is compressed, while above 80% methyl ethyl ketone the elution region is progressively expanded. Since polymer of molecular weight 1×10^6 is eluted at about 82% methyl ethyl ketone, the nonlinear change in the 90-10 solvent mixture should be especially effective for the fractionation of high molecular weight polymers.

Curves 3, 4, and 5 of Figure 1 show the results obtained with an 0.8-g. sample on increasing the mixing vessel size or changing the solvent concentration in the reservoir. In curve 3, where the mixing vessel size is doubled, the fractionation results conform with the reference curve, although there is inversion above 98% of the sample. In curve 4, the effect of extending the solvent gradient by reducing the solvent concentration in the reservoir to 90% by addition of absolute alcohol is shown. The results obtained are consistent with the reference curve, but it will be noted that there is enhanced resolution of the high molecular weight end of the curve, the highest molecular weight fraction being about 15% above that from the previous run. In both these runs, the fractions below 1×10^6 are of the expected size, but several fractions above 1×10^6 are almost twice as large as expected from the 0.4-g. run shown in curve 2. This is reflected in the somewhat larger values of $CM^{1/2}$ listed in Table I for these two runs. Finally, curve 5 shows the points obtained by reducing the solvent concentration in the reservoir to 90% by dilution of MEK with 95% alcohol. Here there is a marked reduction in the size of the fractions, as shown by the value of $CM^{1/2}$ in Table I, and a pronounced extension of the high molecular weight portion of the curve.

In addition to these runs on 0.8-g. samples, 2.-g. samples have been fractionated by use of a proportionately larger mixing vessel (800 cc.) and a 6-cm. diameter column. Fractionation with the methyl ethyl ketone concentration in the reservoir diluted to 90% with absolute alcohol in one case and with 95% alcohol in the other case gave results in complete correspondence with the 0.8-g. runs.

The results presented above show that the distribution curve obtained by the thermal gradient method is quite reproducible under a variety of fractionation conditions and that the fractionation responds to changes in the sample size or eluting volume approximately as expected. The values of $CM^{1/2}$ for this set of runs, shown in Table I, are consistent with the conclusion drawn earlier⁸ that successful fractionation can be obtained when the largest value of $CM^{1/2}$ is less than about 400. However, from the data in Table I, it appears that the value of $CM^{1/2}$ is relatively insensitive to minor changes in the fractionation pattern. It has also been shown that enhanced resolution of the high molecular weight portion of the sample can be obtained by the use of a mixed solvent in the reservoir, presumably as a result of the extension of the solvent gradient in the range where the high molecular weight fractions are eluted.

B. Elution Fractionation of the Styron Sample

The results from the various runs on the Styron sample by the elution method are shown in Figure 2, where the data are plotted against the same reference curve as that used in Figure 1. In the elution runs the polymer-coated beads were placed in the middle of the column. The first run at 15°C., under standard conditions (used for curve 2, Figure 1) but with the sample on 75 g. of beads, gave a distribution which coincided with the reference curve up to a molecular weight of 0.94×10^6 , as curve 1 in Figure 2 shows, but the highest molecular weight was only 1.0×10^6 . In a second run, under otherwise identical conditions, the sample was spread over 200 g. of beads to provide a thinner coating on the beads in the hope of improving the extraction step. There was, however, no improvement in the fractionation, and the curve is not shown. Two runs were then made at 50°C. to determine whether an increase in temperature would be helpful. In the first of these runs, shown as the open circles on curve 2 of Figure 2, the highest molecular weight was 1.35×10^6 , but in the second run, the filled circles on curve 2, the highest molecular weight was 1.2×10^6 . This

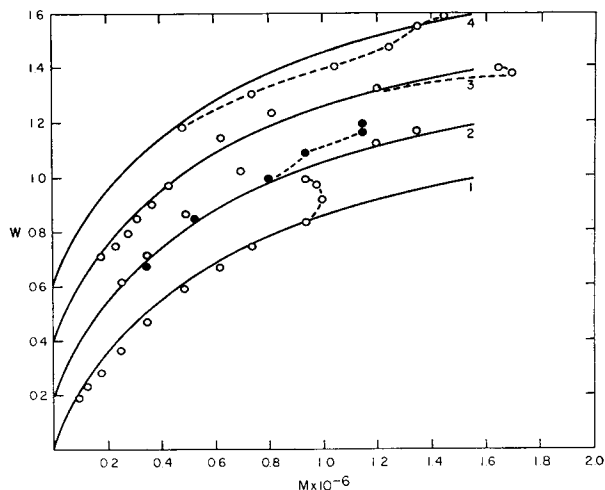


Fig. 2. Elution fractionation of Styron sample. Conditions for curves (1), (2) and (4) are: 300-ml. mixing vessel, 100% methyl ethyl ketone in reservoir, 0.4-g. sample; (1) at 15°C., (2) at 50°C., (4) acetylated sample at 15°C. Conditions for curve (3) are 300 ml. mixing vessel, 90% methyl ethyl ketone in reservoir; 0.6-g. sample run at 50°C.

