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Methods of Fractionating Polymers by Ultrafiltration

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Synopsis

Polyvinylpyrrolidone and dextran polysaccharides have been fractionated by ultrafiltration by using newly developed membranes with sharp molecular weight cutoffs. Three different experimental techniques were employed: (a) batch ultrafiltration by using progressively more retentive membranes; (b) continuous ultrafiltration by using several similar membranes in series; (c) variable pressure ultrafiltration by using a single membrane and varying the applied pressure. Ultrafiltration is shown to be a technique of preparative fraction having the advantages of simplicity, speed, and economy.

INTRODUCTION

Both of the polymer fractionation techniques currently in wide use, fractional precipitation[1] and gel-permeation chromatography,[2] require skilled experimentation while usually producing only a very small quantity (milligram to gram) of fractionated material. The recent development of sharp molecular weight cutoff membranes offers a simple method of obtaining good fractionations of large quantities of polymeric materials.

Traditionally, dialysis has been the most widely used membrane fractionation technique.[3] In dialysis, the molecules diffuse through the membrane down a concentration gradient. The degree of separation is therefore a function of the relative diffusion coefficients and the ratio of molecular diameter to pore diameter. Like most diffusion processes, dialysis is very slow, and this is its major drawback. In order to speed the process, dialysis membranes are made as thin as possible, and the concentration on the downstream side of the membrane is kept as low as possible. This leads to fragile membranes and large volumes of dialysate.

Membranes may also be used as ultrafilters. In ultrafiltration, solvent flows through the membrane under the action of a hydraulic pressure gradient. Solutes which are not retained by the membranes are carried through the membrane by convection with the solvent. Ultrafiltration membranes with high solvent permeability would permit much faster separation between a retained and a non-retained solute than can be achieved via dialysis. In addition (in contrast to dialysis[4,5]) the solute which passes through the membrane is not excessively diluted. Unfortunately, until quite recently the available ultrafiltration membranes suffered from two major drawbacks. First, the membranes rapidly plugged with molecules caught in pore constrictions which were not then able to diffuse back into the solution against the bulk solvent flow. This resulted in a rapid drop in flux.[3,6] In effect,

these membranes acted as depth filters. With dialysis, plugging is not such a severe problem, since a molecule which diffuses into a pore is in the same way able to diffuse out. Second, the ultrafiltration membranes used hitherto discriminated poorly between species with similar molecular weights. For these reasons, dialysis has been the preferred membrane separation technique.

A range of ultrafiltration membranes has now been developed which overcome the problems found with earlier types. These new membranes, unlike previous isotropic structures,[7] consist of a dense layer, 1–2 μ thick, supported upon a 50–100 μ thick porous substructure. The "skin" side, which faces the solution to be filtered, contains a network of microporous capillaries which are capable of retaining molecules with radii from 5 to 40 Å. The supporting substructure has pores with diameters of 1–10 μ. The single function of the substructure is to support the skin layer and it has no effect upon the rejection of solute or the flux rate of solvent. With these anisotropic membranes polymer molecules are held back at the membrane surface and subsequently back-diffuse into the bulk solution. Since the pore length in these membranes is much smaller than in an isotropic membrane, the probability of plugging at pore constrictions is much reduced. It is also probable that there is a gradual increase in pore radius from the outer or skin side of the membrane toward the other substrate side. This results in the majority of pore constrictions being localized right at the membrane surface reducing the possibility of plugging. A further advantage of the thin membrane skin is the very high fluxes attainable, typical values for pure solvents being in the region 0.5–3 $cm^3/min\text{-}cm^2$ at 100 psi.

In this work, these new membranes were used to fractionate polyvinylpyrrolidone and dextran polysaccharide, two water-soluble polymers.

