Molecular concentrators measure colloid osmotic pressure

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In a recent publication, colloid osmotic pressure (C.O.P.) measurements and results for other related variables in the serum of wild animals were reported (Hattingh, de Vos, Bomzon, Marcus, Jooste & Chertkow 1980). Such measurements provide valuable information relating to the forces involved in capillary filtration on a comparative basis. Although various formulae have been published by which theoretical values for C.O.P. may be calculated making use of values for total protein concentration, A/G ratios, etc., (Weisberg 1978), we have shown that C.O.P. can only be accurately determined by measurements (Hattingh *et al.* 1980).

Various methods have been published to measure C.O.P. These vary from screw-on osmometers (Aukland & Johnson 1973; Hansen 1961; Prather, Gaar & Guyton 1968; Wiederheim, Lee & Stromberg 1973) to needle-type (Kakiuchy, Arai, Horimoto, Kikuchy & Koyama 1979) and implantable osmometers (Reed 1979; Reed & Aukland 1978; Wiederheim *et al.* 1973). Common to all these methods is the use of ultra-filtration membranes. A major drawback in all cases is the distinct possibility of membrane fracture during assembly with resultant pressure leakage and thus underestimation of C.O.P. If this can be overcome the measurement of C.O.P. could become routine. Millipore (Bedford, Mass.) make an immersible molecular concentrator consisting of a Pellicon filtration membrane cast on a cylindrical polyethylene core (Figure 1). The retentive surface of the membrane (nominal molecular mass limit of 10 000 daltons) faces outward. Attached to the core is a polyethylene cap. The membrane surface area is 13 cm^2. Because of the molecular mass limitation, the concentrator is normally immersed into a solution and used to concentrate samples of low protein content by applying suction to the polyethylene cap. Water and dissolved molecules of molecular mass less than 10 000 are thus removed and the protein solution concentrated (e.g. urine). The concentrator can withstand temperatures up to 50 °C and may be sterilized with ethylene oxide.

The concentrator has all the requirements for an osmometer. By applying suction and immersing it into a 0,9% NaCl solution all the air inside the polyethylene core may be removed. The concentrator is then connected to a suitable pressure transducer and recorder with saline filled tubing and is ready for use. Calibration, as is the case with other osmometers, is performed in one of two ways.

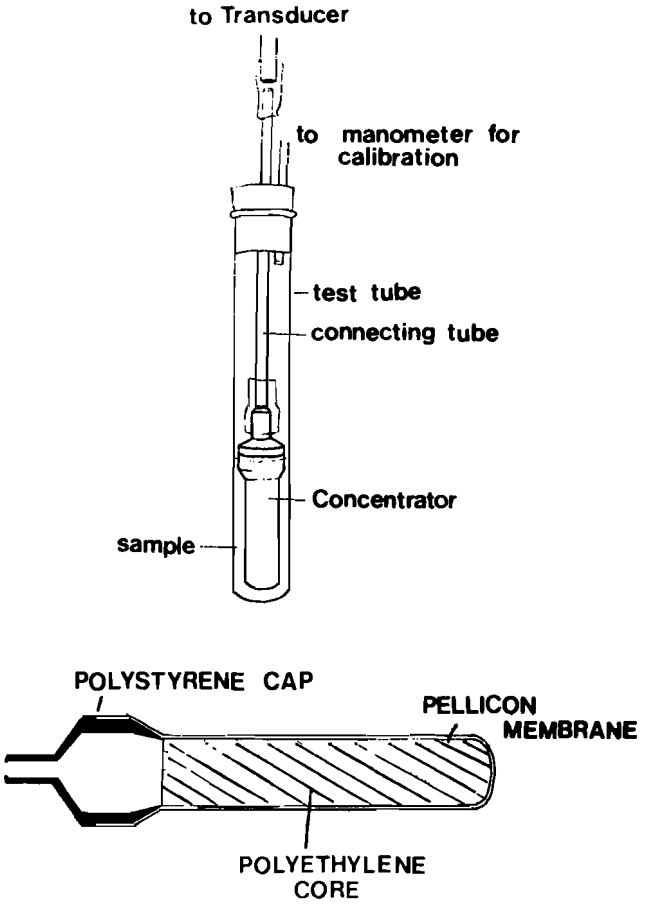

Figure 1 Diagrammatic representation of the Millipore concentrator.

The osmometer is either placed in a series of bovine albumin solutions of known C.O.P. or airtightly sealed in a test tube containing saline connected to a sphyghmomanometer. In the latter instance, any pressure applied to the saline is transmitted to the pressure transducer and is duly recorded.

The C.O.P. of various bovine albumin solutions (BDH) ranging from 20 g/ℓ to 100 g/ℓ were measured with the concentrator and compared to the results obtained with the flow-through osmometer of Prather *et al.* (1968), fitted with a PM 10 (Amicon) membrane. Results from both osmometers were compared to those obtained by calculation (Landis & Pappenheimer 1963) (Figure 2). The measured pressures did not differ significantly at the 1% level from one another but did deviate from the calculated values on average by 30%.

An example of an actual recording is shown in Figure 3. The response time of the concentrator compares well with that of the osmometer, both equilibrating within 30 to 60 s. Reproducibility was found to be within a 1% limit. If temperature control is required the samples to be measured may be placed in a waterbath.

