Calibration of Vapor Pressure Osmometers for Molecular Weight Measurement

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Synopsis

Experimental molecular weights in the range 200 to 100,000 Daltons have been determined by vapor pressure osmometry using three different solvents. For all values in excess of 10,000, molecular weights were also determined by membrane osmometry. The general agreement found for molecular weights determined under a variety of conditions, leads to the conclusion that values as high as 100,000 Daltons can be determined by at least one type of vapor pressure osmometer when it is calibrated with a material of 200 Daltons. Thermistor self-heating and diffusion in the liquid phase are shown to be unimportant for this particular instrument.

INTRODUCTION

Since the introduction of a commercial instrument almost 20 years ago, vapor pressure osmometry has become one of the most common methods for molecular weight measurement in the 100 to 10,000 Dalton range. The ease of measurement, sensitivity, and ability to use a wide range of temperatures and solvents has made it generally preferable to ebulliometry and cryoscopy for most measurements.

However, it is not an equilibrium method, so the instrument must be calibrated with a material of known molecular weight. Many pure compounds have been used for this purpose, most of them having molecular weights between 100 and 500 Daltons. Within the last five years several authors have questioned this procedure because they report a molecular weight dependence of the calibration factor.[1-4] Other workers have not noted such an effect, and indeed Wachter and Simon[5] have reported molecular weight measured as high as 400,000 after calibration with dibenzyl disulfide and sulfonal.

Resolution of this question is important since a molecular weight dependent calibration necessitates availability of standard materials with a wide range of molecular weights and soluble in all useful solvents. The problem is complicated further by the availability of an instrument, based on the design of Dohner, Wachter, and Simon,[6] which can be used for molecular weight measurements at least as high as 100,000 Daltons. If standard materials for the entire molecular weight range are necessary, the applicability of this instrument will be limited.

All authors of refs. 1 through 4 have found that molecular weights determined for high-molecular-weight materials were lower than those expected from information available from the supplier. As a general rule the negative deviation increased with increasing molecular weight. In this regard it is troubling that the results of Wachter and Simon were also lower than those found by membrane osmometry. This difference was attributed to a small amount of low-molecu-

lar-weight material which passed through the membrane thus yielding an erroneous molecular weight by membrane osmometry. Although plausible, that is not the only possible explanation, so the comparison of results by both methods needs to be investigated more closely.

Morris also found a very significant difference in molecular weights of the same material measured in different solvents. Such an effect means that no universal correction can be found for changes of calibration factor with molecular weight.

Bersted found that the change of thermistor resistance with solute concentration was a linear function of the square of the bridge voltage. This effect was attributed to self-heating of the thermistor bead since that heating would also change as the square of the voltage. Thus he concluded that self-heating of the thermistors was an important feature which must be included in any satisfactory description of the mechanism of this measurement. This was in contrast to earlier proposals.

Using a commercially available instrument based on the design of Dohner, Wachter, and Simon we have investigated these three specific points. Calibration factors with materials ranging in molecular weights from 128 to 678 have been determined. The resulting values have been used to determine molecular weights of polystyrene standards up to 110,000 Daltons in three different solvents and the results compared to values found by membrane osmometry. Finally we have investigated the effect of bridge current on the calibration factor.

Drop size effects are not a factor in the cell geometry used since the volume of liquid on the thermistor is determined by the volume held by the fine mesh screen and cannot vary.

EXPERIMENTAL

Solvents: All solvents were "Distilled in Glass" grade from Burdick and Jackson and were used as received.

Solutes: Calibrating materials used as received were biphenyl: Eastman White Label; benzil: Matheson, Coleman, and Bell; and sucrose octaacetate: Matheson, Coleman, and Bell 99+%.

In addition sucrose octaacetate from another lot, not labeled 99+%, was recrystallized and the calibration factor for this material determined to be within 1% of that for the 99+% material.

Polystyrenes from Pressure Chemical Co. with nominal molecular weights of 17,000, 50,000, and 110,000 were used as received.

Apparatus: Membrane osmometry measurements were made with a Wescan Model 231 osmometer using Schleicher and Schuell type RC51 membranes. The vapor pressure osmometer was a Corona/Wescan Model 232A molecular weight apparatus.

Data Treatment: As pointed out by Wachter and Simon, a linear extrapolation of the values of $\Delta V/C$ versus C will not be valid at high molecular weights due to curvature caused by the third virial coefficient. To properly account for this curvature they made measurements at ten or more concentrations and fit the data to a quadratic rather than a linear equation. They also applied the theory of Maron together with the known intrinsic viscosity–molecular weight relationship to get a linear graphical method.

