Acquisition and Processing of Cerebral Blood Flow Data with a Multichannel Analyser and Microcomputer

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SUMMARY

The method of the determination of cerebral blood flow in dogs with radio-isotopes to evaluate ethrane as a new anaesthetic agent is described, as well as the use of a multichannel analyser and the programmes developed for the analysis of data in a conversational mode. Preliminary results of the use of the computer programme are presented.

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Cerebral blood flow is normally increased by the anaesthetic agents used during brain surgery. To evaluate new anaesthetic agents cerebral blood flow has therefore to be investigated to determine if an agent could be found which would not increase blood flow and so reduce bleeding during surgery. This article describes the methodology to determine cerebral blood flow in dogs, using radio-isotopes in order to compare new anaesthetic agents with those in general use. Since during evaluation of new anaesthetic agents the cerebral blood flow is measured before and after administration of the drug, the measurement of absolute cerebral blood flow is not necessary and the use of radio-isotopes is therefore ideal. Different methods of calculation of cerebral blood flow were used, and these methods compared. Comparison of methods is important for extending the use of the gamma camera

to generate additional information by using functional images.

The use of a calculator, which is hardwired to be programmed in Basic to analyse clearance curves of a diffusible radio-isotope, will be described. Results so obtained indicate that the cardiac output and mean brain transit time obtained with a non-diffusible radio-isotope would also be important parameters. For this reason, the programmes to calculate cardiac output and mean transit time are described, although only test studies were done. The ease of programme development and use make this calculator an extremely useful piece of equipment, and this study is given as an example of its usefulness. Similar programmes are also used for the analyses of other clearance curves obtained from lung function, red cell survival and iron plasma clearances.

MATERIALS AND METHODS

Animal Preparation

Adult non-atropinised mongrel dogs—average weight 16 kg—are anaesthetised with pentobarbitone 30 mg/kg intravenously, and are then placed in a supine position on the operating table. An intravenous infusion of a volume-expander or Ringer's lactate by means of a Branule No. 1 needle is set up.

The dog is intubated with a cuffed Magill's oro-endotracheal tube and is connected to a Manley ventilator on a Boyle machine. Nitrous oxide and oxygen ratio 5 : 5 are used for ventilation. To facilitate ventilation and to prevent straining or coughing, the dogs are paralysed with pancuronium bromide 4 - 6 mg intravenously.

The endotracheal tube is connected by a small polythene tube to a Godart capnograph to monitor end-expired CO_2. Minute volume, tidal volume and respiratory rate are adjusted so as to maintain a state of normocapnia. The normocapnic state is frequently checked with arterial blood gas sampling on a Harnoncourts AVL micro blood gas analyser. Acid-base status is similarly monitored.

After skin preparation, the following vessels are cannulated.

(a) A Seldinger catheter is inserted via the femoral artery into the left ventricle of the heart. From this the dp/dt of the heart is recorded on an 8-channel Hewlett Packard (HP) recorder via a HP pressure transducer.

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Heart rate is derived by a rate computer from the dp/dt module and recorded on the 8-channel HP recorder.

(*b*) Through the external jugular vein is cannulated a 16-gauge Intracath positioned in the superior vena cava or right atrium. This is connected to a HP venous pressure transducer and recorder on the HP 8-channel recorder.

(*c*) The internal carotid artery is similarly cannulated with a 16-gauge Intracath, and by means of a 3-way stopcock, arterial pressure is recorded on the HP 8-channel recorder. Through the 3-way stopcock arterial blood sampling can be done and the ^{133}Xe injected for cerebral flow studies.

A cranial skin incision is made, the temporal muscle is deflected, and on achieving haemostasis a burrhole is made through the parietal bone. The lateral ventricle is cannulated with a 16-gauge polythene catheter which is connected to a HP pressure transducer, and the intraventricular pressure is recorded on the 8-channel recorder.

