(Final)   1097

A NON-INVASIVE CONTINUOUS METHOD OF MEASURING

BLOOD VOLUME DURING HAEMODIALYSIS

USING OPTICAL TECHNIQUES.

M P McMahon[1,2], S B Campbell[3],

G F Shannon[2], J S Wilkinson[4], S J Fleming[3]

[1] Department of Physical Sciences, Royal Brisbane Hospital

[2] Department of Electrical and Computing Engineering, Queensland University

[3] Department of Renal Medicine, Royal Brisbane Hospital

[4] Department of Electronics and Computer Science, University of Southampton

Author for Correspondence: Dr. S. J. Fleming,

Department of Renal Medicine.

Royal Brisbane Hospital.

Herston Road.

Herston.   4029.

Australia.

ABSTRACT

Hypotension during haemodialysis and fluid overload between treatments are major problems for haemodialysis patients. Clinical means of assessing hydration state can be relatively imprecise. We describe a non-invasive method of measuring absolute blood volume (BV) during a mock in vitro haemodialysis session which adds objective information to that assessment.

As fluid is removed by ultrafiltration, haemoglobin concentration [Hb] rises proportionately to the fall in BV. An optical monitor clamped across the transparent dialysis tubing gives a continuous readout of near infra-red light transmitted through the blood, and this can be converted to [Hb] values. The net change in BV is the difference between the volume of fluid ultrafiltered and the volume which refills the vascular compartment from the extravascular space. By analysing the change in [Hb] and therefore the change in BV at two different rates of fluid removal the absolute BV can be determined. The accuracy of this method was tested in vitro.

This optical method accurately measures the change in BV over a range of [Hb] from 4 to 15 g/dL and blood circulation pump speeds of 150 to 300 ml/min. A series of ten in vitro experiments was performed. The mean relative difference between the measured BV and the calculated BV, was $5.7 \pm 2.5\%$.

This readily repeatable technique can accurately measure BV during a mock in vitro haemodialysis session, thus providing information for the clinical assessment of the hydration state. Information from these experiments will assist in future in vivo studies.

1. INTRODUCTION

Patients with end-stage renal failure are unable to regulate salt and water balance. Their medical treatment involves careful assessment of their hydration state, so that an appropriate amount of fluid can be removed by ultrafiltration during haemodialysis. Appropriate fluid removal prevents the complication of fluid overload or depletion, and helps to control blood pressure. Hypovolaemia is one of the major contributors to the symptomatic hypotension which complicates 20 to 30% of dialyses.[1]

A clinical assessment of hydration is made by monitoring blood pressure and other signs of excessive or inadequate fluid. A number of devices are being developed to continuously monitor changes in BV to assist in this process. These involve the measurement of viscosity,[2] protein concentration,[3] haemoglobin concentration[4,5] or electrical conductivity[6] of the blood.

A method described by Schneditz[7] was used to measure absolute BV, but requires the use of a sterile in-line device incorporated into the dialysis circuit. Furthermore, the measurements of absolute BV obtained were inadequately validated in that they were compared to those calculated from anthropometric data taken from normal subjects.

Previously described techniques[2-6] only allowed the measurement of changes in BV. We have modified the near infra-red optical monitor described previously[5] to establish a non-invasive technique for measuring absolute BV, rather than simply changes in BV. The theoretical basis for the use of this technique and the results of *in vitro* validation studies are the subject of this paper.

2. PRINCIPLES OF ABSOLUTE BLOOD VOLUME MEASUREMENT

Light travelling through a medium will be attenuated by scattering and absorption effects. At wavelengths in the near infra-red range, haemoglobin, Hb and oxyhaemoglobin, HbO_2 are the main absorbing components.[10] If scattering is neglected, the attenuated power, P_2 can be calculated using a modification of Beer's Law:

$$P_2 = P_1 e^{-(\epsilon.[Hb]+\rho).L} \tag{1}$$

where P_1 is the incident power of the light, ϵ is the coefficient of absorption of the component of concentration c, L is the distance the light travels through the medium and ρ is a scattering factor added by Janssen[11] for the case of a whole blood medium as reflection and refraction of incident light passing through whole blood causes additional attenuation.

