A PHYSIOLOGICAL MODEL FOR THE RENAL EXCRETION OF LABELED COMPOUNDS *

Melvin H. Farmelant, Charles E. Sachs†, Sebastian Genna and Belton A. Burrows

University Hospital, Boston Veterans Administration Hospital and Boston University School of Medicine, Boston University Medical Center, Boston, Massachusetts

A number of labeled compounds, selectively excreted by the kidneys, have been used for clinical studies of renal function by external monitoring techniques (*3–6*). There is no general agreement as to the interpretation of these studies. Since the kidneys receive their radioisotope from arterial blood, the following studies were undertaken to relate the shape of the arterial blood curve to the renal area curve in the hope of developing a model for renal handling of these compounds.

METHODS

Female mongrel dogs weighing 15–20 kg were anesthetized with 25 mg/kg of pentobarbital. An infusion of 2.5% mannitol in saline was maintained at about 4–8 ml/min to keep urine flow from each kidney at 3–5 ml/min, thus eliminating renal curve distortion known to be present at low flow rates (*7–9*). The right carotid artery was shunted to the right external jugular vein after heparinization through a coil of polyethylene tubing (Radicoil, Abbott Laboratories, Inc.) which fits a standard NaI(Tl) well counter, and a flow of about 30–50 ml/min was maintained by a constant-infusion pump. Reduction of renal blood flow was accomplished by closing a Poppin-Blalock clamp around the renal artery or by silk ligature of the pedicle. To measure changing urine concentrations of radioisotope, a ureter was cannulated with a double lumen tube and tied below the openings in the tube. One lumen was connected to a water reservoir; the other was connected to another polyethylene coil fitting a second well counter. A constant-infusion withdrawal pump in the system was set to deliver about 20–30 ml/min of water and to withdraw about 10 ml/min more. The bladder was catheterized with an indwelling Foley catheter, and the catheter, filled with urine, was clamped just before injection of the radioisotope for external measurement of bladder accumulation.

In addition to the two well counters, collimated, matched probes with 1½ × 1-in. NaI(Tl) crystals were used for monitoring various body sites. The individual pulses of each detector were recorded on a four-track magnetic tape system (Ames Atomium Corp., Billerica, Mass.), which has been previously described (*10,11*), and subsequent integration of counts accumulated in various fixed intervals was accomplished by reading the tapes through a multiscaler. Subtractions of room or tissue background counting rates and summations (integration) of successive equal intervals of arterial blood counts were performed electronically.

Iodine-131-hippuran was injected intravenously as a single bolus in about 20–30-μCi doses. When repetitive studies in a single dog were performed, about an hour elapsed between injections to allow excretion of the bulk of the preceding dose. PAH clearances were performed by standard constant-infusion techniques.

Tissue background was subtracted from renal area curves (*11,12*) to yield net kidney curves. Intercomparison of kidney, bladder, urine counts and arterial blood counts required normalization because of different sensitivities of the well and probe detectors and different geometrical arrangements of the probes. Normalization was accomplished by multiplication of the appropriate curve by a factor selected to provide the best visual fit of semilogarithmic plots of the curves being compared during the first 2–3 min.

RESULTS

Shape of arterial concentration curves. Curves of arterial blood concentration of ^{131}I-hippuran were obtained in intact dogs and following unilateral and bilateral nephrectomies. Curves of a representative dog are presented in Fig. 1. Arterial concentration diminished to $\frac{1}{20}$ of its initial value in 10 min. Curves of an integral of arterial blood concentration, obtained by subtotaling successive 20-sec intervals, are also shown for the intact and uninephrectomized states.

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For reprints contact: Melvin H. Farmelant, St. Vincent Hospital, 25 Winthrop St., Worcester, Mass. 01610.

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† Present address: Hôpital Neckar, Paris, France.