A rectal probe is positioned and the rectal temperature recorded on the 8-channel recorder. The dog's temperature is maintained by means of the electrically heated operation table. The scintillation counter is positioned over the dog's cranium and the first ^{133}Xe flow study performed under this basal state.

The inhalational anaesthetic is now added to the inspired gas mixture, and when the inspired and expired mixtures reach equilibrium, as determined by a Dräger Narkotest M analyser, further cerebral flow studies with ^{133}Xe are performed.

Any changes in other parameters monitored are also recorded on the 8-channel recorder.

Equipment and Methods

Since the effect on the average cerebral blood flow of a new anaesthetic agent was to be evaluated, only one scintillation detector was used over the head of the dog. The detector was connected to an Ortec NIM amplifier and pulse height analyser (Fig. 1). The output of the pulse
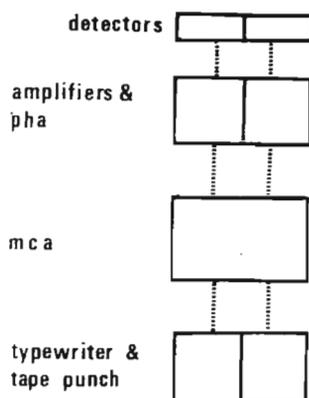
Fig. 1. Block diagram of the electronic equipment used in the investigation: scintillation detectors, amplifiers and pulse height analysers, multichannel analyser, typewriter and tape punch.

height analyser is connected to a Nuclear Data multichannel analyser operated in the multiscaling mode. The 1 024 channels of the analyser were divided into 4 equal groups of 256 channels each. Counts were collected for 5-second intervals up to 800 seconds. A second detector was used for the determination of cardiac output. The multichannel analyser has facility for 4 inputs.

From the data stored in the analyser a print-out was obtained and read into a Hewlett Packard Model 30 computer with a 4k byte memory, hardwired and programmed in Basic. The computer has a thermal printer output, which is fast and silent. It is therefore used for the plotting of graphs as well as for alpha numeric output. Data and programmes can also be stored on a casette recorder. Information of up to 36 characters can be visually displayed.

Principle of Cerebral Blood Flow Determination with Diffusible Radio-isotopes

Cerebral blood flow can be measured by recording the clearance of the radio-isotope ^{133}Xe from brain tissue after an intracarotid injection. Xenon is readily diffusible and the equilibration of blood and brain tissue occurs rapidly after a bolus injection. Since xenon is biologically inert the rate of disappearance from the brain depends only on blood flow. There is no significant recirculation due to the high solubility of xenon in air and 90% of tracer in the blood is excreted by the lungs. The count rate versus time curve recorded by measuring the radioactive xenon in the brain can generally be represented by a double exponential function, where the faster component is taken to represent flow in grey matter and the slower component to represent flow in white matter.

The activity versus time clearance curve can be analysed in several ways:

1. The easiest method of calculating an approximate mean cerebral blood flow F_i is by using the initial slope or half-time T_i of the clearance curve (Fig. 2):

$$F_i = \frac{\lambda \ln 2}{T_i}$$

where λ = tissue blood partition coefficient for brain, and

T_i = initial clearance half-time of xenon in the brain.

2. The mean cerebral blood flow F_a can be calculated from (Fig. 3):

$$F_a = \frac{\lambda H}{A}$$

where λ = tissue blood partition coefficient for brain,

H = maximum height of clearance curve,

A = area under curve.