As discussed previously[5], by measuring the ratio of the attenuated to the incident power at the isobestic wavelength, changes in the blood volume can be calculated.

If it is assumed that [Hb] changes only because of fluid movement into or out of the vascular compartment, and if BV(0) and BV(t) are the blood volumes at times 0 and t minutes respectively, and [Hb(0)] and [Hb(t)] are the haemoglobin concentrations at times 0 and t minutes respectively, then:

$$\frac{BV(0)}{BV(t)} = \frac{[Hb(t)]}{[Hb(0)]} \tag{2}$$

which we shall subsequently refer to as the proportional BV.

In a patient undergoing haemodialysis the system is not closed. As fluid is removed from the intravascular space during ultrafiltration, a lesser volume of fluid is drawn from the extravascular space partially by the increase in plasma protein osmotic (oncotic) pressure that results, and refills the intravascular compartment.

The rate of change of BV is equal to the refilling rate, Q_r, less the ultrafiltration rate, Q_u.

$$\frac{d\ BV(t)}{dt} = Q_r - Q_u \tag{3}$$

If Q_r is assumed to remain constant for a short period of time before and after a change in ultrafiltration rate, the unknown refilling rate can be eliminated from the calculations. Using 1 and 2 to denote rates before and after a change in ultrafiltration rate, equation (3) may be written for both time periods and the difference found:

$$\left[\frac{d\ BV(t)}{dt}\right]_2 - \left[\frac{d\ BV(t)}{dt}\right]_1 = Q_{u_1} - Q_{u_2} \tag{4}$$

Because the absolute BV is not known, but the proportional BV (and its rate of change) is, both sides are divided through by the BV at time t=0, BV(0), to give the known values:

$$\left[\frac{d\frac{BV(t)}{BV(0)}}{dt}\right]_2 - \left[\frac{d\frac{BV(t)}{BV(0)}}{dt}\right]_1 = \frac{Q_{u_1} - Q_{u_2}}{BV(0)} \tag{5}$$

The left hand side now contains only the rate of change of the proportional BV which can be obtained from the output of the optical monitor and a limited number of blood samples to calibrate its output. The two ultrafiltration rates are known, leaving only the absolute BV at time t=0 to be found by rearranging equation (5) as:

$$BV(0) = \frac{Q_{u_1} - Q_{u_2}}{\left[\dfrac{d\dfrac{BV(t)}{BV(0)}}{dt}\right]_2 - \left[\dfrac{d\dfrac{BV(t)}{BV(0)}}{dt}\right]_1} \qquad (6)$$

The denominator of this equation is the difference between the rates of change of the proportional BV at the two different ultrafiltration rates. This calculation can be simplified if one of the ultrafiltration rate is zero.

3. METHODS

3.1 Optical Monitor Design

The optical monitor is based on the design of Wilkinson et al.[5] A schematic diagram of the electronic circuit is illustrated in Figure 1. The GaAlAs light emitting diode (LED) used has a peak spectral output of 822 nm, which exceeds the isobestic wavelength of 805 nm, but is the closest readily available commercial device. We have previously demonstrated *in vitro* that the error introduced into calculations of BV is insignificant.[5] A square wave of 200 Hz with a precise 1:1 mark to space ratio is used as a means of minimising the ambient light effects. In order to reduce the effects of long-term drift of the transmitted power, the output was determined as ratio of the received signal transmitted through the blood and a reference signal taken prior to transmission. The

reference signal originates from a secondary photodetector described previously.[5]

3.2 Monitor Calibration

A number of blood samples was taken during each experiment to calibrate the received optical signal. [Hb] was measured using a Coulter S Counter and plotted against the natural log of the normalized output signal, $Ln(P_a)$. An example of this is shown in Figure 2(a). Linear regression analysis was used to determine the relationship so that the [Hb] could be calculated for any optical signal measured during that experiment. The monitor was recalibrated in this way for each experiment. Five, seven or ten samples were taken in earlier studies but a negligible improvement in accuracy resulted when using more than five samples. It was therefore decided to base the calibration on five blood samples.