indicated a slight beneficial effect of increased temperature but poor reproducibility. It is important to state that in both runs the size of certain of the fractions was larger than in the 15°C. runs and almost 50% larger than in the thermal gradient run.

To determine whether an increase in the ratio of the eluting volume to the sample size would improve the fractionation as it had done in the thermal gradient method, a run was made at 50°C. with the use of a 90-10 mixture of methyl ethyl ketone-ethyl alcohol in the reservoir. The sample size was reduced from the 0.8 g. used under these conditions with the thermal gradient method to 0.6 g. to insure that the concentration in the fractions would be sufficiently low. Under these conditions satisfactory resolution was obtained, as the results on curve 3 of Figure 2 show. The highest fraction has increased to 1.7×10^6 , equal to the value obtained with the thermal gradient method; most of the points follow the reference curve reasonably well. Again, it should be noted that, even though the sample size was reduced from 0.8 to 0.6 g., some of the fractions are as much as 70% larger than fractions at a corresponding molecular weight for the comparable thermal gradient run. The remaining curve in Figure 2, curve 4, will be considered later.

The differences noted above in the size of the fractions obtained from the elution runs compared to those from the thermal gradient runs indicate that the thermal gradient does bring about a re-

distribution of polymer in the fractions, and the fractionation results show that resolution and reproducibility are better on the thermal gradient column. However, it is noteworthy that, in all the runs with a rapidly changing solvent gradient, a considerable degree of fractionation is obtained on the elution column. By using an extended solvent gradient and a sample somewhat smaller than that

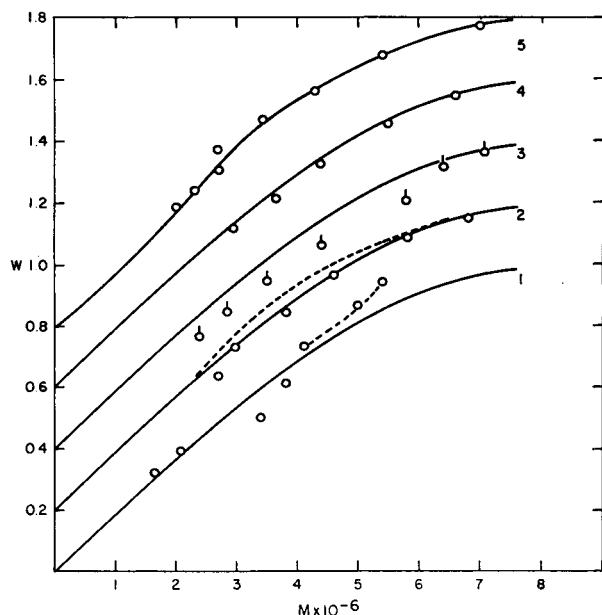


Fig. 3. Fractionation of autopolymerized sample A by thermal gradient and elution methods. Conditions for all runs are: 550-cc. mixing vessel, 89% methyl ethyl ketone in reservoir, 0.2 g. sample; curves (1), (2), (3), and (4) were run by elution at 15°C. (1) No inhibitor; (2) inhibitor added; (3) sample reacted with inhibitor, no added inhibitor for run; (4) acetylated sample, no added inhibitor. Curve (5) is for thermal gradient run with no added inhibitor.

on the thermal gradient run, fractionation is obtained which compares favorably with that from the thermal gradient method. These results suggest that the difference in performance of the two methods in this range of molecular weights is not as large as one would expect.