EXPERIMENTAL

The apparatus used for the investigation is shown in Figure 1. It consisted primarily of an Amicon Model 400 cell having a cell volume of ca. 450

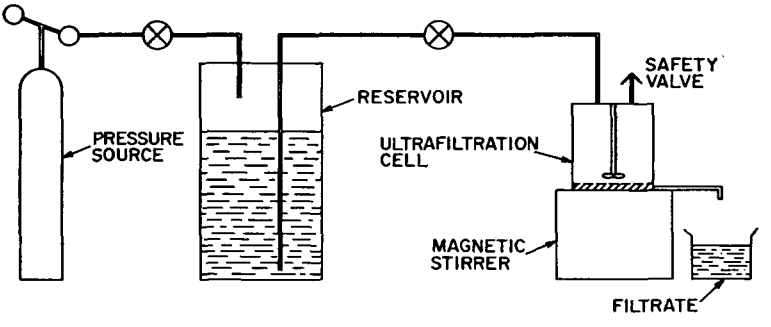

Fig. 1. Schematic diagram of a batch cell apparatus.

cm^3 and an effective membrane area of ca. 33 cm^2. The cell was provided with a magnetic stirrer to dissipate the filter cake of retained solutes which otherwise form on the membrane surface and which, if not removed, lower the flux. The membranes used were Amicon XM-4B and UM-1 Diaflo membranes (Amicon Corp., Lexington, Mass.) with molecular weight cutoffs for random coil polymers of ca. 50000 and 10000, respectively. Viscosities of the solutions were measured with an Ostwald viscometer at 25°C in order to characterize the molecular weights. The concentrations of the solutions were measured, to an accuracy of $\pm 0.01\%$, with a Brice-Phoenix differential refractometer.

The XM-4B membrane was initially stabilized at 100 psi by passing water through it for 10–15 min. During this time, there was a sharp drop in the flux due to compression of the membrane. This stabilization procedure was also followed with the UM-1 membranes. In this case, however, the water flux remained steady at its initial value. Typical fluxes with polymer solutions were in the region of 0.15–0.2 $cm^3/min\text{-}cm^2$ for both membranes. The two polymers fractionated were: (A) a polyvinylpyrrolidone (PVP), obtained from the Light Chemical Company, Type K25, with a molecular weight of 30,000 and (B) dextran polysaccharides, obtained from Pharmacia Fine Chemicals, Inc. who also provided the following molecular weight data: Dextran-40, $\bar{M}_w = 40000$, $\bar{M}_n = 25600$; Dextran-80, $\bar{M}_w = 86000$, $\bar{M}_n = 44000$; Dextran-110, $\bar{M}_w = 100000$, $\bar{M}_n = 62000$.

RESULTS AND DISCUSSION

Batch Fractionation with the Use of Increasingly More Retentive Membranes

The aim of this fractionation experiment was to produce a medium molecular weight sample of PVP containing no very high and no very low molecular weight fractions. The presence of these high and low molecular weight fractions has hitherto hindered the use of PVP as a blood expander.[8] A schematic outline of the experimental procedure is shown in Figure 2. In steps I and II, the high molecular weight fraction was removed with an XM-4B membrane, the lower molecular weight fractions being eluted in the filtrate. In step III the more retentive UM-1 membrane was used to retain the high molecular weight fraction and to pass the low molecular weight fraction. A similar fractionation procedure has been used by Blatt et al. with various protein mixtures.[9,10]

In step I, 4 liters of 2% polymer solution were concentrated with the XM-4B membrane to 425 cm^3.

In step II, the filtrate from step I was passed through another XM-4B membrane to yield a retentate of 450 cm^3 and a filtrate of approximately 3 liters.