4. MONITOR PERFORMANCE

4.1 Scattering at Low Haemoglobins

Janssen[11] suggested that Beer's Law would cease to hold at low [Hb] because of scattering effects. In order to validate our use of linear regression analysis to calculate [Hb] from the monitor's output, as described in section 3.2, we sought to demonstrate that within the accuracy required a linear relationship existed at all physiological [Hb] levels.

We examined this by measuring the received optical signal, while the [Hb] of the blood circulating in our experimental circuit was serially reduced by the infusion of physiological normal saline (0.9% NaCl).

A peristaltic pump circulated blood, from a reservoir of approximately 500ml, through PVC tubing. The optical head, monitoring [Hb] changes was attached to this tubing. The blood was returned to the reservoir where it was mixed with a magnetic stirrer. Aliquots of normal saline were slowly added so that the [Hb] was reduced by approximately 1 g/dl between each reading. Time was allowed for mixing between the saline additions. [Hb] and optical response were recorded at the start of the test and following each dilution. Figure 2(b) illustrates the optical response from one of these tests.

At low [Hb] (below 2g/dL), the transmitted light saturated the detector. To overcome this, different optical filters [KODAK Wratten Gelatin Filters] were placed in the optical head to reduce the signal by a known factor. The optical head was modified to enable the filters to be placed in the beam path with no distortion to the tubing.

The results illustrate that scattering is an influence on the readings, causing nonlinearity particularly at [Hb] levels outside the range of 3-10 g/dL. A linear relationship between [Hb] and $Ln(P_a)$ can be assumed over the small range of [Hb] variation usually encountered in haemodialysis procedures (approximately 1g/dL). This was verified by the strong linear correlations between [Hb] and $Ln(P_a)$ [$r > 0.98$] we achieved with each test.

4.2 Pump Speed Effect

A similar experiment to that described in 4.1 was performed with serial dilution of 500ml of whole blood to test whether the speed of the circulation pump would affect the monitor's reading. At each [Hb] level, the transmitted power was measured at three different pump speeds (150, 200, 300 ml/min), all in the range that would normally be used for haemodialysis.

Changes in pump speed had no appreciable influence on the optical response with a maximum apparent change in [Hb] of 0.06 g/dl observed based on a calibration curve calculated using the 200ml/min output. These results are illustrated in Table 1. Pump pulsatility effects were easily filtered out of the signal at all times.

4.3 Monitor Position Effects

The experimental system was the same as that described in 4.1 and 4.2 except this time a haemofilter (Gambro, FH88H) was incorporated into the circuit. Optical heads were placed at various positions around the test circuit. It was found that the head had to be positioned between the blood reservoir and the haemofilter. Because blood with a higher [Hb] coming out of the haemofilter had not had the opportunity to mix with the total blood reservoir, the head gave unreliable results when placed after the haemofilter.

5.0 Blood Volume Measurement

5.1 Methods

Blood from a reservoir of approximately 1500ml was circulated via a peristaltic pump through a circuit containing a haemofilter (Gambro FH88H) (Figure 4). The blood in the reservoir was constantly mixed by a magnetic stirrer.

A second pump, attached to the ultrafiltrate port of the haemofilter, removed fluid at 8 ml/min. As the BV used in these studies was approximately one quarter to one third of that of an adult haemodialysis patient, the ultrafiltration rate was chosen to be a similar fraction of the 30 ml/min to be used in future *in vivo* studies.

A separate rate-controlled pump allowed the infusion of physiological normal saline into the blood flask to simulate the internal vascular refilling which occurs when fluid is removed by ultrafiltration *in vivo*.

Ten experiments were performed to compare calculated and measured BV (BV_{calc} and BV_{meas}). After allowing the blood to circulate for ten minutes to ensure that it was completely mixed, ultrafiltration was carried out for thirty minutes. In five of these experiments there was no infusion of saline into the reservoir (no refilling) while in the other five a refilling rate of 4 ml/min was used. The optical monitor signal was recorded at a sampling rate of one per second. Five blood samples were taken at regular time intervals to measure [Hb] for calibration of the monitor.