TABLE 1. RATIOS OF NORMALIZED BLADDER AND INTEGRAL OF ARTERIAL BLOOD RADIOACTIVITIES (BLADDER/BLOOD)

Dog No.	Exp.	Ratio at 2 min	4 min	8 min	12 min
9	2-I*	1.00	1.12	1.09	1.01
10	1-I	1.00	1.08	1.08	1.03
	2-U†	1.00	1.05	1.05	1.0
11	1-I	1.00	1.07	1.09	1.08
	2-U	1.00	1.07	1.02	0.97
	3-U	1.00	1.05	1.04	0.99
12	1-I	1.00	0.93	0.90	0.85
13	1-I	1.00	0.99	0.89	0.82
16	1-I	1.00	1.01	0.97	0.93

* Intact renal arteries.
† Unilateral nephrectomy or arterial clamping.

Comparison of bladder and integral of arterial concentration curves. Simultaneous curves of arterial blood concentration of ^{131}I-hippuran and bladder accumulation were obtained in nine studies including three performed with a renal artery clamped. Figure 2 shows a semilogarithmic plot of the bladder curve and the subtotaled arterial concentration curve obtained by cumulatively adding counts from successive 20-sec intervals. The bladder curve has been multiplied throughout by a constant factor to normalize the curves as described in Methods. The bladder curve has been replotted transposed in time by an interval (labeled "passage time") corresponding to that time between the first appearance of radioactivity in the arterial blood and the first appearance of radioactivity in the bladder. The identity of the shape of the two curves is evident. Table 1 gives the ratio of the normalized curve of an integral of arterial concentration to the bladder curve, displaced in time as in Fig. 2. Ratios were calculated at 2, 4, 8 and 12 min after the first appearance of radioactivity. The constancy of the ratios indicates the identity of the shape of the curves during a 4–5-fold rise in bladder radioactivity.

Comparison of arterial blood concentration with urine concentration. In eight experiments in four dogs the curve of arterial radioisotope concentration was compared to that of radioisotope concentration in the urine issuing from one kidney. A constant flush with water was maintained as described in Methods to minimize intrapelvic mixing of radioisotope. The urine concentration curve was displaced in time so that the first appearance of radioactivity coincided with the time of injection to eliminate the effect of renal passage time. Figure 3 shows the results of each experiment. The urinary concentration shows a period of rise about 1.5 min and a decline initially

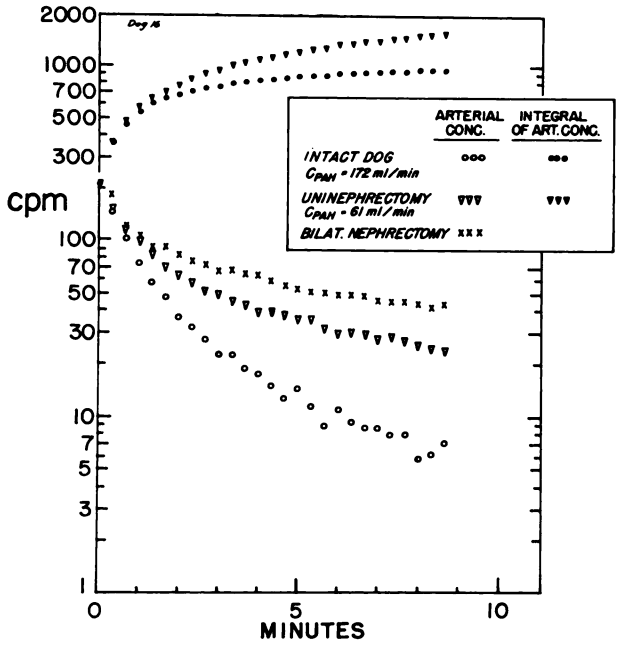

FIG. 1. Arterial blood concentration curves in single dog made with ^{131}I-hippuran. Curves are for intact animal and following single and bilateral nephrectomy.