C. Thermal Gradient Fractionation of Autopolymerized Polystyrenes

Following a number of unsuccessful attempts to fractionate sample A, a successful run was finally obtained by a reduction of the sample size from 0.4 g. to 0.2 g. The fractionation was run with the

use of a 550-cc. mixing vessel and an 89–11 mixture of methyl ethyl ketone–ethyl alcohol in the reservoir, conditions which produced fractions of less than 0.1% concentration. The initial solvent composition in the mixing vessel was increased to 70% methyl ethyl ketone to speed the fractionation, and 20-cc. fractions were collected. The distribution curve for a typical run is shown in curve 5 of Figure 3. The points fall on a smooth curve and the highest fraction has a molecular weight of 7×10^6 , approximately twice the molecular weight of the original sample. Two other runs made under the same conditions at different times gave similar distribution curves but the reproducibility was not as good as with the Styron sample, differences being noted principally in the molecular weight of the highest fraction. Probably these differences are due to some degradation by peroxides in the methyl ethyl ketone. At any rate, these runs show that under proper conditions satisfactory fractionation can be obtained by the thermal gradient method even on such a high molecular weight polymer.

Since the remaining amount of sample A was too small for the work which was planned, a number of subsequent thermal gradient runs were made on sample B which had a molecular weight of 5×10^6 , 30% higher. With the fractionation conditions described above, a run on sample B produced fractions of progressively increasing molecular weight (curve 1, Fig. 4). However, the highest fraction was only 6.4×10^6 , not very much above the molecular weight of the original sample. This suggested that the sample was more sensitive than sample A to degradation by peroxides in the methyl ethyl ketone. When the fractionation was carried out with 0.12% *tert*-butyl catechol added to the solvents, the fractions were shifted by about 30% to higher molecular weights and a fraction of 8.3×10^6 was obtained at 94% of the sample (curve 2, Fig. 4, open circles). A second run with 0.02% *tert*-butyl catechol and with methyl ethyl ketone freed of peroxides by passage over alumina gave results in good agreement with the first run (curve 2, Fig. 4, filled circles). The last measurable fraction in this run had a molecular weight of 1.0×10^7 at 96% of the sample. These two runs show that reproducible fractionation of this more sensitive high molecular weight sample can be obtained in the presence of an inhibitor. Therefore, it was concluded that satisfactory conditions had been obtained for a comparison with the elution fractionation.

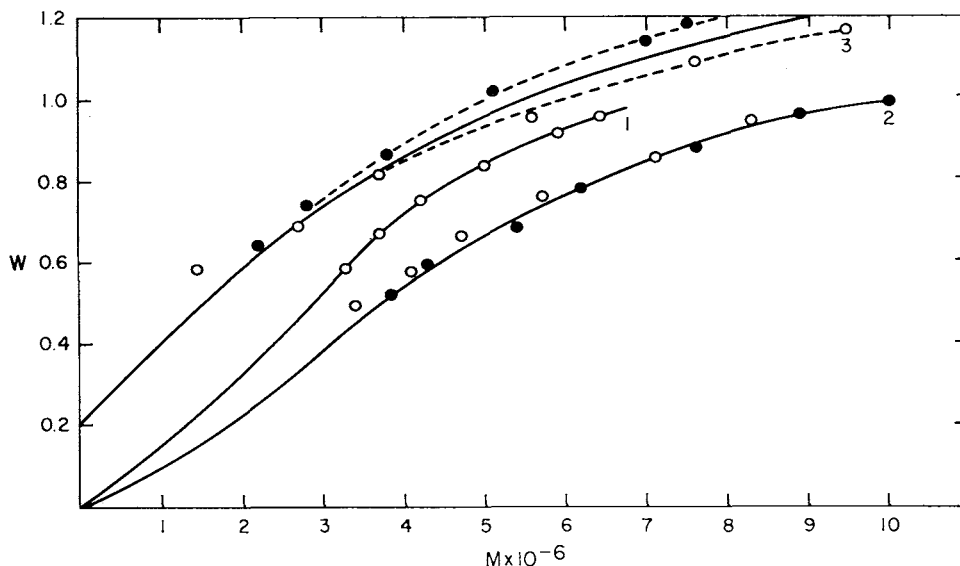


Fig. 4. Fractionation of autopolymerized sample B by thermal gradient and elution methods. Conditions for all runs are: 550 cc. mixing vessel, 89% methyl ethyl ketone in reservoir, 0.2-g. sample. Curves (1) and (2) are thermal gradient runs. (1) No added inhibitor, (2) inhibitor added. Curve (3) is for elution runs at 15°C. with inhibitor; (●) sample deposited on 75 g. of beads; (○) sample deposited on 250 g. of beads.