Ultrafiltration was stopped after 30 minutes and the optical signal was recorded for a further ten minutes. The blood was then removed from the circuit and its volume measured. The result was compared with the volume calculated using equation 9 with Qu_1= 8 ml/min and Qu_2= 0 ml/min.

5.2 Results

A sample of the optical monitor's output from one of the five experiments in which saline was being infused at 4 ml/min while ultrafiltration was progressing is shown in Figure 5.

Results of the ten experiments performed are summarised in Table 2. The mean relative percentage difference between BV_{calc} and BV_{meas}, determined as $[(BV_{calc} - BV_{meas}) * 100 / BV_{meas}]$, was 5.7 ± 2.5%. The largest discrepancies between BV_{meas} and BV_{calc} were found in tests 6 and 9. In test 6 older stored blood was used and the presence of haemolysed blood caused some haemoglobin to be transferred with the ultrafiltrate. In test 9 there was an unidentified problem with the system. The optical signal clearly did not follow the pattern seen in other experiments resulting in a larger error. Even though the exact cause of this problem is unknown, it was obvious during the performance of the experiment that the signal was spurious and if carried out on a patient would have been disregarded and repeated.

6. DISCUSSION

Despite the importance of absolute BV in the development of symptomatic hypotension during dialysis, currently there are no objective and easily repeatable methods of measuring this variable. The best method available is measurement of red cell and plasma volumes by dilution of radioactively labelled blood elements. The time consuming nature of this test and the exposure to radiation makes it unsuitable for routine clinical practice. An alternative technique[7] has not been validated against other objective methods of BV measurement.

The present study has validated, *in vitro*, a non-invasive optical technique to measure absolute BV that can be carried out quickly and inexpensively and involves no radioactive materials.

The *in vitro* system designed to test the method represented a haemodialysis circuit as closely as possible. A volume of 1500 ml was circulated, approximately one quarter of the average adult's blood volume. The rate of ultrafiltration used, 8 ml/min, was reduced by a similar fraction from 30 ml/min as used in *in vivo* tests. Fluid was infused into the system during five of the studies to simulate the plasma refilling that occurs *in vivo* during ultrafiltration. This method reliably estimates the volume of blood *in vitro* with a mean relative difference of 5.7 ± 2.5% over a series of experiments.

These *in vitro* tests provided relatively noise free signals as indicated in Figure 4. Calculation of the rate of change was thus easy. With *in vivo* studies, more noise is

present, and rates of change need to be averaged over a number of samples (5000 will be used.)

Different pump speeds and monitor positions were tested and were found not to influence the results. A recent report[12] has shown that the optical attenuation of blood changes significantly with flow rate in the case of relatively low blood velocity. This confirms our earlier observation[5] that, for our large bore tubing, flow rates less than 150 ml/min should not be used if flow-rate dependent attenuation is to be avoided. The flow rates used in the present work correspond to blood velocities between 1.5 and 2.5 times the maximum employed by Lindberg. At flow-rates above 150 ml/min it is expected that phenomena such as the orientation and deformation of red cells, which affect optical scattering, will be approaching asymptotic values, and will not change substantially for flow-rates up to 250 ml/min.

The potential influence of changes in red cell volume on received optical signal was avoided by carrying out the study using ultrafiltration alone without concurrent dialysis. This widely used clinical technique does not change the osmolality of the filtered fluid significantly. It is change in osmolality, particularly that due to change in serum sodium, that can cause alterations in red cell volume during dialysis.[13]

This technique has the potential to be used in clinical nephrology to assist the assessment of a patient's state of hydration, thereby helping to predict and avoid some of the complications that can occur if incorrect amounts of fluid are removed.

REFERENCES

1) HENDERSON LW, KOCH, AM, DINARELLO CA, SHALDON S. Haemodialysis hypotension: the interleukin hypothesis. *Blood Purification* 1983; 1: 3-8.

2) GREENWOOD RN, ALDRIDGE C, CATTELL WR. Serial blood water estimations and in-line viscometry: The continuous measurement of blood volume during dialysis procedures. *Clin Sci* 1984; 66: 575-583.