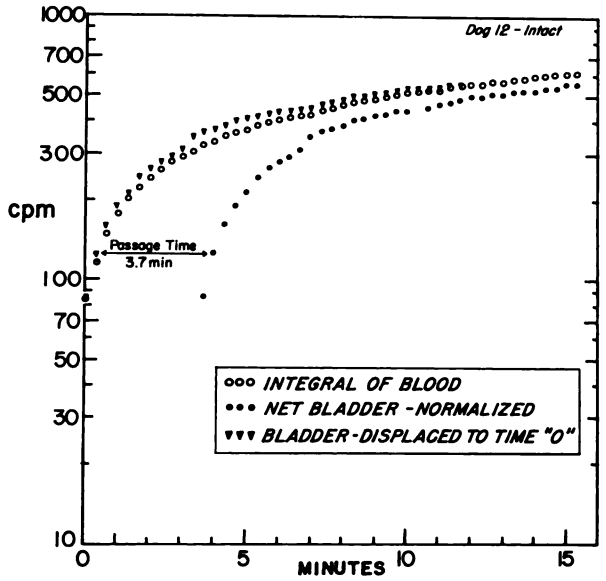

FIG. 2. Comparison of integral of blood concentration with bladder curve with ^{131}I-hippuran in dog. Bladder curve has been normalized to initial equal radioactivity and displaced in time to facilitate comparison.

somewhat slower and terminally more rapid than that of the blood concentration. The decrease in arterial concentration in the interval of comparison is about tenfold. Methylene blue dye dilution curves obtained from the pump system alone are shown.

The relationship of the kidney curve to arterial blood concentration. Curves of renal radioactivity normally show increasing uptake for some 2–5 min and then progressive decrease. Curves were obtained