If the measurement of the xenon clearance in the brain is stopped before complete clearance, the flow can be approximated by:

$$F_a = \frac{\lambda (H - H_{10})}{A_{10}}$$
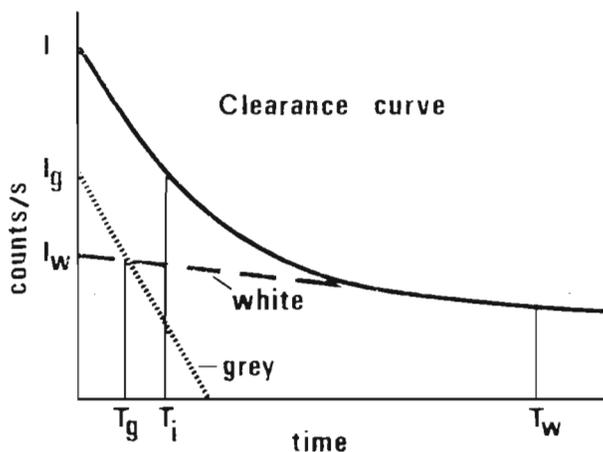
Fig. 2. Clearance curve to illustrate the initial half-time (T,) and the analysis of the double component curve to obtain grey matter (T$_g$) and the white matter (T$_w$) half-times.

where H$_{10}$ = height of clearance curve after 10 min,
      A$_{10}$ = area under clearance curve after 10 min.

This approximation leads to errors of 15% on average. If these errors are not acceptable the value of the area under the curve is obtained by extrapolation, thus:

$$A = A_{10} + \frac{I_{10} \ T_w}{\ln 2}$$

where T$_w$ = clearance half-time of the tail end of the curve, which represents the blood flow in white matter. Such calculation is fast, but requires a longer time for measuring the clearance curves.
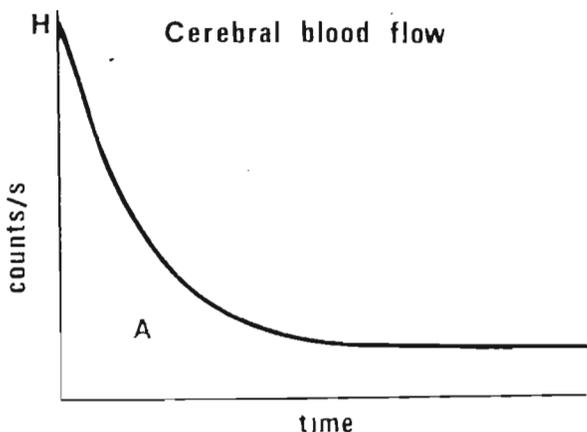
Fig. 3. The parameters of height (H) and area (A) to calculate the mean blood flow (F$_a$).

3. Cerebral blood flow can also be obtained by the assumption that the xenon from the brain is cleared to a double exponential function (Fig. 2).

(a) By analysing the double exponential curve, the blood flow through the white matter F$_w$ and grey matter F$_g$ can be obtained from the clearance half-times of the slow and fast components:

$$F_g = \frac{\lambda_g \ \ln 2}{T_g} \quad \text{and} \quad F_w = \frac{\lambda_w \ \ln 2}{T_w}$$

where λ_g and λ_w = tissue partition coefficient for grey and white matter, and T$_g$ and T$_w$ = clearance half-times for xenon in grey and white matter respectively.

(b) The relative mass of grey and white matter can be calculated from:

$$M_g = \frac{I_g / F_g}{I_g / F_g + I_w / F_w}$$

where M$_g$ = relative mass of grey matter, and I$_g$ and I$_w$ = intercepts of the fast and slow components on the Y axis.

(c) The weighted mean blood flow can be calculated from:

$$F_m = W_g F_g + (1 - W_g) \ F_w.$$

The analysis of the clearance curve according to a double exponential function takes longer, but it renders more information regarding the blood flow in the white and grey matter.

Principle of Cerebral Blood Flow with Non-Diffusible Radio-isotopes

When a radionuclide bolus is injected intravenously, the bolus enters the brain as a stream-out bolus. The count-rate, measured with an external scintillation detector, rises to a peak and falls as the radionuclide leaves the volume being detected. The mean transit time, which is inversely related to brain blood flow, can be calculated. The relationship is non-linear because the pool volume changes with flow rate. For the study ^{111}InCl at pH 1,5 or ^{99m}Tc-labelled human serum albumin was used.

$$\bar{t} = \frac{\sum\limits_{i=1}^{\infty} I_i \ t_i}{\sum\limits_{i=1}^{\infty} I_i}$$

I$_i$ = count rate at the time t$_i$ from start.

