

Reference samples of poly(vinylalcohol) for molar mass measurement in aqueous solution

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The preparation and characterization are described of reference samples of poly(vinylalcohol) spanning the range of molar mass 20–350 kg mol⁻¹. Evidence is presented that long-chain branching has no significant effect on the relation to gel permeation chromatography of the molar masses of the poly(vinylalcohol) samples and those of the corresponding poly(vinylacetate) samples. The measured molar masses are shown to be consistent with the elution behaviour in aqueous gel permeation chromatography with two column materials.

INTRODUCTION

With the introduction of new column materials that offer greater resolution and more reproducible behaviour than the established cross-linked dextrans, gel permeation chromatography (g.p.c.) in aqueous media is gaining in importance¹. In its present state of development, the method requires calibration of the elution volume scale with reference samples, the molar masses of which have been measured independently by light-scattering photometry, membrane osmometry or ultracentrifugation. Two series of calibrants have been used hitherto for aqueous g.p.c., but both have disadvantages. Thus, the commercially available dextran samples have relatively broad distributions of molar mass², and may have significant degrees of long-chain branching^{2,3}; and, although controlled sulphonation of polystyrene prepared by anionic polymerization can yield⁴ poly(styrene sulphonic acid) samples of narrow molar mass distribution and linear structure, their elution volumes in g.p.c. are very sensitive¹ to ionic strength. This communication describes the production and use of a series of reference samples of poly(vinylalcohol) that are free from those disadvantages; they were prepared in an extension of the work⁵ leading to the provision of the NPL reference samples of poly(vinylacetate).

Elution volume in g.p.c. reflects solvated molecular size, which depends upon molar mass and long-chain branching. Reference samples for g.p.c. should be of essentially linear molecular structure; the calibration established with such samples can be related to those for other linear polymers and, when combined with ancillary measurements, can be used to estimate branching quantitatively. Poly(vinylalcohol) is prepared most readily by the saponification of poly(vinylacetate). Long branches are formed^{6,7} in the polymerization of vinylacetate by chain transfer to polymer to an extent increasing with temperature and conversion. The NPL reference samples of poly(vinylacetate) were prepared⁵ by fractionation of polymer prepared under conditions (< 7% conversion at -24°C) that are agreed^{8,9,10} to minimize long-chain branching. Saponification of those samples produced poly(vinylalcohol) in a form insoluble in water. Solubility of poly(vinylalcohol) depends¹¹ upon such structural

features as tacticity, the proportion and distribution of vicinal hydroxyl groups and short-chain branching. Those features are known to depend upon the conditions of polymerization of vinylacetate. Preliminary experiments showed that poly(vinylalcohol) produced by the complete saponification of poly(vinylacetate) prepared to low conversion ($\leq 5\%$) at 0°C was sufficiently soluble in water to serve as reference material for molar mass measurement.

EXPERIMENTAL

Preparation

Poly(vinylacetate) whole polymers were prepared at 0°C by photochemical polymerization as described elsewhere⁵; conversions were limited to 5%. Fractions were produced by preparative g.p.c.⁵ Procedures for analytical g.p.c. and ultracentrifugation¹² of poly(vinylacetate) have been described, together with data treatment for the latter method.

Selected poly(vinylacetate) fractions were saponified by adding excess (50% molar) of aqueous sodium hydroxide (in 10M solution) to stirred solution at 40°C of the polymer in methanol (0.1M in acetate). After 1 h the mixture was filtered, and the precipitated poly(vinylalcohol) was washed exhaustively with methanol (Soxhlet) to remove salts. The polymer was dissolved in water heated slowly to 95°C and then kept at 95°C for 1 h. The solution was filtered, and the filtrate was freeze-dried to yield the poly(vinylalcohol) reference materials.

Molar mass measurement

Solutions for characterization were prepared in volumetric flasks by the method described above, with magnetic stirring, and were used at 35°C within 24 h of preparation.