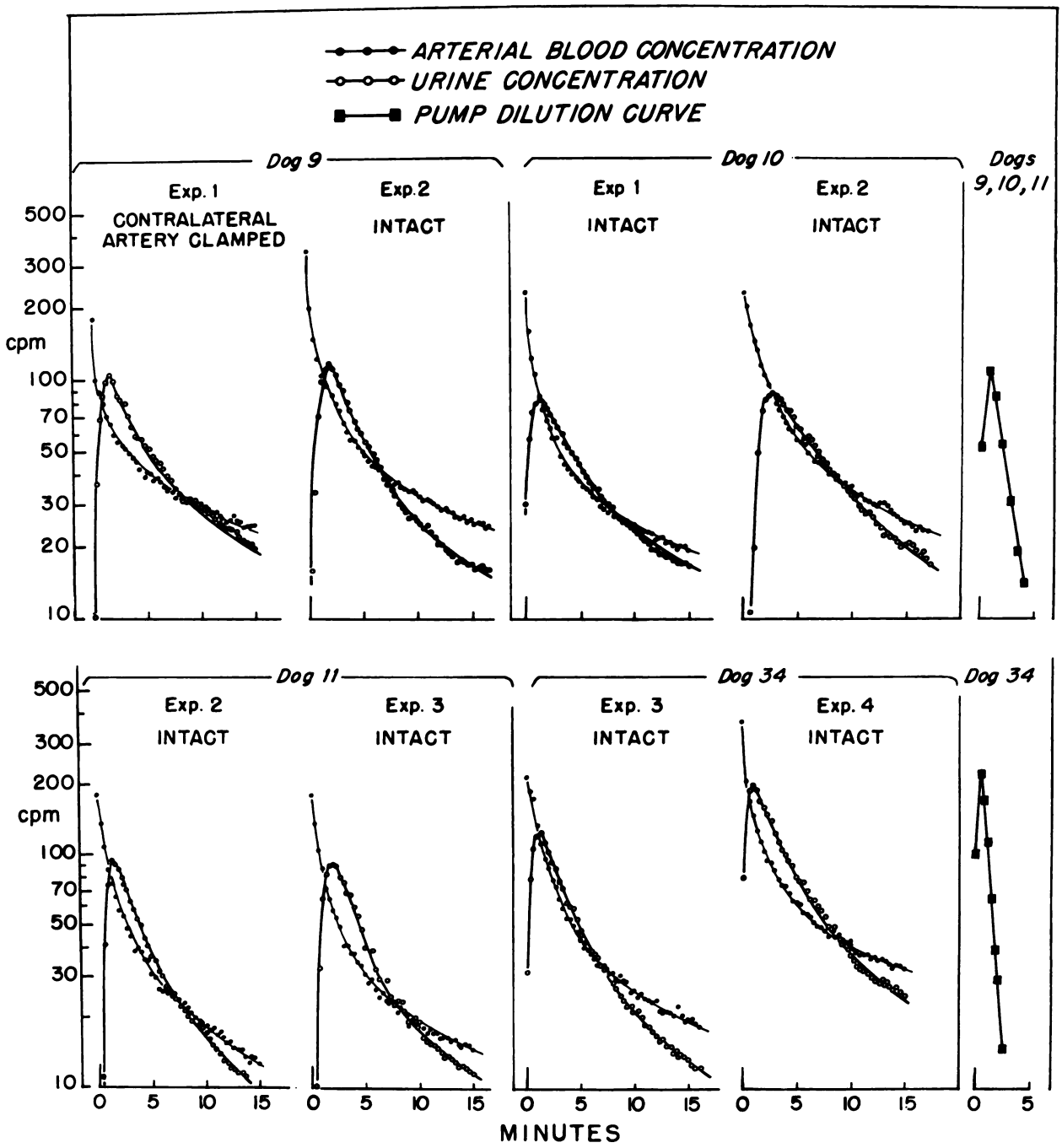

FIG. 3. Comparison of arterial blood and urine concentration curves with ^{131}I-hippuran in dog. Urine curves have been displaced in time to facilitate comparison.

from five dogs in five studies with intact renal circulation and two studies with occlusion of the renal artery contralateral to the observed kidney. The net kidney curves were compared with the integral of arterial blood concentration. The curve of the integral of blood concentration was normalized to the rising portion of the net kidney curve and plotted for the entire period of observation. In all studies the two curves superimposed during the period of renal rise.

During the period of declining renal radioactivity the renal curve was subtracted from the continuously rising curve of the integral of arterial radioisotope concentration, and the differences were plotted and replotted displaced by an interval corresponding to the passage time. It was found in each study that the curve of the differences was identical to that of the integral of the arterial blood concentration (Fig. 4, Table 2).

Converse studies were performed. In four studies in three dogs the curve of bladder accumulation was normalized to the curve of the integral of arterial concentration and was subtracted from this without

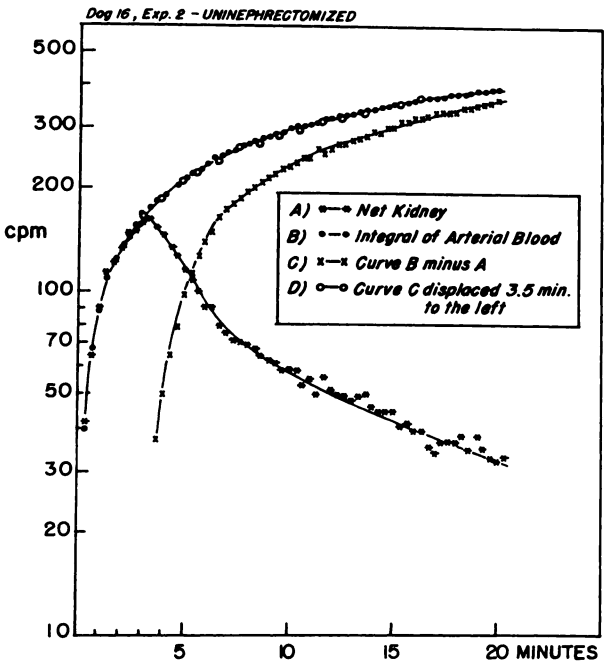

FIG. 4. Comparison of integral of blood concentration and difference between kidney and integral of blood concentration with ^{131}I-hippuran in dog. Curve of differences has been displaced in time to facilitate comparison.

transposition in time. The net kidney curve was also normalized so that the first 2 min corresponded to the integral of arterial concentration. The difference between the bladder and integrated arterial concentration curves was plotted and corresponded closely to the renal curve in each case (Table 3). Figure 5 illustrates this in a dog with intact kidneys and following uninephrectomy. The flattening of the net kidney curve associated with reduction of total renal blood flow is evident. PAH clearances obtained during each study are shown.

Comparison of bladder and renal curves in man. A group of 10 tests was selected for analysis from a large series of studies in patients and subjects in whom curves of the two kidneys, the bladder and upper chest were available. The two kidneys had similar times of peak (passage times) in the selected group because differences in passage time are known to distort the bladder curve (*13*). The bladder curve was taken to represent the curve of the integral of arterial blood concentration as was shown in the dog and was displaced backward in time to eliminate the effect of passage time. The net kidney curve was normalized to this, and the difference between the displaced bladder curve and the descending portion of the net kidney curve was obtained. This curve, also displaced to time zero, corresponded to the original bladder curve as shown by ratios of points on the two curves taken at 2, 4, 8, 12, 16 and 20 min (Table 4, Fig. 6).