The cerebral blood flow can be calculated from:

$$F_t = \frac{1}{t} \quad \text{expressed as ml/ml brain blood volume/minute.}$$

Principle of Cardiac Output

The principle of cardiac output calculation is as illustrated in Fig. 4.

Cardiac output can be measured by recording a bolus of radionuclide through the heart and by determining the count rate over the same region after complete mixing of the bolus with the blood volume has occurred. The cardiac output is then calculated from

$$C = \frac{I_o \times B}{S_1 + S_2}$$

where B = blood volume

I_e = count rate at equilibrium

S_1 = total counts or area under curve up to recirculation.

$$S_2 = \frac{I_r T_r}{\ln 2}$$

i_r = count rate at recirculation

T_r = clearance half time of curve obtained over left heart.
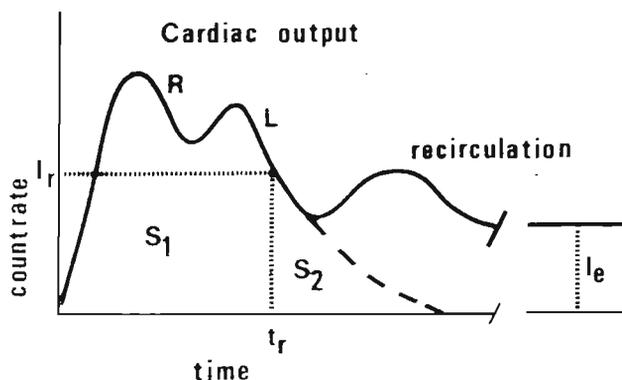
Fig. 4. The areas S_1 and S_2 and the count rate I_r at time t_r before recirculation has occurred, and I_e the equilibrium count rate.

PROCESSING RESULTS

For the analysis of clearance curves obtained with diffusible radioactive xenon, the following procedure was followed:

1. Data were read from the cassette tape, natural logs taken and all the data normalised to a maximum of 100. These curves were then stored on tape for further analysis.

2. The initial part of the curve is fitted to obtain the initial clearance half-time, the slow component half-time, and the data for the calculation of flow according to the height-area method.

The clearance half-times are computed with a developed plotting and curve fitting routine. The log data are first plotted with the thermal printer.

From the graph the tail region over which the graph is linear is selected for calculation of the clearance half-time of white matter. The subroutine calculated the best fit according to a linear least square method, and then printed the observed and calculated data for both linear and log values. The observed and fitted values of the log data are then plotted for visual inspection, and regression constants are printed. The computer also requests whether another curve is to be fitted. This procedure can be repeated until the best fit is obtained. The curve was usually fitted over the range 300 - 600 seconds. At the end of the print-out the sum of the count rate up to where the slow component is fitted is given, as well as the value of the graph at the

end time. These data were used for the calculation of the flow from the height-area method.

3. The initial clearance half-time is obtained by using the same data in the computer memory as above, selecting the initial part of the curve where the clearance curve is linear.

4. The fast component is obtained by stripping the slow component from the curve. This is done by a subroutine which requests the initial count rate and clearance half-time of the slow component and calculates the slow component values to be subtracted from the original linear normalised data, stored on tape. The resultant fast component data are then stored on tape. The curve-fitting routine is called in and the curve fitted over the linear region. This then results in the initial value I_g and clearance half-time T_g of the grey matter fast component.

5. All the clearance half-times and other values are entered for a particular dog and stored on a magnetic tape and filed for later retrieval. From this data either the original clearance half-times or initial count rates can be retrieved, or they can be used to calculate the cerebral flow rates.

The calculator with thermal printer is ideally suited for analysing brain blood flow data. The speed and silence of the thermal printer produces valuable graphs for meaningful stripping of the double exponential curves.