3) SCHNEDITZ D, POGGLITSCH H, HORINA J, BINSWANGER U. A blood protein monitor for the continuous measurement of blood volume changes during haemodialysis. *Kidney Int* 1990; 38: 343-346.

4) SCHALLENBERG U, STILLER S, MANN H. A new method of continuous haemo-globinometric measurement of blood volume during haemodialysis. *Life Support Systems* 1987; 5: 293-305.

5) WILKINSON JS, FLEMING SJ, GREENWOOD RN, CATTELL WR, and ALDRIDGE C. Continuous measurement of blood hydration during ultrafiltration using optical methods. *Med & Biol Eng & Comput* 1987; 25: 317-323.

6) DE VRIES PMJM, KOUW PM, MEIJER JH, OE LP, SCHNEIDER H, DONKER AJM. Changes in blood parameters during hemodialysis as determined by conductivity measurements. *Trans Am Soc Artif Intern Organs* 1988; 34: 623-626.

7) SCHNEDITZ D, ROOB J, OSWALD M, POGGLITSCH H, MOSER M, KENNER T, BINSWANGER U.   Nature and rate of vascular refilling during hemodialysis and ultrafiltration   *Kidney Int* 1992; **42**: 1425-1433.

8) LINS LE, HEDENBORG G, JACOBSON H, SAMUELSON K, TEDNER B, ZETTERHOLM UB, LJUNGQVIST O. Blood pressure reduction during haemodialysis correlates to intradialytic changes in plasma volume. *Clin Nephrol* 1992; **37**: 308-313.

9) KIM KE, NEFF M, COHEN B, et al.   Blood volume changes and hypotension during hemodialysis. *Trans Am Soc Artif Intern Organs* 1970; **16**: 508-514.

10) ANDERSON NM, SEKELJ, P. Studies on light transmission of nonhaemolysed whole blood. Determination of oxygen saturation. *J Lab & Clin Med* 1965; **65**: 153-166.

11) JANSSEN F.   A study of the absorption and scattering factors of light in whole blood. *Med & Biol Eng* 1972; **10**: 231-240.

12)LINDBERG L-G, ÖBERG PA. Optical properties of blood in motion. *Optical Engineering* 1993; **32**: 253-257.

13) FLEMING SJ, WILKINSON JS, ALDRIDGE C, GREENWOOD RN, MUGGLESTON SD, BAKER LRI, CATTELL WR. Dialysis-induced change in erythrocyte volume; effect on change in blood volume calculated from packed cell volume. *Clin Nephrol* 1988; **29**: 63-68.

CAPTIONS TO FIGURES:

Figure 1 Optical Monitor Block Diagram.

Figure 2(a) A typical calibration curve obtained from the blood samples: The natural log of the normalized output signal, Ln (P_a) as a function of [Hb].

Figure 2(b) Optical Monitor Response to changing [Hb], showing Ln(P_a) as a function of [Hb].

Figure 3 *In Vitro* Blood Volume Measurement Circuit.

Figure 4 Plot of experimental test data with a pump flow rate of 200 ml/min, ultra-filtration rate of 8 ml/min and a refilling rate of 4 ml/min.
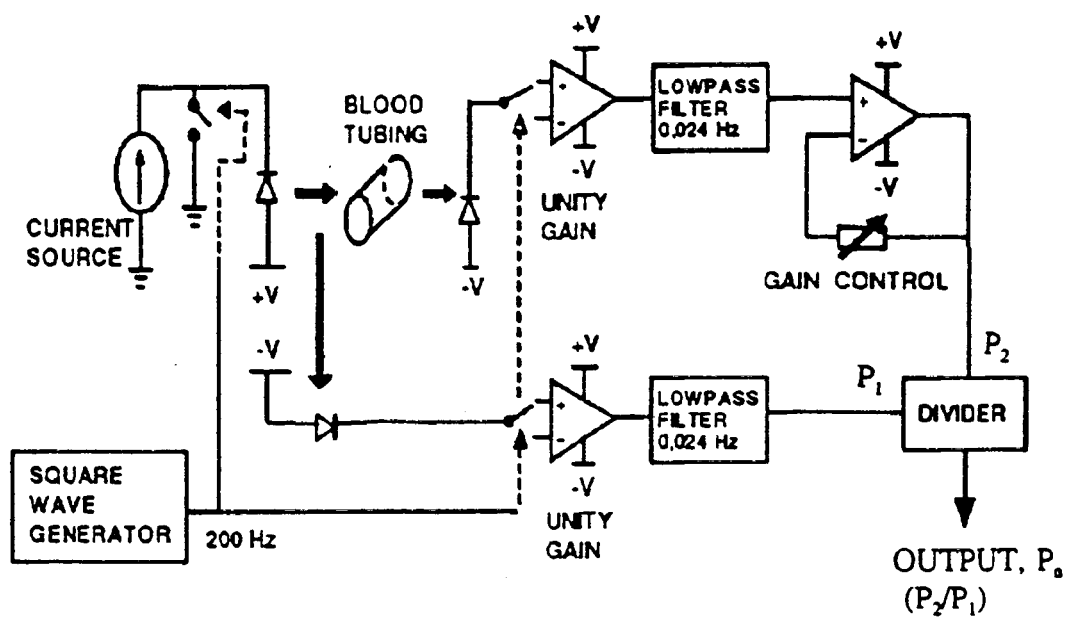
RUNNING HEADLINE:

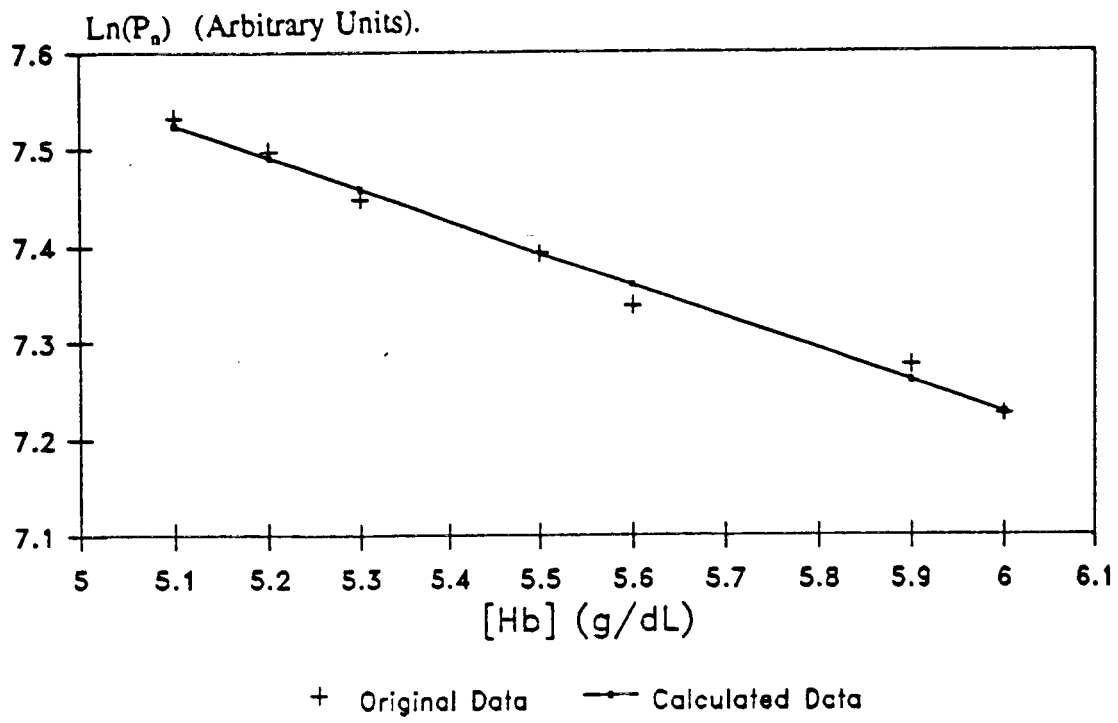Blood Volume During Haemodialysis.
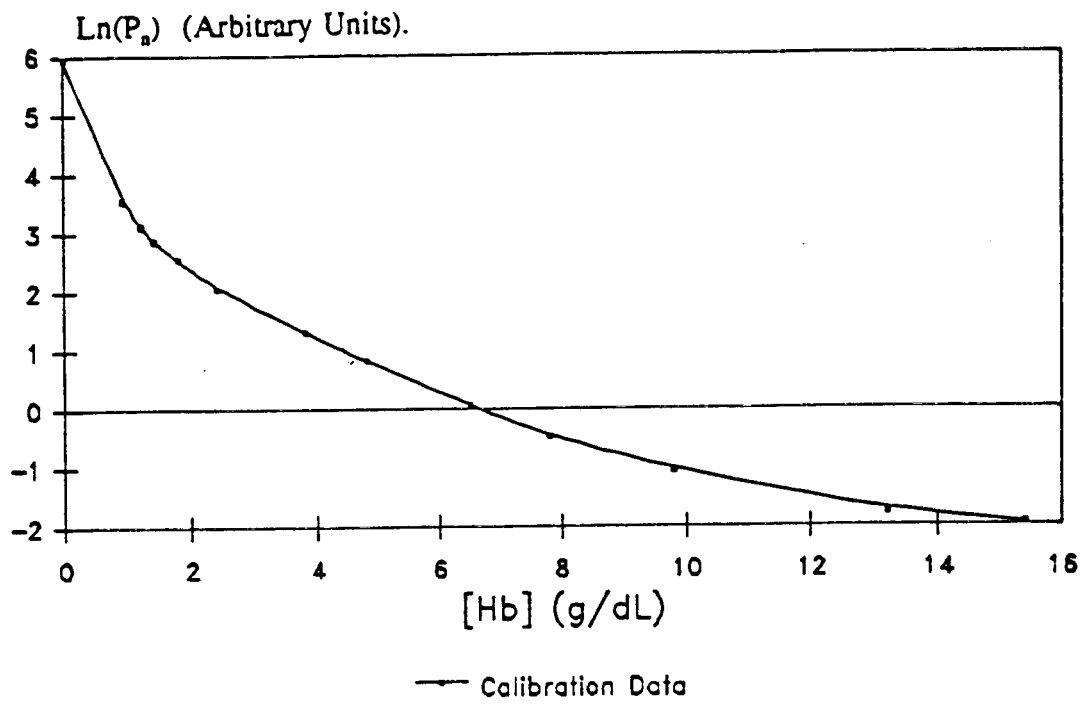