TABLE 2. RATIO OF INTEGRAL BLOOD RADIOACTIVITY AND DIFFERENCE BETWEEN INTEGRAL OF BLOOD RADIOACTIVITY AND KIDNEY RADIOACTIVITY (BLOOD/DIFFERENCE)

Dog No.	Exp.	Ratio at 1 min	2 min	4 min	6 min	8 min	12 min
8	1-I*	1.00	1.00	0.98	0.99	0.99	1.00
12	1-I	0.87	0.96	1.00	1.01	1.01	1.01
13	1-I	1.03	1.02	1.01	1.01	1.00	1.00
	2-U†	1.16	1.00	1.03	1.03	1.03	1.03
14	1-I	1.00	1.00	0.95	0.94	0.92	0.87
16	1-I	0.80	0.875	0.93	0.94	0.95	0.95
	2-U	0.93	1.00	1.00	0.98	0.99	0.99

* Intact renal arteries.
† Unilateral nephrectomy or arterial clamping.

TABLE 3. RATIO OF KIDNEY RADIOACTIVITY AND DIFFERENCE BETWEEN INTEGRAL OF BLOOD RADIOACTIVITY AND NORMALIZED BLADDER RADIOACTIVITY (DIFFERENCE/KIDNEY)

Dog No.	Exp.	Ratio at P*+2 min	P+4 min	P+6 min	P+8 min	P+12 min
12	1-I†	0.79	0.77	0.80	0.865	0.95
14	1-I	1.00	0.86	1.00	0.97	
16	1-I	0.85	0.84	0.94	1.05	
	2-U‡	0.97	0.82	0.80	0.87	

* Time of peak activity.
† Intact renal arteries.
‡ Clamped contralateral renal artery.

TABLE 4. RATIO OF DIFFERENCE BETWEEN NORMALIZED BLADDER AND KIDNEY CURVES AND NORMALIZED BLADDER CURVE

Subject	Ratio at 2 min	4 min	8 min	12 min	16 min	20 min
EP	1.00	1.02	1.02	1.01	1.01	1.00
JP	1.00	0.97	1.00	0.98	0.98	0.99
JO	1.00	0.99	0.97	0.97	0.96	0.96
BM	1.06	1.00	1.01	0.98	0.97	0.97
ER	1.00	1.03	0.98	0.96	0.97	
RL	1.00	0.99	0.97	0.99	0.98	
DS	1.00	1.00	0.95	0.99	1.00	
EW	1.00	1.00	1.00	0.99	0.98	0.97
HM	1.00	1.00	0.98	0.97	1.00	0.99
CA	1.00	1.00	1.04	1.02	0.99	1.00

DISCUSSION

These studies demonstrate that renal accumulation of ^{131}I-hippuran can be expressed by a constant times the integral of the arterial blood concentration ($k\int Bdt$). This would result if a kidney continuously