The cerebral blood flow determined with a non-diffusible radio-isotope is calculated with a routine which is much shorter. The data obtained during the transit of the bolus through the brain are entered into the computer and the mean circulation time and cerebral blood flow are calculated.

For the calculation of the cardiac output another subroutine was developed to operate along the same lines as the analysis of the clearance curves. Here the graph of observed data was used to select the clearance curve region before recirculation. An exponential curve is fitted to these data to obtain the clearance half-time, and to calculate the region under this curve where the curve is affected by recirculation. Once this calculation is performed the sample count rates to calculate the blood volume are entered, and the cardiac output is calculated.

RESULTS

Cerebral blood flow was determined in 12 dogs, and 31 measurements were made.

From the average values and standard deviations of the initial 27 determinations it is evident that the initial flow, F_i, does not represent the absolute values of the mean F_a, and weighted mean, F_m. The initial method is biased towards flow in the grey matter which represents only 7% of the total mass (Table I).

TABLE I. CEREBRAL BLOOD FLOW (ml 100 g^{-1} min^{-1})

	F_i	F_a	F_m	F_g	F_w	M_g	M_w
Flow	58	20	21	96	11	7	82
SD	26	9	7	37	3	6	10

The correlation coefficient between the initial and weighted flow was 0,54, and the percentage standard deviation of the difference between the initial flow and the weighted mean flow was 40,2%. The correlation coefficient between the mean and weighted mean flow was 0,81, with a percentage standard deviation from the difference of 22,5% (Fig. 5).
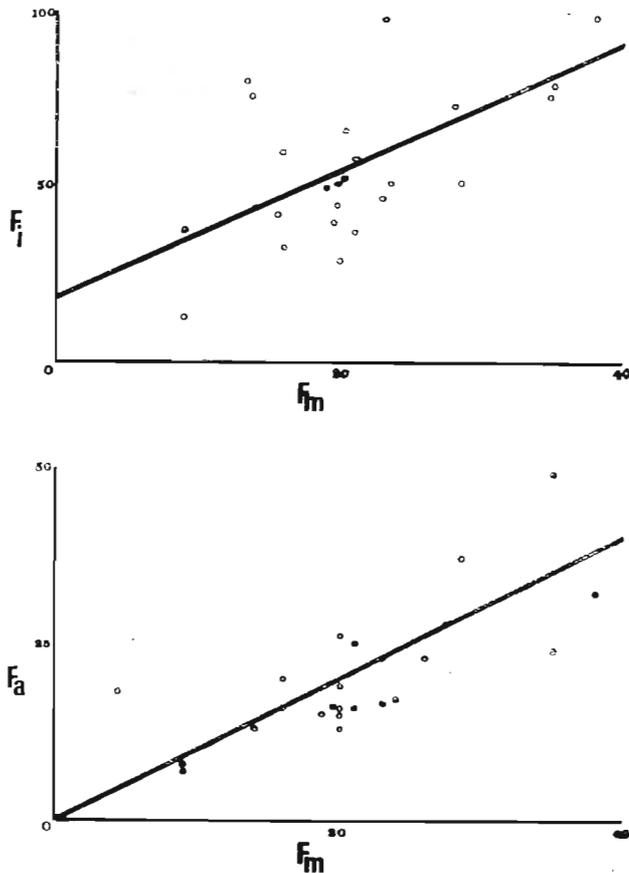
Fig. 5. Scatter diagrams of the initial blood flow F_i and mean blood flow (F_a) against the weighted mean blood flow F_m.