Number-average molar masses were measured by membrane osmometry. A Knauer osmometer was used with Schleicher and Schüll B19 membranes. At least five separately prepared solutions in the concentration range 10⁻³–10⁻⁴ g cm⁻³ were used for each sample; duplicate measurements were made with each solution. No decrease was evident of osmotic head, π , once established (1 h) even

with samples of small molar mass. Data were reduced by relating $(\pi/c)^{1/2}$ to concentration, c . The standard deviations of fit (linear least squares) ranged from 0.9% to 5.0% and increased with molar mass, with the exception of the two samples of lowest molar mass. There were two sources of systematic error difficult to quantify: small amounts of material may have diffused undetected through the membrane, and the linear relation of $(\pi/c)^{1/2}$ and c follows¹⁴ from a relation between virial coefficients with little theoretical foundation.

The refractive index increment of two fractions was measured (number-average molar masses 90 and 140 kg mol⁻¹). A Brice-Phoenix differential refractometer was calibrated by measurements with aqueous potassium chloride. The anomalies reported¹⁵ in refractometry of poly(vinylalcohol) were not observed, and there was no significant difference between the fractions. Linear least squares analysis gave the refractive index increment 0.151 ± 0.007 cm³ g⁻¹ at 546 nm and 35°C; the \pm term combines the standard errors of fit of the calibration, the reference data for potassium chloride and the measurements (7 concentrations in the range $6-14 \times 10^{-3}$ g cm⁻³).

Mass-average molar masses were measured by light-scattering photometry. A Sofica 50 photometer was used with vertically polarized incident light of wavelength 546 nm and a narrow band-pass filter of that wavelength preceding the photomultiplier. At least five separately prepared solutions were used for each sample in the concentration range 2×10^{-4} – 5×10^{-3} g cm⁻³. Solutions were filtered successively through cellulose triacetate filters (Gelman-Hawksley) of pore size 0.2 μ m and 0.1 μ m directly into the cells. Measurements were made at eleven angles in the range 30°–150°. The optical alignment was checked using aqueous fluorescein, and the irradiance scale was calibrated by measurements with benzene. Data were reduced by fitting (least squares) a bivariate polynomial based upon the Debye equation¹⁶. The total random uncertainties were calculated by combining those of the bivariate fits, the refractive index increment and the calibration of the photometer, and lay within the range 6–7%. Systematic error may have been introduced by the calibration procedure, and by curvature of the angular dependence below the experimentally accessible angles.

Gel permeation chromatography

Spheron (Koch-Light) was packed under pressure in degassed slurries into stainless steel columns (1.2 m long and 1 cm in diameter). Three such columns were used, packed separately with Spheron of porosity grades P100, P300 and P1000, and of bead size 40–63 μ m. The columns were employed with a Waters Associates' 6000 pump, conventional loop injection (2 cm³) refractometric detection and a siphon to monitor elution volume. Aqueous sodium azide (2×10^{-4} g cm⁻³) was used as eluant at a flow rate of 1 cm³ min⁻¹. The injected concentration was 1×10^{-3} g cm⁻³.

TSK gel PW was supplied in prepacked columns (60 \times 1 cm), two of porosity grade G 5000 PW and one of porosity grade G 3000 PW; the bead size was ca. 10 μ m. The columns were used with conventional high pressure liquid chromatographic instrumentation (Waters Associates) and aqueous sodium azide as eluant at 1.2 cm³ min⁻¹. Sucrose (5×10^{-3} g cm⁻³) was added to the injected solutions of concentration 2×10^{-3} – 6×10^{-4} g cm⁻³, decreasing with increasing molar mass; the injected

volume was increased correspondingly from 0.3 to 0.6 cm³.

Procedures for g.p.c. and ultracentrifugation of poly(vinylacetate) have been described elsewhere^{5,12}, together with data treatment for the latter method^{12,13}

RESULTS AND DISCUSSION

It was necessary to show that poly(vinylacetate) prepared to low conversion at 0°C was free of long-chain branching significant hydrodynamically, so that a calibration in g.p.c. established with poly(vinylalcohol) prepared from that poly(vinylacetate) would be indistinguishable from one established with linear polymer. Branching can be quantified most reliably in polymer fractions. Recent work^{12,13} has shown that branching in poly(vinylacetate) can be estimated by parallel experiments in g.p.c. and sedimentation velocity, without appeal to measurements of molar mass. The method¹³ leads to estimates of h , the ratio of the hydrodynamic radius of the branched molecule to that of its linear isomer, both in a theta-solvent for linear molecules. Application to the eight linear fractions⁵ upon which the method is based gave h in the range 0.99–1.01. Four fractions of poly(vinylacetate) prepared to low conversion at 0°C gave¹² h between 0.96 and 0.98. The estimated uncertainty in the determination of h is some $\pm 8\%$ so that the method reveals no significant branching in the fractions of polymer prepared at 0°C. By contrast, fractions of commercial branched poly(vinylacetate) yielded $0.80 < h < 0.91$ ¹².