D. The Elution Fractionation of the Autopolymerized Polystyrene

Two runs were made on sample B at 15°C. under conditions duplicating those for the thermal gradient fractionation with 0.02% *tert*-butyl catechol and methyl ethyl ketone freed of peroxides. In the first run, the 0.2-g. sample was deposited on 250 g. of beads while in the second run the sample was deposited on 75 g. of beads to duplicate the ratio of polymer to beads used in the thermal gradient run. Although the size of some of the fractions was 50% larger than corresponding fractions from the thermal gradient runs, both of these runs on the elution column produced very good fractionation with terminal fractions of high molecular weight. The results are shown in curve 3, Figure 4, where the open circles are from the run on 250 g. of beads, the filled circles on 75 g. of beads, and the solid line is the curve from the thermal gradient run, included for comparison purposes. Thus, it appears that fractionation of this very high molecular weight sample on the elution column gives results as good as or better than those obtained by fractionation on the thermal gradient column.

These results are surprising and appear to be inconsistent with the poorer resolution obtained by the elution method on the Styron sample at

15°C., since with increasing molecular weight it becomes more difficult to obtain efficient fractionation by any conventional single-stage process. The problem arises as to whether this unexpected improvement in the quality of the elution fractionation is characteristic of the behavior of very high molecular weight polymers or whether the results are anomalous. Support for the latter possibility came from the results of special tests in which it was found that the viscosity of the autopolymerized polystyrene in toluene increased after incubation in methyl ethyl ketone containing *tert*-butyl catechol. Although the concentration of inhibitor used for these tests was about twenty times that used in the elution runs and the viscosity increase was not found to be reproducible, the results did indicate that there might be a reaction on the column of the inhibitor with the polymer.

In the experiments carried out to determine whether a possible interaction of the inhibitor with the polymer affected fractionation on the elution column, we returned to the use of the remaining portion of sample A because its greater stability permitted reliable fractionation in the absence of an inhibitor. When this sample was fractionated at 15°C. on the elution column under the same conditions used with sample B, but omitting the inhibitor, fractionation was poor (curve 1, Fig. 3). The points were somewhat

erratic, there was inversion in the terminal fractions, and the highest molecular weight fraction was 5.4×10^6 compared to 7.0×10^6 obtained on the thermal gradient column. A second run was conducted with 0.1% *tert*-butyl catechol added to the solvents. In this run the points were on a smooth curve, there was no inversion, and the molecular weight of the fraction at 95% of the sample was 6.8×10^6 . Clearly, the inhibitor had a specific effect on the fractionation process. The results of this run are shown in curve 2, Figure 3, together with a comparison with results from the thermal gradient run represented by the dotted line. Curve 2 has been used as the reference line in the other curves on this figure except for curve 5.

The question remained as to whether the effect of the inhibitor was due to a nonspecific aggregation of the polymer, a specific aggregation of the fractions or to some other action on the polymer. A portion of sample A was incubated for two days in methyl ethyl ketone containing 0.3% *tert*-butyl catechol. The polymer was precipitated with methanol, centrifuged, and washed thoroughly with methanol before drying. The intrinsic viscosity of the recovered sample checked that of the original within 1% indicating that no aggregation occurred. However, when this sample was run on the elution column, without additional inhibitor, satisfactory fractionation was obtained. Although the curve did not match the previous elution run in the presence of inhibitor, the points correspond to a smooth curve and the highest fraction had a molecular weight of 7.1×10^6 (curve 3, Fig. 3). This suggested that there were some groups in the polymer which were altered by reaction with the *tert*-butyl catechol resulting in an improvement in fractionation.

Since the possible nature of the reaction with *tert*-butyl catechol is obscure, a portion of sample A was acetylated under the conditions used by Trementozzi, Steiner, and Doty⁹ to determine if this would produce a similar improvement in fractionation, thereby indicating the presence of hydroxyl groups. It should be noted that these authors have shown that trace amounts of hydroxyl groups, below the level detectable in the infrared, are present in polystyrene prepared by emulsion polymerization. The presence of these polar groups leads to an anomalously high molecular weight in toluene which decreases to a normal value following acetylation of the polystyrene. When the acetylated portion of sample A was fractionated at 15°C. on the elution column in the absence of

inhibitor in the solvents, satisfactory fractionation was obtained (curve 4, Fig. 3). Moreover, the points lie on the reference curve which was obtained from the elution run on the original polymer in the presence of inhibitor. Thus, it appears that there are trace amounts of hydroxyl groups in this polymer which adversely affect fractionation. Blocking the groups by acetylation improves fractionation, and presumably the effect of *tert*-butyl catechol also arises from a similar reaction with the hydroxyl groups.