TABLE I

Nominal Molecular Weight	Membrane Osmometry	Vapor Pressure Osmometry		
		Solvent	Linear Plot	Square Root Plot
17,500	16,700	Toluene	17,400	17,800
		Cyclohexane	15,500	15,500
		Dichloroethane	16,900	17,200
50,000	45,300	Toluene	46,300	46,400
		Cyclohexane	47,200	47,200
		Dichloroethane	44,900	43,900
110,000	108,000	Toluene	145,000	100,000
		Cyclohexane	104,000	103,000

However, for practical purposes it is desirable to have a simple graphical procedure which can be applied to four data points to get a reliable molecular weight on samples for which little other data is available. Such a procedure has been suggested by Stockmayer and Cassasa[7] and applied successfully by others. That procedure is to plot $(\Delta V/C)^{1/2}$ versus concentration, obtain the best fit to a straight line and get the intercept $(\Delta V/C)^{1/2}$ at $c = 0$. The intercept value is then squared and that value used for calculation. Molecular weights found by both procedures are given in Table I. Within experimental error the results from both data treatment methods are the same except for the highest molecular weight in toluene. In that case the combination of high molecular weight and high concentrations leads to a significant contribution by the third virial coefficient. Appropriate correction for this is supplied by the square-root plot but not by the linear plot.

For the molecular weight range covered, a linear plot was satisfactory for the membrane osmometry data.

RESULTS AND DISCUSSION

The effect of bridge current was examined in toluene at several concentrations of sucrose octaacetate. In the Model 232A, bridge current can be adjusted and read directly from a panel meter. In the operating mode, the meter reading is proportional to the bridge unbalance voltage. For constant current this voltage will vary directly with resistance of the measurement thermistor, so $\Delta V/i$ is directly proportional to ΔR found in other instruments such as the Mechrolab 301.

We have measured ΔV, the unbalanced voltage, at several currents and several concentrations. The results are given in Table II where the values are $\Delta V/i$. At each concentration it can be seen that this quantity is independent of bridge current, in contrast to the results of Bersted where ΔR was found to be a strong function of bridge voltage. For the instrument used here we find no evidence for Bersted's contention that thermistor self heating is an important parameter in instrument sensitivity.

Molecular weights of the three polystyrene samples were determined by membrane osmometry in toluene at 40°C. The results were in good agreement with those reported by the manufacturer as shown in Table I. For each polymer,

the osmotic pressure π of four concentrations was determined, π/C plotted versus concentration and a best-fit linear extrapolation made to zero concentration. This intercept was used for molecular weight calculation.

Calibrations of the Model 232A molecular weight apparatus for subsequent molecular weight measurements were done with both benzil and sucrose octa-acetate in toluene. Similar calibrations have been done in several instruments over a period of more than a year and found to agree within one percent when several concentrations are used and the resulting $\Delta V/C$ values extrapolated to zero concentration. It is essential that benzil solutions be prepared just before use because they are not stable. As a result of this work, and many other cali-brations in several solvents, we have found sucrose octaacetate to be a superior standard material for the purpose. It is soluble in most organic solvents, its solutions are stable, and its vapor pressure is low enough for it to be used up to 140°C. We have therefore adopted it as our most commonly used standard and it alone was used for calibrations in dichloroethane.

Because sucrose octaacetate is not soluble in cyclohexane, biphenyl, and benzil were used as standards for that solvent. Calibrations with these materials over a time period of two weeks agreed within 2.5%.

Molecular weights of the three polystyrenes were determined by vapor pressure osmometry using the calibration constants obtained as just described. Each of the three samples was run in toluene at 50°C, and in cyclohexane at 35°C. Those with nominal molecular weights of 17,000 and 50,000 were also measured in 1,2-dichloroethane at 35°C.

In each case at least four solute concentrations were used. Determined values of $\Delta V/C$ were plotted against C and a linear extrapolation to zero concentration used to determine the intercept. An alternative method plotted $(\Delta V/C)^{1/2}$ vs C and a linear extrapolation used as suggested by Stockmayer and Cassasa.[7] The results are summarized in Table I.

In all cases but one, the molecular weights found by linear extrapolation agree within experimental error with those found by the method of Stockmayer and Cassasa. The sample with nominal molecular weight 110,000 in toluene gave a molecular weight about 30% too high when best fit linear extrapolation was used. However the square root plot gave the expected molecular weight. This is presumably due to an appreciable third virial coefficient contribution at the highest concentrations used.

On the basis of this result, and others not pertinent to this work, we feel that

TABLE II
Variation of Instrument Sensitivity $\Delta V/i$ With Bridge Current

Sample	Bridge Current (μA)		
	30	38	60
Polyol, MW = 2.7×10^3, in $CHCl_3$ at 38°C, 12.4 g/l	5.9	5.7	5.8
Sucrose octaacetate in toluene at 50°C, 9.80 g/l		12.1	12.1
Sucrose octaacetate in toluene at 50°C, 3.95 g/l	5.0	5.0	5.0
Sucrose octaacetate in toluene at 50°C, 0.66 g/l		0.83	0.86
Sucrose octaacetate in toluene at 50°C, 0.11 g/l		0.14	0.13
Polystyrene, MW = 45,000, in toluene at 50°C, 1.60 g/l		0.52	0.52

the method of Stockmayer and Cassasa provides a simple graphical method to permit molecular weight determinations as high as 100,000 by vapor pressure osmometry even in thermodynamically good solvents.