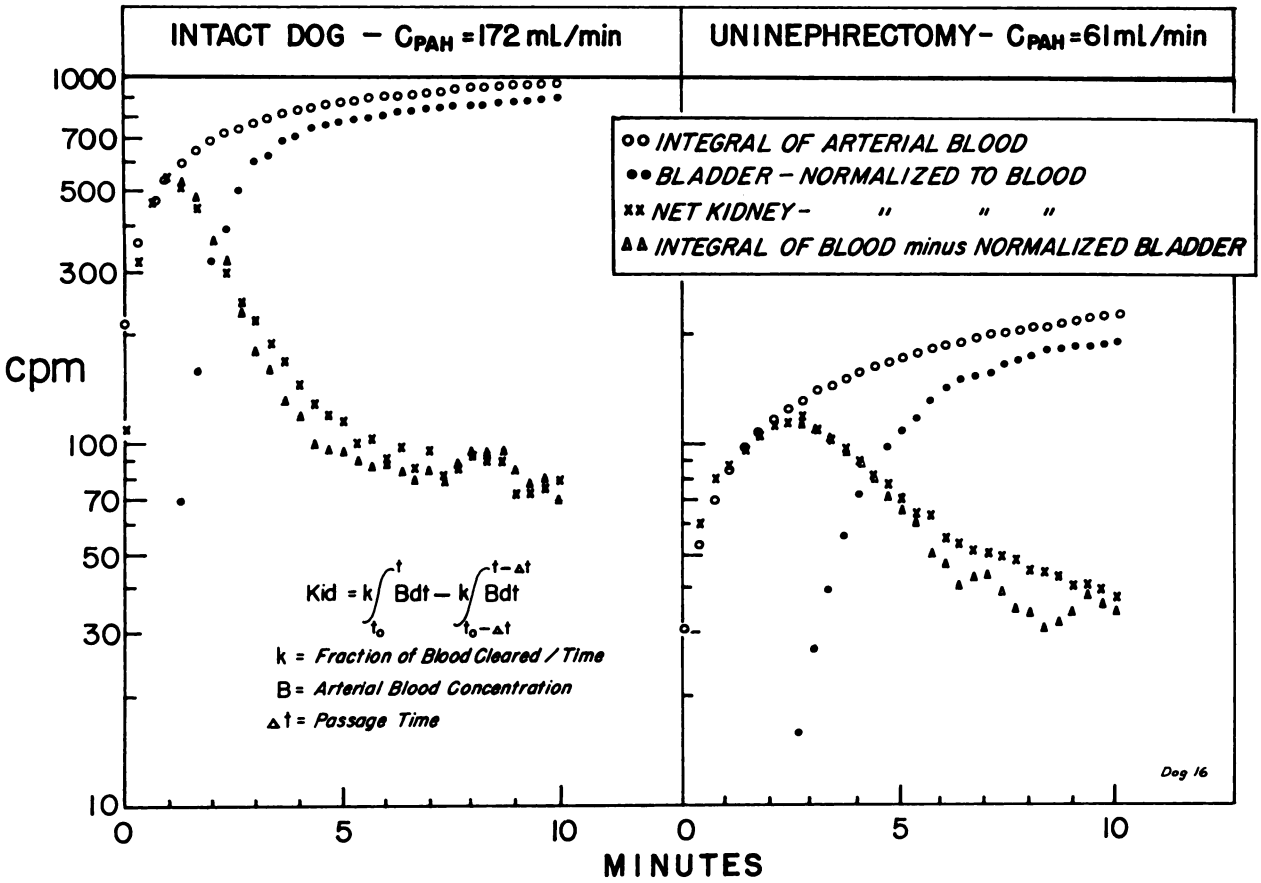

FIG. 5. Comparison of net kidney curve and differences between integral of blood concentration and bladder radioactivity with ^{131}I-hippuran in dog. Time relationships have not been altered. After uninephrectomy, curves are flatter, but correspondence of curves of difference and kidney is preserved.

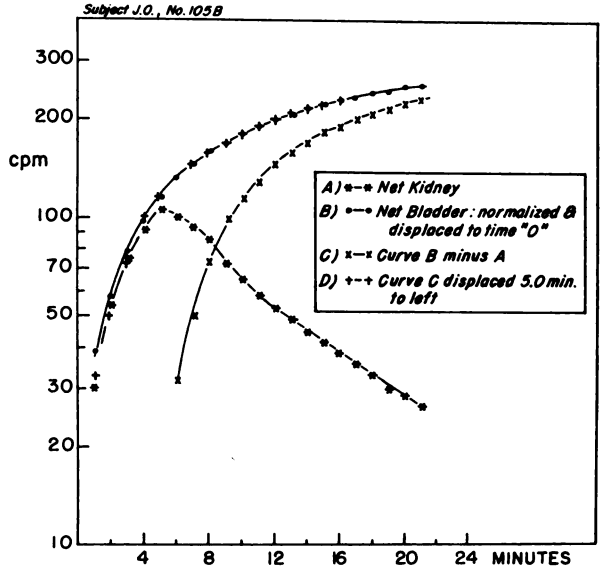

FIG. 6. Comparison of bladder curve and differences between bladder and net kidney curves with ^{131}I-hippuran in man. Curve of differences has been displaced in time to allow comparison.