The design of the present osmometer is obviously much simpler than that of other osmometers (Aukland & Johnson 1973; Prather *et al.* 1968). It compares in respect of reliability, response time and reproducibility. In addition it is inexpensive, much less delicate, easier to assemble, the membrane is pre-cast and it may be used with routine laboratory pressure transducers and recorders. With a decrease in size, it would be suitable for microsample analysis, in which case its application can be ex-

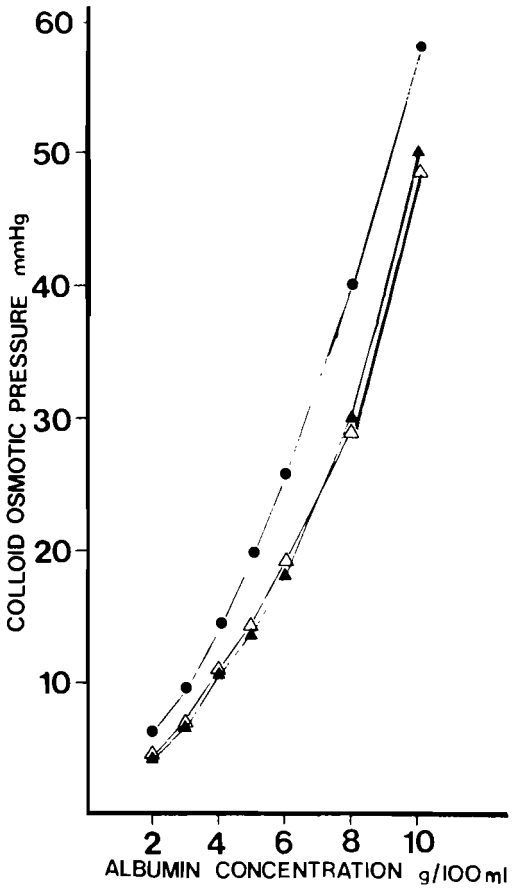

Figure 2 C.O.P. of albumin solutions obtained with the concentrator (Δ—Δ), Prather *et al.* osmometer (▲—▲) and by calculation (●—●). For clarity S.D. is not shown (n = 6 for each set of measurements).

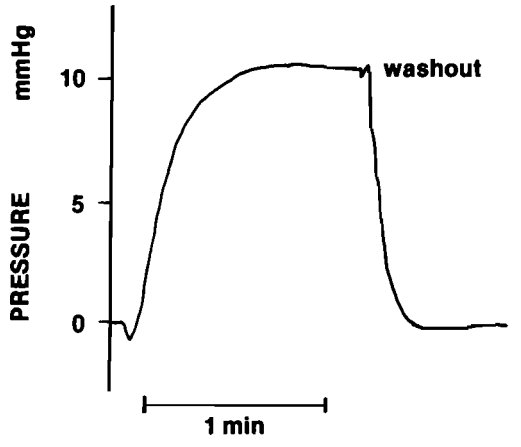

Figure 3 C.O.P. of an albumin solution measured with the concentrator showing response time.

tended to the measurement of C.O.P. of plasma, lymph and tissue fluid samples from small laboratory animals.

The concentrator also manifests potential in being adapted as an implantable osmometer such as described by Reed (1979). The polyethylene core would resist any deformatory forces applied on the membrane, ensuring that an equilibrium is obtained which would accurately reflect tissue C.O.P. The only possible disadvantage at present is that care must be exercised not to allow air into the system (through the membrane) and the concentrator should thus always be stored in saline.

It is therefore apparent that the concentrator provides a means for rapid, accurate estimation of C.O.P. On a comparative basis much remains to be learnt concerning this important variable, not only from plasma but also other body fluids. The methodology reported here, makes this possible.

Acknowledgement

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References

AUKLAND, K. & JOHNSON, H.M. 1973. A colloid osmometer for small fluid samples. *Acta. Physiol. Scand.* 19: 485 – 490.

HANSEN, A.T. 1961. A self-recording electronic osmometer for quick, direct measurements of colloid pressure in small fluid samples. *Acta. Physiol. Scand.* 53: 197 – 213.

HATTINGH, J., DE VOS, V., BOMZON, L., MARCUS, E., JOOSTE, C. & CHERTKOW, S. 1980. Comparative physiology of colloid osmotic pressure. *Comp. Biochem. Physiol.* 67A: 203 – 206.

KAKIUCHY, Y., ARAI, T., HORIMOTO, M., KIKUCHY, Y. & KOYAMA, T. 1979. A new needle-type colloid osmometer for continuous determination of blood oncotic pressure. *Am. J. Physiol.* 236: F419 – F422.

LANDIS, E.M. & PAPPENHEIMER, J.R. 1963. Exchange of substances through capillary walls. In: Handbook of Physiology. Circulation. Sect. 2, Vol. II. 961 – 1034. *Am. Physiol. Society.*

PRATHER, J.W., GAAR, K.A. & GUYTON, A.C. 1968. Direct continuous recording of plasma colloid osmotic pressure of whole blood. *J. Appl. Physiol.* 24: 602 – 605.

REED, R.K. 1979. An implantable colloid osmometer. *Microvas Res.* 18: 83 – 94.

REED, R.K. & AUKLAND, K. 1978. An implantable osmometer for determination of interstitial fluid colloid osmotic pressure of serum proteins. *Acta. Physiol. Scand.* 102: 46A.

WEISBERG, H.F. 1978. Osmotic pressure of serum proteins. *Ann. Clin. Lab. Sci.* 8: 155 – 164.

WIEDERHEIM, C.A., LEE, D.R. & STROMBERG, D.D. 1973. A membrane osmometer for microliter samples. *J. Appl. Physiol.* 35: 432 – 435.

A pellet mould for small powdery specimens for X-ray microanalysis in the SEM

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This paper describes a simple mould to press small powdered samples into pellets for X-ray microanalysis. The pellet can be pressed on the sample holder (stub), but in some cases it has to be glued to the sample holder or stub. The drawings indicate how the mould is made (Figure 1).