It is of interest to calculate the sensitivity of molar mass determination by g.p.c. to long-chain branching equivalent to the extreme values of h (0.96 and 0.98) measured for fractions of polymer prepared at 0°C. Elution behaviour in g.p.c. is related to hydrodynamic volume. Provided that the possible difference in expansions of branched and linear molecules can be neglected¹², the ratio of the hydrodynamic volumes of a branched molecule and its linear isomer in the good solvent used for g.p.c. can be identified with h^3 . The hydrodynamic volumes of the fractions of poly(vinylacetate) prepared at 0°C are thus 0.85–0.94 of those of the linear isomers; note, however, that the uncertainty of these estimates is large ($\pm 27\%$).

The consequences for g.p.c. of those differences in volume follow readily for the case with a linear calibration

$$\ln M_r = a - bv.$$

Here M_r represents relative molecular mass, v is elution volume and a and b are empirical constants. Hydrodynamic volume, V_h , is proportional to the product $[\eta]M_r$, where $[\eta]$ is the limiting viscosity number, which is related to M_r by the Mark-Houwink equation for linear molecules,

$$[\eta] = KM_r^\alpha$$

where K and α are empirical constants. It follows that V_h is proportional to $KM_r^{1+\alpha}$, so that

$$\frac{d \ln V_h}{dv} = -(1 + \alpha)b$$

and hence

$$\frac{d \ln M_r}{d \ln V_h} = \frac{1}{1 + \alpha}$$

Table 1 Molar masses of poly(vinylalcohol) reference samples measured directly and calculated from g.p.c. of the corresponding poly(vinylacetate) samples

Sample descriptor	Number-average molar mass/kg mol ⁻¹		Mass-average molar mass/kg mol ⁻¹	
	measured	calculated	measured	calculated
PVA/25	23.4 ± 2.3	19.0 ± 1.9	26.1 ± 1.3	27.1 ± 2.4
PVA/40	38.7 ± 3.1	38.2 ± 3.8	44.6 ± 2.2	51.2 ± 4.6
PVA/70	71.1 ± 1.2	68.0 ± 5.4	73.7 ± 3.8	85.4 ± 6.0
PVA/80	65.5 ± 0.9	61.9 ± 5.0	96.1 ± 4.8	91.6 ± 6.4
PVA/115	100 ± 3	95.1 ± 5.7	129 ± 7	140 ± 7
PVA/165	146 ± 3	140 ± 8	186 ± 9	196 ± 10
PVA/215	190 ± 4	201 ± 16	238 ± 12	268 ± 9
PVA/270	230 ± 13	256 ± 26	322 ± 17	353 ± 32

For linear poly(vinylacetate) in tetrahydrofuran at 35°C, α has been shown⁵ to be 0.724 ± 0.011 , so that molar masses of the poly(vinylacetate) samples with degrees of branching corresponding to $h = 0.98$ and 0.96 are underestimated by the calibration in g.p.c. for linear molecules by 3% and 7%, respectively. Such differences are barely significant in the relation of directly measured molar masses to estimates from g.p.c. It follows that the calibration established in g.p.c. with the NPL reference samples of poly(vinylacetate) can be used to estimate the molar masses of the fractions of poly(vinylacetate) prepared to low conversion at 0°C. The estimated molar masses in turn can be used to calculate those of the poly(vinylalcohol) samples formed by saponification. Comparison of those calculated values with molar masses measured directly, by light-scattering photometry and membrane osmometry, provides a test of consistency and further evidence for low levels of branching in the poly(vinylacetate).