The results for an elution run at 15°C. on an acetylated Styron sample, in the absence of an inhibitor, are shown in curve 4 of Figure 2. Although the points do not lie on the reference curve, the highest molecular weight fraction is 1.45×10^6 . This represents a marked improvement over the elution fractionation of the original sample at 15°C., where the molecular weight of the highest fraction was 1.0×10^6 . These results show that there are also trace amounts of hydroxyl groups in the Styron sample which interfere with the elution fractionation.

Discussion

Having clarified the role of the inhibitor and the effect of chemical heterogeneity on fractionation, we can reach a conclusion which is consistent with results for both the Styron and the autopolymerized polystyrenes on the relative efficiency of fractionation on the thermal gradient and elution columns. For the original samples, in the absence of inhibitor, the thermal gradient column definitely provides superior resolution and reproducibility for fractionations of both the Styron and the autopolymerized samples. Apparently this method is less sensitive to the chemical heterogeneity which is present in both types of polystyrenes used in this study. However, under a number of specific conditions there is a marked improvement in fractionation on the elution column. In fact, elution fractionation of the autopolymerized sample following acetylation or in the presence of *tert*-butyl catechol gives results as good as those from the thermal gradient column. Even under ordinary conditions, 80% or more of the samples can be resolved, and with a decrease in the ratio of the sample size to eluting volume, complete fractionation is obtained on the Styron sample.

These results raise a number of problems. First, it appears that the difference in performance of the two column methods is less than that expected on

the basis of the elementary considerations outlined in the introduction. Possibly, this reflects the unexpected capacity to deliver fractionation which was shown by the elution method. However, it is also possible that this is due to factors which limit the efficiency of the precipitation stages of the thermal gradient method, although the present experiments do not give any direct information about this. Second, if it is correct to assume that the elution method is a single-stage extraction process, then these results indicate that the extraction step, as carried out here, is very selective. This conclusion is difficult to reconcile with the fact that the polymer sample is present in the form of particles and not as a thin, uniform coating on the beads. A related problem concerns the effect of acetylation on the fractionation obtained by the elution method. It is not possible to explain this in terms of an improvement in the extraction process, since the conversion of hydroxyl groups to ester groups would hardly affect the solubility of the polymer in the methyl ethyl ketone-ethyl alcohol solvent.

A plausible explanation of these problems could be given in terms of an adsorption on the surface of the beads, especially if the adsorption increases with molecular weight. As previously mentioned, Krigbaum and Kurz² have shown that the support used in the column fractionation of polymers cannot be considered inert. They found that the solubility of polystyrene in cyclohexane was influenced by adsorption on the coarse-grained sand used to pack the column and that adsorption increased with molecular weight. Hodben and Jellinek¹⁰ report an inverse molecular weight dependence for the adsorption of polystyrene from methyl ethyl ketone-ethyl alcohol mixtures onto charcoal. This is probably due to the porous nature of the adsorbent. In another study,¹¹ polymer adsorption on a porous surface followed an inverse molecular weight dependence but adsorption on a smooth surface was found to increase with molecular weight. Nonetheless, direct evidence on this is needed for the present system.

Adsorption of the polystyrene on the surface of the beads without reference to the molecular weight dependence provides an explanation of the behavior of the acetylated samples. The presence of hydroxyl groups in the nonpolar polystyrene molecule would result in stronger adsorption on the surface of the glass beads; this would interfere with fractionation, particularly if the polar groups are not distributed in proportion to molecular weight. The adsorption mediated by polar groups

in the Styron sample could also provide an explanation for the inversion observed in the terminal fractions of the otherwise satisfactory runs shown in Figure 1. The results of Koral, Ullman, and Eirich¹¹ on the adsorption of polyvinyl acetate on iron powder lend support to this explanation. They found that the adsorption coefficient of 13% hydrolyzed polyvinyl acetate increased by a factor of 2.7 over that for the unhydrolyzed polymer but that further hydrolysis had relatively little effect. These results imply that adsorption of this polymer is strongly affected by small numbers of hydroxyl groups just as postulated for the polystyrene samples.