In step III, the filtrate from step II was split into several portions for convenience. Each portion was then concentrated to half its initial volume with the UM-1 membrane. The retentate was then diluted back to the

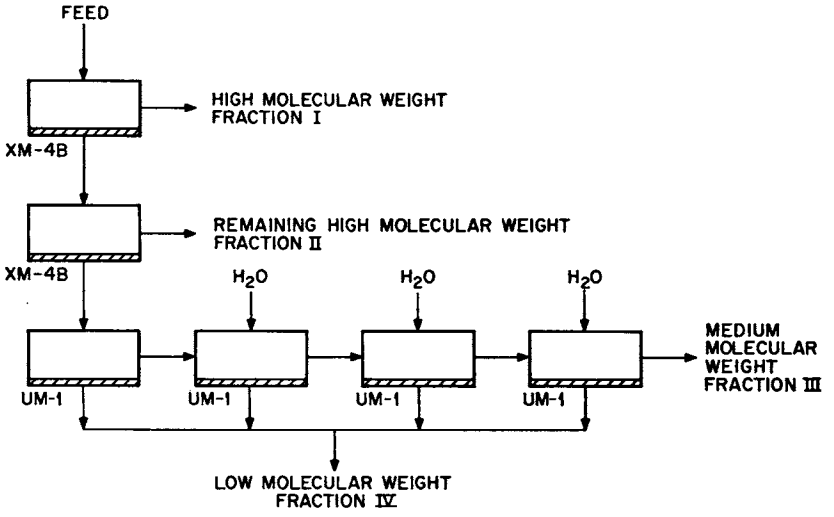

Fig. 2. Schematic flow diagram of the system used to fractionate PVP by batch fractionation with use of increasingly more retentive membranes.

initial volume and the process repeated three more times. In this way all the low molecular weight fraction was washed through the UM-1 membrane.

The results of this experiment are shown in Table I. During the first and second stages, most of the high molecular weight ends were removed. The intrinsic viscosity of the filtrate from these two stages is less than half that of the original. After removal of the low molecular weight ends in fraction IV the intrinsic viscosity of the remainder fraction III rose again slightly.

TABLE I
Fractionation Results Obtained with PVP

Fraction	Approximate % of original feed	Intrinsic viscosity, dl/g
Original material	—	0.60
Residue from first XM-4B membrane (fraction I)	35.0	0.75
Filtrate from first XM-4B membrane	—	0.47
Residue from second XM-4B membrane (fraction II)	30.0	0.50
Filtrate from second XM-4B membrane	—	0.29
Residue from UM-1 membranes (fraction III)	30.0	0.33
Filtrate from UM-1 membranes (Fraction IV)	5.0	<0.1

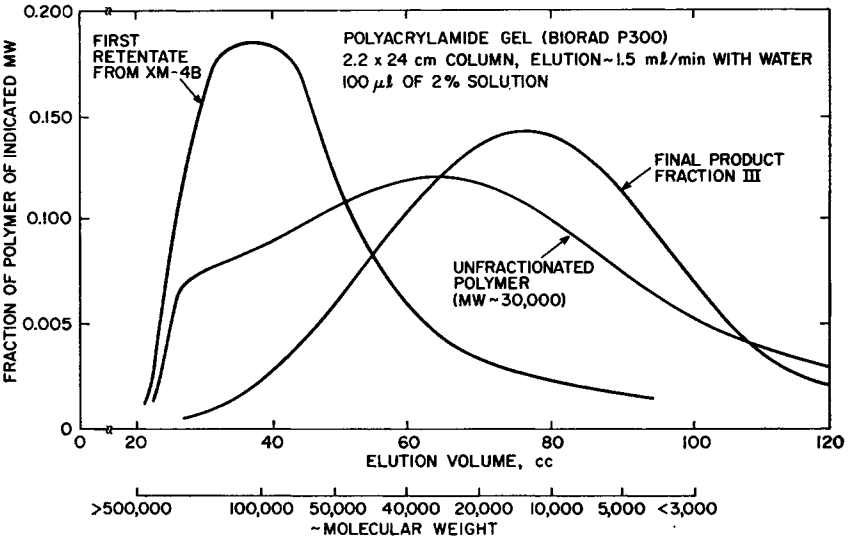

Fig. 3. Gel chromatography molecular weight distribution of ultrafiltered polyvinylpyrolidone fractions.