Long-chain branches are formed in the polymerization of vinylacetate by chain transfer to polymer at two possible sites. Branches attached to a carbon atom in the chain will remain in the corresponding poly(vinylalcohol); branches attached to methyl carbon atoms of the pendant acetate groups will be removed by saponification, so that the degree of polymerization of a poly(vinylalcohol) sample will then be smaller than that of the poly(vinylacetate) from which it is formed. The proportions of branches of the two sorts are controversial; the published estimates^{17,18} vary widely with the analytical method. However, all estimates agree that hydrolysable branching is a significant fraction of the total.

The Table compares the molar masses of the poly(vinylalcohol) samples measured directly with the estimates from g.p.c. of the corresponding poly(vinylacetate) samples and the stoichiometry of saponification. The uncertainty limits given for the directly measured values are the root mean squares of the standard deviations of fit of the respective least squares extrapolations. Those of the values derived from g.p.c. combine similarly the standard error of prediction of the primary calibration and the replication error of the chromatograms. The difference between the directly measured and inferred molar mass in no case exceeds by more than 5% the sum of the respective standard errors. The molar masses calculated from g.p.c. and the stoichiometry of saponification are traceable to membrane osmometry and light-scattering photometry of the NPL reference samples of poly(vinylacetate). The Table shows clearly the coherence of molar mass measurements in water and in

organic solvents, and confirms the absence of significant branching removable by hydrolysis in poly(vinylacetate) prepared to low conversion at 0°C.

There is published evidence⁹ that such poly(vinylacetate) is without unhydrolysable branching significant hydrodynamically; fractionation following saponification and reacylation yielded material that conformed to the Mark-Houwink relation established¹⁰ for poly(vinylacetate) prepared to low conversion at -19°C. Taken together with the evidence (Table 1) of no significant hydrolysable branching, that result is consistent with the above demonstration of linearity of molecular structure by sedimentation velocity and g.p.c.

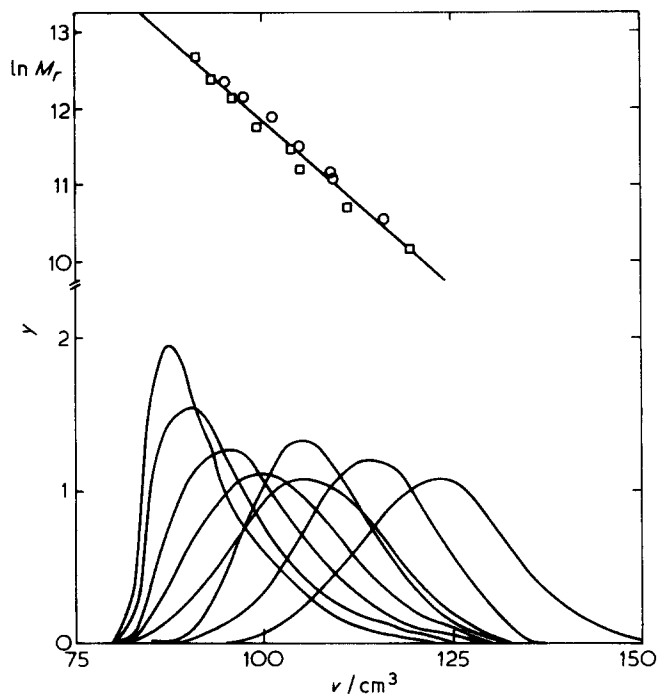


Figure 1 Normalized g.p.c. traces (chromatogram height, y , versus elution volume, v) and derived calibration (relative molecular mass, M_r , versus v) for poly(vinylalcohol) samples and Spheron gel: \circ number-average and \square mass-average relative molecular mass