TABLE II. AVERAGE PERCENTAGE INCREASE IN BLOOD FLOW

	F_i	F_m	F_a	F_g	F_w
1% ethrane	48,2	61,6	55,7	73,3	38,6
SD	74,7	62,3	59,2	121,5	39,3
1,5% ethrane ...	−0,3	−0,7	−5,3	−16,0	−7,0
SD	45	28	16,8	18,8	25,5

After administration of the anaesthetic agents, the average blood flow increased with all the methods employed for calculating blood flow (Table II). These results are confirmed by the transducer measurements (Fig. 6). The intracranial and the central venous pressure increase with a decrease in the arterial pressure and dp/dt. This increase in pressure is reversed when the ethrane administration is stopped (Fig. 7).
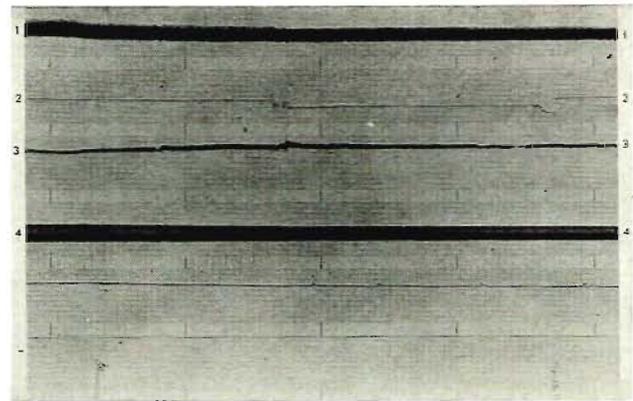
Fig. 6. The change in pressure caused by the administration of 1% ethrane.
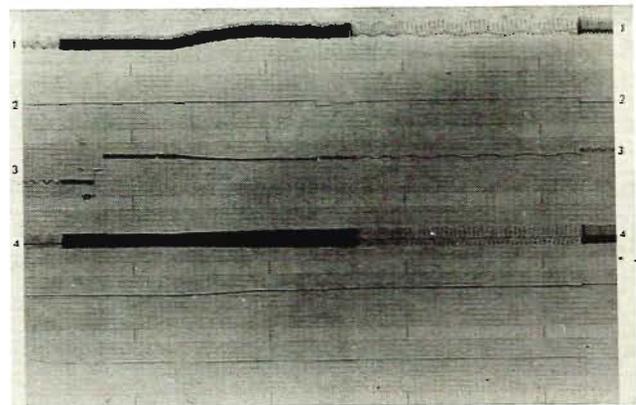
Fig. 7. The reversal of the process in Fig. 6 when the ethrane administration is stopped. The symbols on the figures are as follows: 1 = arterial pressure, 2 = central venous pressure, 3 = intracraniai pressure and 4 = dp/dt.

The results further indicated that the average blood flow in grey matter increased more than in white matter, although there was a great variation. However, when a higher concentration of ethrane is administered there seems to be no effect or a slight decrease in cerebral blood flow.

DISCUSSION

The preliminary results obtained from this study by means of radio-isotopes indicate that 1% ethrane increases cerebral blood flow. These results were confirmed by the measurements obtained in the brain with pressure transducers.

To investigate the mechanism of the change in cerebral blood flow, the measurement of cardiac output is useful. In addition the brain transit time measurement should also be obtained. The transit time represents cerebral blood flow in unit volume flow/second/unit brain blood volume. The cerebral blood flow with diffusible xenon is expressed as the unit volume flow per second per unit organ mass.

By measurement of blood flow with both these methods, an indication of increase in brain blood volume can be obtained.

Another method to be considered is the measurement of the distribution of cardiac output to the brain by the use of the microspheres labelled with different radio-isotopes.

The increasing use of on-line digital computers with the gamma camera has made possible the display of regional blood flow as a functional image for the whole of an organ. Preliminary investigations indicate good correlation between the height-area method and the double exponential method. The height-area method would prob-ably be best suited for display of the functional images. The height-area method requires less calculation, and since a large number of clearance curves are analysed, this is an important advantage. The application of functional curves is not limited to the brain, but could be used to display lung function where similar clearance curves are observed. Functional images of the kidneys for accumu-lation and clearance can be displayed during renograms as well as clearance images during glomerular filtration studies.