The intrinsic viscosity results were confirmed by the gel chromatography elution curves of the original material—the retentate from step I (fraction I) containing most of the high molecular weight polymer, and the final product (fraction III) containing no high and no low molecular weight polymer. These curves are shown in Figure 3.

Fractionation by Using Cells in Series

A second method of fractionation is that of connecting several cells in series, each cell containing the same type of membrane. Since ultrafiltration membranes have a retention spectrum rather than an absolute discrete cutoff, this arrangement operates in a manner analogous to a chromatographic column, i.e., it provides a method of obtaining continuous enrichment from a separation medium whose molecular discriminating capacity has a finite molecular weight band width. The sample is placed in the first cell and the filtrate from this cell passes directly into the second, and so on. Hence the smaller molecules, which are less effectively retained by the membranes, are separated from the larger molecules. The greater the number of cells, the greater the separation between the different fractions.

In order to test this technique, three cells were placed in series. Each cell had a volume of ca. 450 cm^3 and was fitted with an XM-4B membrane. The apparatus is shown schematically in Figure 4. The first cell was filled with a 2% solution of Dextran-80 and the other two cells with water. A pressure of 40 psi was then applied across the three-cell array and 7 liters of water passed through the system. The experiment was then stopped and the concentration and intrinsic viscosity of the material in all three cells and

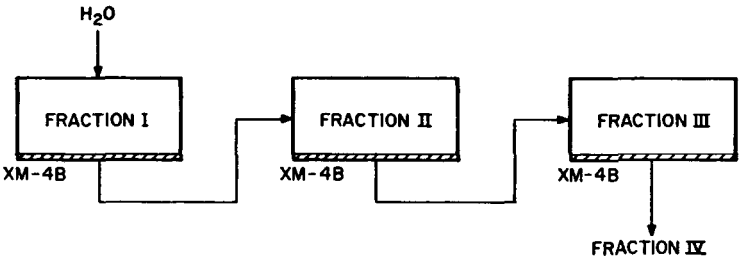

Fig. 4. Schematic flow diagram of the system used to fractionate Dextran-80 with the use of three cells in series.

in the filtrate measured. The experimental results are summarized in Table II. A considerable fractionation was obtained. The molecular weight of the four fractions as characterized by their intrinsic viscosities shows a steady fall from fraction I to fraction IV as we would expect.

These data are especially noteworthy when it is remembered that the original Dextran-80 is already well fractionated, having, according to the manufacturer a $\bar{M}_w/\bar{M}_n$ of less than 2.

Fractionation with the Use of Variable Pressure

Another method of fractionation utilizes the recently discovered dependence of the molecular weight cutoff of these membranes on the applied pressure. A quantitative measure of the retention characteristics of a membrane is given by the rejection coefficient R; thus

$$R = [1 - (c_f/c_B)] \times 100\% \quad (1)$$

where c_f denotes the concentration in the filtrate at time t and c_B the concentration in the feed solution at time t.

A high rejection coefficient shows that the macromolecular species cannot pass through the pores of a particular membrane, while a low rejection coefficient indicates the reverse. The rejection coefficient can be measured by filling a cell with a known concentration of liquid and then applying a fixed pressure. Sufficient filtrate is removed until the dead space between

TABLE II
Fractionation Results Obtained by Using Three Membranes in Series

Fraction	% of original Dextran-80[a]	Intrinsic viscosity, dl/g
Original Dextran-80 solution	—	0.11_7
First cell (fraction I)	26	0.15_3
Second cell (fraction II)	20	0.14_7
Third cell (fraction III)	44	0.11_1
Filtrate (fraction IV)	5	0.08_6

[a] 5% material loss.