extracts a fixed proportion of the amount of radioisotope present in a constant mixing pool of blood (the effective blood volume). The value of the constant would reflect the fraction of the blood volume being cleared by the kidney and, with external measurements of renal radioactive material, would also include attenuation, geometrical and sensitivity factors inherent in such measurements. Accumulation of radioactivity in the bladder is an integration of the amounts of radioactive material issuing from the kidneys. Our studies have shown that this integral curve is indeed proportional to the integral of arterial blood concentration. The implication of this observation and another previously reported (*11*) is that in hydrated subjects the radioisotope taken up by the kidney passes through the renal tubule and renal pelvis with little mixing.

If this is so, the time-concentration curves of ureteral urine and arterial blood radioactivity should be identical but displaced in time. Our data show fair correspondence over a tenfold change in concentration but a definite difference in that the urine curve has a short period of rise and then initially falls more gradually and finally more rapidly than the blood curve. A part of the divergence reflects mixing within the pumping system which was demonstrated by obtaining dilution curves of a bolus of methylene blue (Fig. 3). A more significant factor

may be transient storage of radioisotope in renal tubular cells. Chinard (*14*) has shown a 70-sec delay in the mean of PAH excretion when compared with glomerular substances when both are simultaneously injected in a renal artery. He concludes that the fraction of PAH which is filtered comes out in the urine as do other filtered materials, but the fraction that is secreted by the tubular cells is transiently delayed by about 1½ min. If about 80% of the ^{131}I-hippuran is delayed in its passage through the kidney by 1–1½ min, the discrepancies in our arterial and urine concentration curves may be explained.

With this model as a basis, certain portions of the curve can be analyzed to provide quantitative information which may be related to overall renal function. Using the descending slope of a nonobstructed kidney with an adequate urine flow as an index, the response to surgery in patients with unilateral renal arterial stenosis could be predicted (*18*). The assumption of a single passage time through all the nephrons is an oversimplification that probably does not markedly influence the relationships between arterial, renal and bladder curves expressed by the model. Approximate homogeneity of behavior in nephrons of disparate size has been suggested by other studies in dogs (*19*). Observations on the effect of altered renal hemodynamics and changes in solute excretion on the spread of transit times through a dog kidney suggest that deviations from this model are small (*20*).

Subtraction of the net kidney curve from the curve of the integral of blood concentration after renal emptying begins yielded a curve similar to the original integral curve transposed in time. Subtraction of the bladder curve (which is the integral of the losses from kidneys) from the curves of the integral of blood concentration (which represents the integral of uptake) yielded values similar to the original net kidney curve. In man, substituting the bladder curve for that of the integral of arterial blood concentration and subtracting the net kidney curve from this yields the original bladder curve again. These data indicate that after an interval (the passage time) radioactivity is released from the kidney in the *same* time-concentration order in which it is taken up from the blood. Uptake has been expressed before as $k\int Bdt$. We can now state that in the hydrated subject losses from a normal kidney follow the same expression. The equation that would fit the entire kidney curve, accounting for passage time would be

$$Kid = k\int_{t_o}^{t} Bdt - k\int_{t_o-\Delta t}^{t-\Delta t} Bdt$$

in which Kid is net counting rate of the kidney at time t; k combines the fraction of the effective blood volume being cleared by the kidneys per unit time with probe sensitivity factors; B is the arterial blood concentration; t is the time of observation after initial appearance of radioactivity t_o; and Δt is the passage time through the kidney. This equation assumes that little mixing of radioisotope occurs in passage through the kidney which we have shown in the nonobstructed kidney with adequate urine flow.

This equation ignores a difference in delay time between passage of the secreted and filtered portions of ^{131}I-hippuran and assumes a single passage time through all the nephrons of a kidney. Despite these simplifications, the closeness of fit of the data makes this model useful in describing the kidney curve.

Figure 7 is a simplified model of the events described. The kidney is shown as an irregular tubular structure. As uptake proceeds, the concentration of ^{131}I-hippuran in the proximal portion diminishes continuously as a consequence of falling arterial concentration. The ^{131}I-hippuran is carried down the kidney without mixing. Since the entire kidney is viewed by the detector, the renal area curve rises. Once emptying of radioisotope begins, urine with the highest concentration of radioisotope is delivered to the bladder to be replaced proximally by smaller amounts, and the kidney curve falls.