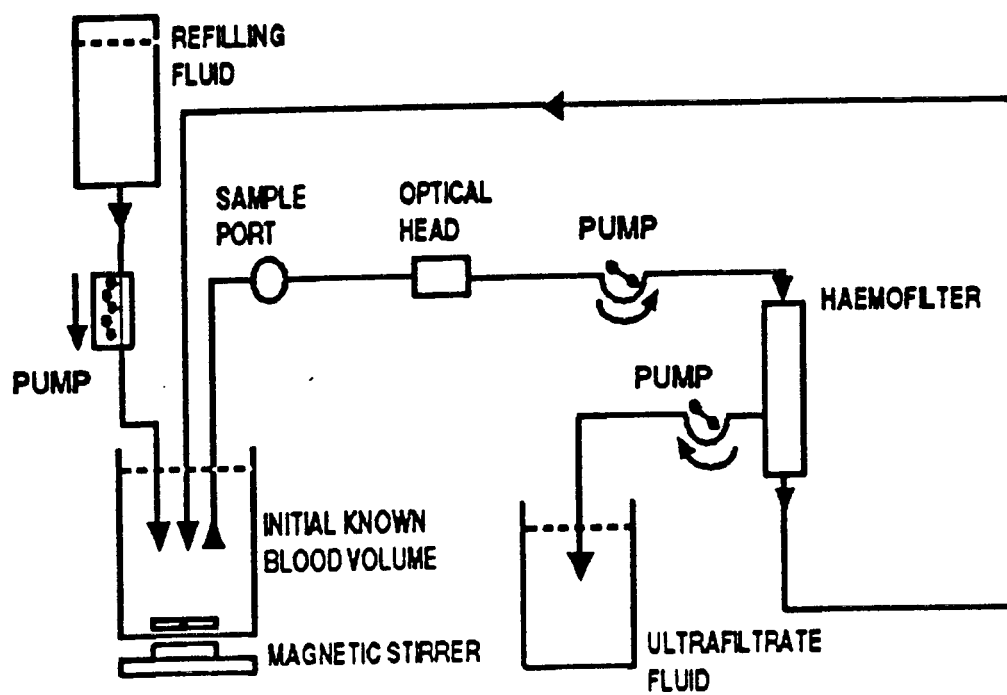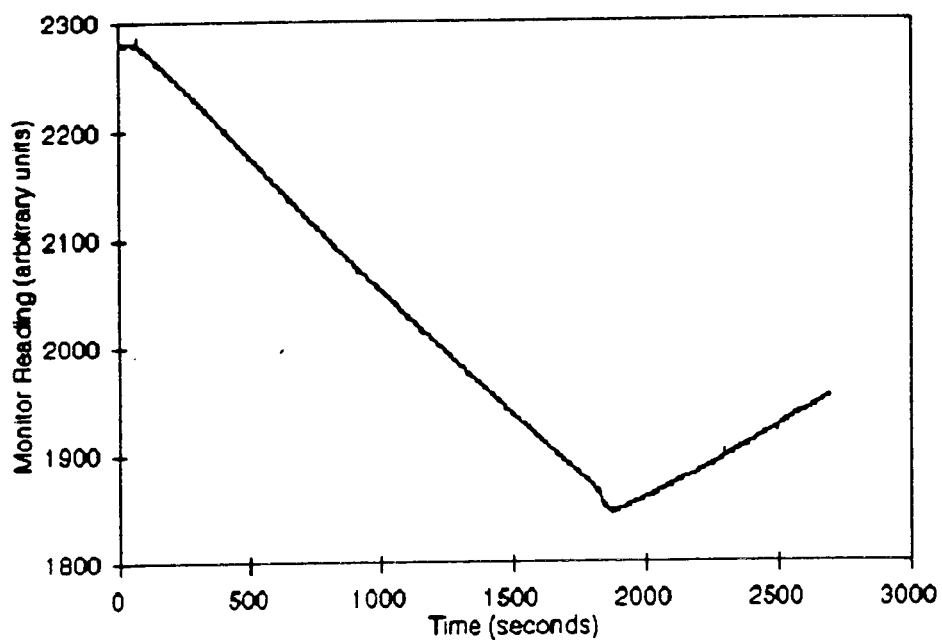
Figure 1