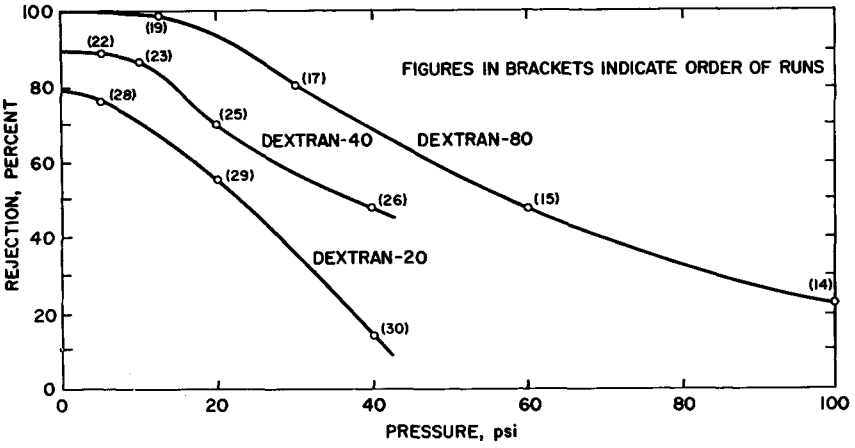

Fig. 5. Rejection as a function of pressure for 1% dextran solutions with an XM4B membrane.

the membrane and the collection cylinder has been adequately flushed (usually 25 cm^3). The next 5 cm^3 of filtrate is collected and the concentration of the solute in the sample measured. Since the bulk concentration is little changed by the removal of such a small volume of filtrate, the rejection coefficient may be directly calculated from eq. (1). The cell is then emptied. fresh dextran solution added, and the experiment repeated at a different pressure.

Typical data obtained in this way are shown in Figure 5. It is apparent that the rejection falls markedly with increasing pressure. It was also established that this is not a hysteresis effect. The phenomenon will be elaborated on further in a later paper. It follows from this effect that if a cell is filled with a polymer solution and several volumes of water passed through it at a low pressure, only the smallest molecules will be eluted into the filtrate. On raising the pressure, the next higher molecular weight fraction will be eluted, and so on. In this way a single membrane can be used to obtain a series of different molecular weight fractions.

TABLE III
Fractionation Results Obtained by Varying the Applied Pressure Across a Membrane

Pressure at which fraction was obtained, psi	% of original dextran[a]	Intrinsic viscosity, dl/g
5	19	0.08_2
10	12	0.08_7
20	14	0.10_3
40	16	0.10_9
Residue in cell	35	0.12_2
Original dextran-40 solution	—	0.10_5

[a] 4% material loss.

A 2% dextran-40 solution was fractionated in this way by using an XM-4B membrane. The results are given in Table III. A good fractionation has again been obtained, the molecular weight of successive fractions (as characterized by the intrinsic viscosity) rising in the expected order. The fractionation obtained was not as good as that with the three cells in series.

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References

1. H. Tampa, *Polymer Solutions*, Butterworths London, 1956.
2. J. Cayes, *J. Chem. Educ.*, **43**, A567 (1966).
3. L. C. Craig, *Advan. Anal. Chem. Instrumentation*, **4**, 35 (1965).
4. J. D. Ferry, *Chem. Rev.*, **18**, 373 (1935).
5. C. J. O. R. Morris, and P. Morris, *Separation Methods in Biochemistry*, Pitman, 1964.
6. F. Migasato and V. E. Pollak, *J. Lab. Clin .Med.*, **67**, 1036 (1966).
7. H. J. Preusser, *Kolloid-Z.*, **218**, 129 (1967).
8. L. W. Burnette, *Proc. Sci. Sect. Toilet Goods Association*, **38**, 1 (1962).
9. W. F. Blatt, B. G. Hudson, S. M. Robinson, and E. M. Zipilivan, *Nature*, **216**, 512 (1967).
10. W. F. Blatt, S. M. Robinson, F. M. Robbins, and C. A. Saravis *Anal. Biochem.*, **18**, 81 (1967).

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