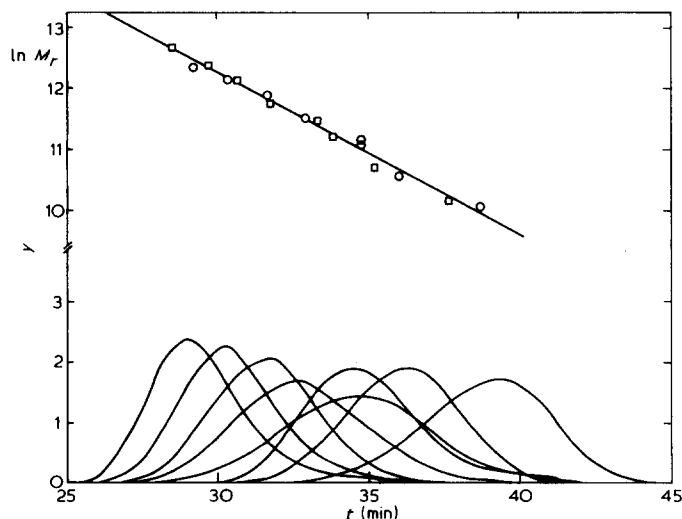


Figure 2 Normalized g.p.c. traces (chromatogram height, y , versus elution time, t) and derived calibration (relative molecular mass, M_r , versus t) for poly(vinylalcohol) and TSK-gel: \circ number-average and \square mass-average relative molecular mass

The utility of the poly(vinylalcohol) samples as calibrants in g.p.c. was tested with column materials of poly(2-hydroxyethylmethacrylate) available commercially (Spheron; Koch-Light) and a proprietary gel of undisclosed structure (Toya Soda Company TSK-gel PW). Figures 1 and 2 show the normalized elution curves and the calibration relations derived from them by an iterative procedure described elsewhere¹⁹. The resolution of the TSK-gel columns was greater than that of those packed with Spheron, as would be expected from the difference in particle size. The elution behaviour of the poly(vinylalcohol) reference samples was consistent with the measured molar masses in both cases. There was no evidence of tailing ascribable to adsorption. The utility of the new reference samples is therefore established.

Comparison of the directly measured molar masses and the g.p.c. traces reveals a likely systematic error in the number-average molar mass of PVA/25 as measured by membrane osmometry. The measured polydispersity (1.12) is clearly inconsistent with the breadth of the g.p.c. traces (Figures 1 and 2). Diffusion of polymer through the membrane is most likely with that sample, and would lead to an overestimate of number-average molar mass.

REFERENCES

- 1 Cooper, A. R. and van Derveer, D. S. *J. Liq. Chromatogr.* 1978, **1**, 693
- 2 Soeteman, A. A., Roels, J. P. M., van Dijk, J. A. P. P. and Smit, J. A. M. *J. Polym. Sci. Polym. Phys.* 1978, **16**, 2147
- 3 Granath, K. A. *J. Colloid Sci.* 1958, **13**, 308
- 4 Carroll, W. R. and Eisenberg, H. *J. Polym. Sci. (A-2)* 1966, **4**, 599
- 5 Atkinson, C. M. L. and Dietz, R. *Europ. Polym. J.* 1979, **15**, 21
- 6 Lindemann, M. K. in 'Vinyl Polymerization' (Ed. G. E. Ham), Dekker, 1967, p. 207
- 7 Graessley, W. W., Hartung, R. D. and Uy, W. C. *J. Polym. Sci. (A-2)* 1969, **7**, 1919
- 8 Matsumoto, M. and Ohyanagi, Y. *J. Polym. Sci.* 1960, **46**, 520
- 9 Morishima, Y., Kim, W. and Nozakura, S. *Polym. J.* 1976, **8**, 196
- 10 Hobbs, L. M. and Long, V. C. *Polymer* 1963, **4**, 479
- 11 'Polyvinyl Alcohol' (Ed. C. A. Finch) Wiley 1973 Ch. 2 and 10
- 12 Dietz, R. and Francis, M. A. *Polymer* 1979, **20**, 450
- 13 Dietz, R. *J. Appl. Polym. Sci.* in press (1980)
- 14 Coll, H. and Stross, F. H. 'Characterization of Macromolecular Structure' (Ed. D. McIntyre) National Academy of Sciences, Washington, 1968, p 10
- 15 Matsumoto, M. and Ohyanagi, Y. *J. Polym. Sci.* 1958, **31**, 225
- 16 Miller, W. and Stepto, R. F. T. *Europ. Polym. J.* 1971, **7**, 65
- 17 Small, P. A. *Adv. Polym. Sci.* 1975, **18**, 1
- 18 Nozakura, S., Morishima, Y. and Murahashi, S. *J. Polym. Sci. (A-1)* 1972, **10**, 2781
- 19 Atkinson, C. M. L. and Dietz, R. *Polymer* 1977, **18**, 408