One consequence of this analysis is noteworthy. Not only is the shape of the rising portion of the renal curve dependent on the rate of change in arterial blood concentration *but the falling portion is also*. As is seen in Figs. 1 and 5, when total renal blood flow is reduced by uninephrectomy, arterial blood concentration falls more slowly than in the

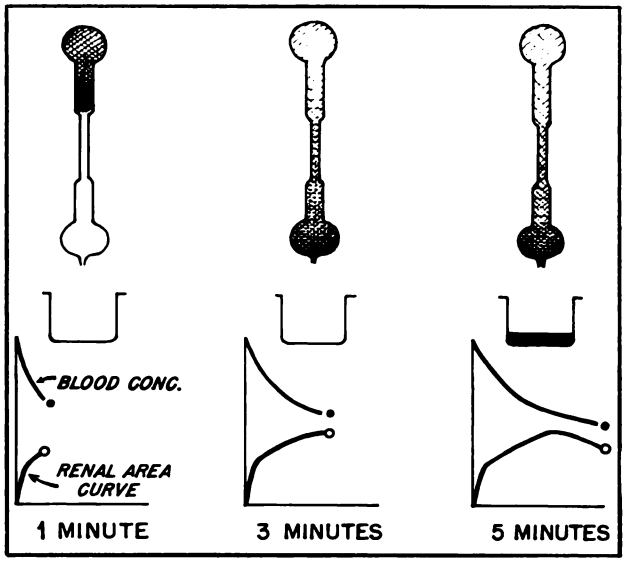

FIG. 7. Model of ^{131}I-hippuran renal curves.

intact animal, and the falling portion of the kidney curve is also considerably less steep.

Because an individual's two kidneys sample the same arterial concentration, differences between the kidney curves, which occur in many patients, must be explained. The value of k may differ either because of differences of counting sensitivity or of renal clearance. Attempts to reduce the variability of renal counting rate caused by geometric factors have been made by increasing detector-to-skin distance and altering collimation of the probes (*9,15,16*). However, as yet no arrangement of detectors has been shown whereby the ratio of the magnitude of uptake by the two kidneys correlates with relative renal blood flow, and differences in counting rate cannot be ascribed with certainty to differences in function of the kidneys.

The only other variable in the equation that could differ between the two kidneys is the time of passage (Δt). However, convincing data have been presented which show that at *low* urine flow rates the descending portion of the kidney curve is considerably flattened (*7–9*). With low urine flow or with mechanical interference with emptying from the renal pelvis, the curve of decreasing renal radioactivity no longer follows the model represented by the equation, presumably because of the presence of significant intrarenal mixing of radioisotope which introduces an additional exponential component. Both prolonged passage time (Δt) and unilateral low urine flow despite hydration may be consequences of unilateral renal arterial stenosis. The success in detecting this disorder obtained by methods that evaluate the ^{131}I-hippuran curves after initial release of radioisotope from the kidney is understandable (*17*).

SUMMARY

Following ^{131}I-hippuran injection, renal curves, urine concentration and bladder radioactivity curves were related to arterial blood concentration curves. Renal uptake, until emptying begins, and bladder accumulation closely approximate the integral of arterial blood concentration. The difference between the integral of blood concentration (renal uptake) and bladder (integrated renal losses) approximates the net kidney curve after loss from the kidney begins. Likewise, the difference between renal radioactivity and the integral of blood concentration yields the original integral curve of arterial blood concentration. The curve from a nonobstructed kidney with adequate urine flow can be expressed as the integral of blood concentration minus the same integral displaced in time (time of passage of radioisotope through the kidney). We can conclude that with adequate urine flow rates the rate of fall as well as the rate of rise of the kidney curve reflects the rate of fall in arterial blood concentration; moreover, the passage of radioisotope through renal tubule and pelvis involves little mixing. Differences between the kidneys in retention of radioisotope reflect both differences in passage time and often marked unilateral reduction of urine flow rate.

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