The results presented here permit two conclusions:

(a) An instrument designed according to Dohner, Wachter, and Simon, when calibrated with low-molecular-weight standards, can be used to measure molecular weights as high as 100,000.

(b) A difference in performance exists between this instrument and the Mechrolab 300 series instruments which were used in the work of refs. 1 through 4.

Thus, the problem posed by earlier work appears to be partially resolved by the fact that different results are associated with different instrument designs. We cannot at this time provide a quantitative explanation for the deviation in results between the two designs.

The theory of Bersted[4] yields three separate cases under different experimental conditions. Our results were not obtained under conditions suited to case I. The predictions of his case II are more in agreement with his experimental observations and those of refs. 1 and 4 so that case does not explain our results. Our data can only be described by his case III where condensation is assumed to predominate over that of self-heating. For the results given in Table II the power generated by ohmic heating in each thermistor varies from 20 to 40 μW. The molar concentrations vary from about 1×10^{-4} to 1×10^{-2}. This same concentration range applies to the molecular weight measurements summarized in Table I.

Bersteds experimental results[3] were obtained in this same concentration range. We believe the resistance of the thermistors in his instrument was about 3000 ohms from which one calculates a power generation of 5 to 20 μW. Therefore, the concentration and ohmic power generation conditions in his experiments and ours were very similar. It would appear that the difference in results must reside in the details of sensor construction.

In the instrument used for this work each thermistor is totally enclosed in glass and supported on a glass tube. These thermistor probes protrude upward through the bottom of the chamber with the active element located in the upper tip of the glass probe. A small piece of very fine platinum or stainless steel screen is shaped to fit tightly over the upper end of the probe so that it will hold a fixed volume of liquid in intimate contact with the thermistor element.

In this configuration the liquid volume on the thermistor never varies since any excess simply drains away into the bottom of the chamber. Because the volume of liquid is determined by the void volume of screen, the surface to volume ratio for the liquid is much larger than is the case with a suspended drop as used in earlier instruments. Furthermore, the presence of the metallic screen promotes thermal equilibration throughout the liquid with very little dependence on convection or conduction in the liquid.

The larger liquid surface area in this geometry, relative to the pendant drop, will cause solvent condensation heating to predominate, over that of Ohmic heating of the thermistors, down to lower solution concentrations. In that case it could fit Bersted's case III and also the theory of Tomlinson et al.[8]

It is clear that certain geometric features of the pendant drop geometry are

either nonexistent or greatly reduced in the design of Wachter and Simon, but one feature is the same for both. Condensation of solvent into the solution disturbs the concentration equilibrium in the liquid. Indeed it appears that this effect should be more severe in the platinum screen geometry than in the pendant drop. In the free drop, thermal inhomogeneities will cause convective stirring and thus minimize the importance of the diffusion process. The wire screen promotes thermal equilibration and at the same time obstructs convective mixing. Therefore, diffusion in the liquid would be more important in the screen than in the free drop. Such diffusion would certainly be molecular weight dependent, so the absence of any effect on the calibration constant clearly indicates that liquid diffusion is not a rate controlling mechanism in this process.

This argument is not new,[8] but experimental observations given here provide the most definitive evidence yet reported for its validity.

One further point deserves mention. Kamide et al.[9] have developed a theory to show that unsaturation of the solvent atmosphere with respect to the solvent on the thermistor, will not have an appreciable effect on the calibration constant. Bersted on the other hand found a rather large effect when octacosane was added to the solvent reservoir. Presumably this effect was caused by vapor pressure lowering. However, Kamide assumed that unsaturation was due to the slightly higher temperature of the thermistors caused by self-heating. This temperature difference would be no more than 0.02°C in the design of Wachter and Simon, although it could be somewhat higher in the instrument used by Bersted.

The lowest concentration of octacosane used by Bersted would give a vapor pressure lowering equivalent to a temperature decrease of 0.2°C, a factor of 10 higher than the self-heating effect. At this lowest concentration, the effect observed by Bersted was small and it only became large at even higher concentrations. Thus there is not necessarily a conflict between the theoretical prediction of Kamide (if applied to realistic instrumental conditions) and the observations of Bersted which resulted from rather extreme conditions.

CONCLUSIONS

On the basis of these results we conclude that a vapor pressure osmometer based on the design of Dohner, Wachter, and Simon can be used for measurement of molecular weights at least as high as 100,000 Daltons when calibrated with solutes having molecular weight as low as 200 Daltons. This is in agreement with the results of Wachter and Simon but our data show closer agreement between membrane osmometry and vapor pressure osmometry thus permitting the claim to accurate results without the slightest equivocation.

We have also found sucrose octaacetate to be a superior calibration material for most organic solvents used for vapor pressure osmometry.

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