If adsorption increases with molecular weight, then the fractionation on the elution column might be the result of selective adsorption on the surface of the beads superimposed on the separation achieved in the extraction step. It has been found that the dependence of adsorption on molecular weight is more pronounced in a poor than in a good solvent.¹¹ The fact that in both column methods the polymer is extracted near saturation into a poor solvent would accentuate the dependence of adsorption on molecular weight. Thus, selective adsorption might also contribute to fractionation on the thermal gradient column, playing a proportionately larger role as the size of the fractions decreases or the molecular weight increases.

References

1. Baker, C. A., and R. J. P. Williams, *J. Chem. Soc.*, **1956**, 2352.
2. Krigbaum, W. R., and J. E. Kurz, *J. Polymer Sci.*, **41**, 275 (1959).
3. Schneider, N. S., L. G. Holmes, C. F. Mijal, and J. D. Loconti, *J. Polymer Sci.*, **37**, 551 (1959).
4. Schneider, N. S., *J. Polymer Sci.*, **32**, 253 (1958).
5. Hart, V. E., *J. Polymer Sci.*, **17**, 215 (1955).
6. Alfrey, T., A. Bartovics, and H. Mark, *J. Am. Chem. Soc.*, **65**, 2319 (1943).
7. Pepper, D. C., and P. P. Rutherford, *J. Appl. Polymer Sci.*, **2**, 100 (1959).
8. Schneider, N. S., J. D. Loconti, and L. G. Holmes, *J. Appl. Polymer Sci.*, **3**, 251 (1960).
9. Trementozzi, Q. A., R. F. Steiner, and P. Doty, *J. Am. Chem. Soc.*, **74**, 2070 (1952).
10. Hodben, J. F., and H. H. G. Jellinek, *J. Polymer Sci.*, **11**, 365 (1953).
11. Koral, J., R. Ullman, and F. R. Eirich, *J. Phys. Chem.*, **62**, 541 (1958).

Synopsis

The elution method is essentially a single-stage process, whereas the thermal gradient method is a multistage process which depends upon a thermal gradient to bring about reprecipitation of polymer in the fractions. As a test of the ef-

fectiveness of the thermal gradient, comparisons have been made of fractionation by these two column methods on high molecular weight polystyrene samples. It was found that the thermal gradient method definitely provides superior resolution and reproducibility, as expected. However, the degree of fractionation obtained by the elution method was surprising, accounting for at least 80% of the sample under the usual conditions and giving complete fractionation with certain modifications of conditions. These results indicate the difference in performance of the two methods is less than expected from an elementary consideration of the operation of the columns, and fractionation by the elution method, as conducted here, exceeds that expected for a single-stage extraction process. Although the reasons for the observed behavior are not clear, the following conclusions have been reached about certain factors which influence fractionation. Alternative methods of controlling the concentration of polymer in the fractions give almost equivalent results but enhanced resolution of the high molecular weight portion of the sample is obtained with extended solvent gradients. The inhibitor, *tert*-butyl catechol, which it was necessary to add to the solvents to limit degradation of the very high molecular weight sample, plays a specific role in the fractionation due to a reaction with the polystyrene which alters the fractionation behavior without affecting the molecular weight. Also, trace amounts of chemical heterogeneity in the polymers, presumably hydroxyl groups, have a marked adverse effect on fractionation by the elution method and probably account for molecular weight reversals observed in some fractionations by the thermal gradient method. It is suggested that adsorption on the surface of the beads is responsible for the adverse effect of chemical heterogeneity on the fractionation and that possibly an adsorption which increases with molecular weight contributes to fractionation by the elution and thermal gradient methods.

Résumé

La méthode d'éluion est essentiellement un processus en une étape tandis que la méthode du gradient thermique est un processus en plusieurs étapes qui se base sur un gradient thermique pour permettre la réprécipitation du polymère en fractions successives. Comme test de l'efficacité du gradient thermique, on a fait des comparaisons de fractionnement par ces deux méthodes colonne sur des échantillons de polystyrène à haut poids moléculaire. On a trouvé que la méthode du gradient thermique donnait, sans aucun doute, un pouvoir de résolution et une reproductibilité supérieure, comme prévu. Cependant, le degré de fractionnement obtenu par la méthode d'éluion était suprenant; elle donnait au moins 80% de l'échantillon dans les conditions ordinaires et un fractionnement complet en modifiant certaines conditions opératoires. Ces résultats montrent que la différence de performance des deux méthodes est moindre que celle qu'on attendait en considérant simplement le fonctionnement des colonnes, et le fractionnement par la méthode d'éluion, comme il a été effectué ici, dépasse celui attendu pour un processus d'extraction en une seule étape. Bien que les raisons du comportement observé ne soient pas claires, les conclusions suivantes ont été avancées en ce qui concerne certains facteurs qui influencent le fractionnement. Différentes méthodes de contrôle de la concentration en polymère dans les fractions donnent presque des résultats équivalents mais on obtient une résolution supérieure de la portion de haut poids moléculaire de l'échantillon avec des gradients de solvant plus étendus. L'inhibiteur, le *tert*-butyl-catéchol,