Figure 2(a)

Figure 2(b)

Figure 3

Figure 4

Table 1 Output from the optical monitor for selected [Hb], at three different pump speeds - 150, 200, 300 ml/min.

MEASURED [Hb]	SIGNAL OUTPUT FROM OPTICAL MONITOR		
	150 ml/min	200 ml/min	300 ml/min
7.4	3.19	3.17	3.14
6.9	3.51	3.52	3.52
6.1	4.22	4.21	4.18
5.2	5.5	5.47	5.47
3.9	8.41	8.38	8.32

Table 2 *In vitro* experimental results for tests with ultrafiltration only and tests with a refilling component. BV_{meas} = measured blood volume, BV_{calc} = calculated blood volume.

Test	Procedure	BV_{meas} (ml)	BV_{calc} (ml)	$\dfrac{BV_{calc}-BV_{meas}}{BV_{meas}} \times 100$
1	Ultrafiltration	1655	1715	3.6
2	Ultrafiltration	1630	1497	-8.2
3	Ultrafiltration	2040	1969	-3.5
4	Ultrafiltration	1515	1563	3.2
5	Ultrafiltration	1635	1552	-5.0
6	Ultrafiltration and Refilling	1675	1811	8.1
7	Ultrafiltration and Refilling	1680	1761	4.8
8	Ultrafiltration and Refilling	1643	1700	3.5
9	Ultrafiltration and Refilling	1211	1087	-10.2
10	Ultrafiltration and Refilling	1617	1727	6.7
Mean		1630	1638	5.7
Standard Deviation, δ_{n-1}		201	238	2.5
Minimum		1211	1087	3.2
Maximum		2040	1969	-10.2