qu'il était nécessaire d'ajouter aux solvants pour limiter la dégradation d'un échantillon de très haut poids moléculaire, joue un rôle spécifique dans le fractionnement, du fait qu'il réagit avec le polystyrène et altère le comportement du fractionnement sans affecter le poids moléculaire. Une trace d'hétérogénéité chimique dans le polymère, probablement des groupes hydroxyles, a un effet contraire prononcé sur le fractionnement par la méthode d'éluion et entre en ligne de compte pour les renversements effectués par la méthode du gradient thermique. On a suggéré que l'adsorption à la surface des grains était responsable de l'effet contraire de l'hétérogénéité chimique sur le fractionnement et qu'une adsorption qui augmente avec le poids moléculaire contribue au fractionnement par les méthodes d'éluion et du gradient thermique.

Zusammenfassung

Die Eluierungsmethode ist im wesentlichen ein Einstufenprozess, während die Temperaturgradientmethode ein Vielstufenprozess ist, welcher auf der Wiederausfällung des Polymeren in den Fraktionen durch einen Temperaturgradienten beruht. Als Wirksamkeitstest für den Temperaturgradienten wurde ein Vergleich der Fraktionierung an hochmolekularen Polystyrolproben nach den beiden Säulenmethoden durchgeführt. Es wurde gefunden, dass die Temperaturgradientenmethode, wie erwartet, in bezug auf Auflösung und Reproduzierbarkeit deutlich überlegen ist. Es war aber doch der mit der Eluierungsmethode erhaltene Fraktionierungseffekt überraschend hoch; die Fraktionierung erfasste unter den üblichen Bedingungen mindestens 80% der Probe und bei bestimmten Modifizierungen der Bedingungen wurde vollständige Fraktionierung erhalten. Die Ergebnisse zeigen, dass (1) der Unterschied in der Wirksamkeit der beiden Methoden geringer ist als nach einer elementaren Betrachtung der Wirkungswiese der Säulen zu erwarten wäre und (2) Fraktionierung nach der Eluierungsmethode, so wie sie hier durchgeführt wird, besser ist als für einen Einstufenprozess erwartet werden kann. Die Gründe für das beobachtete Verhalten sind zwar nicht klar, es waren aber doch folgende Schlüsse auf gewisse, die Fraktionierung beeinflussende Faktoren möglich. Verschiedene Methoden zur Festlegung der Polymerkonzentration in den Fraktionen liefern fast gleichwertige Ergebnisse, eine erhöhte Auflösung des hochmolekularen Anteils der Probe wird jedoch mit flachen Lösungsmittelgradienten erhalten. Der Inhibitor, *tert*-Butylbrenzkatechin, der den Lösungsmitteln zur Herabsetzung des Abbaus der sehr hochmolekularen Probe zugesetzt werden musste, spielt bei der Fraktionierung wegen einer Reaktion mit dem Polystyrol, welche das Verhalten bei der Fraktionierung beeinflusst, ohne das Molekulargewicht zu ändern, eine spezifische Rolle. Spurenweise chemische Heterogenität der Polymeren, wahrscheinlich durch Hydroxylgruppen, hat einen merklich nachteiligen Einfluss auf die Fraktionierung nach der Elutionsmethode und ist wahrscheinlich für die bei einigen Fraktionierungen nach der Temperaturgradientmethode beobachtete Molekulargewichtsumkehr verantwortlich. Es wird angenommen, dass Adsorption an der Oberfläche der Kügelchen für den nachteiligen Einfluss der chemischen Heterogenität auf die Fraktionierung verantwortlich ist und dass möglicherweise eine mit dem Molekulargewicht zunehmende Adsorption zur Fraktionierung nach der Elutions- und Temperaturgradientenmethode beiträgt.

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