PHYSIOLOGICAL DETERMINANTS OF RENAL TUBULAR PASSAGE TIMES

Melvin H. Farmelant, Karel Bakos* and Belton A. Burrows

University Hospital, Boston University Medical Center and
Boston Veterans Administration Hospital, Boston, Massachusetts

The most constant renographic finding in renal arterial stenosis is prolonged retention of the radioactive tracer in the affected kidney (*1,2*). This prolonged retention is considerably reduced or eliminated by osmotic loads, a phenomenon that has been diagnostically useful in differentiating arterial from parenchymal unilateral renal disease (*1*). The mechanisms responsible for these effects have not been clearly elucidated. In dogs, previous studies of the relationships of renogram curves to the arterial blood concentration curve led to the development of a model of these studies (*3,4*). The data indicated that renogram curves in both ascending and descending portions could be described with a high degree of accuracy as a permutation of the blood concentration curve which was considered the "input" into the kidney. The model implied that transit of radioisotope through the kidney occurred through a single tube with a single velocity and without significant spreading or mixing of temporal events in the tube. Small deviations of the data from predictions of the model exist near the peak of the curve. Presumptively, these differences represent an intrarenal distortion of the input function (arterial blood concentration curve) produced during transit.

Therefore we performed studies in dogs to investigate the significance of the distortion imposed on a bolus of radiohippuran introduced directly into the renal artery. Studies were performed at varied urine flows, with graded diminutions of renal blood flow and over a wide range of osmotic loads to clarify the physiological consequences and renographic effect of these stimuli.

METHODS

Eleven mongrel dogs of both sexes, weighing 10–20 kg, were the subjects for these studies. They were anesthetized with 50 mg/kg sodium pentobarbital intravenously, and supplemental doses were given as needed to maintain anesthesia. Bilateral flank incisions were made, and the ureters were catheterized with polyethylene tubing. One femoral artery was cannulated with plastic tubing which was connected to a mercury manometer; the vena cava was cannulated through the femoral vein for collection of blood samples. A clamp with adjustable jaws (Poppin-Blalock) was placed around the aorta above the renal arteries, and a bent 25-gage scalp vein needle connected to an infusion set was introduced into the renal artery. Only dogs with kidneys supplied by a single renal artery were studied.

A constant infusion peristaltic pump delivered about 3 mg/min PAH and 9 mg/min creatinine intravenously following a priming dose. One hour was allowed for equilibration. An additional intravenous infusion of 10% mannitol or 5% dextrose or normal saline was administered during the studies to alter the rate of water and solute excretion.

Each experimental observation consisted of at least two clearance periods and the results of monitoring the kidneys following an injection of 0.2–0.4 ml containing about 20 μCi ^{131}I-labeled hippuran as a bolus into the renal artery. Two matched flat-field collimated probes with 1½ × 1-in. NaI(Tl) crystal detectors were used to monitor the kidney regions. Pulses from the detectors were recorded on a magnetic-tape system and subsequently read through a magnetic-core memory pulse-height analyzer used in a multiscaler mode to yield counts accumulated per selected time interval. Counts obtained from the uninjected control kidney were subtracted electronically by the analyzer from the counts of the injected kidney at corresponding times to remove the effect of background and recirculation of the nonextracted radioisotope.

Counts were integrated at 16-sec intervals and plotted with Cartesian coordinates. For about 1.0–1.5 min following the appearance of radioactivity in the renal area, the counting rate remained fairly constant and then decreased rapidly for several minutes and finally diminished quite slowly (Fig. 1). As-

---

Received Jan. 9, 1969; original accepted April 25, 1969.

For reprints contact: Melvin H. Farmelant, Saint Vincent Hospital, 25 Winthrop St., Worcester, Mass. 01604.

* Present address: Charles University, Prague, Czechoslovakia.

suming that passage through the kidney approximated a distribution function, the curve of diminishing activity was treated as the integral of such a function. Differentiation of the curve invariably yielded a skewed frequency distribution. The time of peak of the differentiated curve representing the modal passage time (MPT) was noted. An index of the spread of the passage times was determined graphically. From the total height of the curve (the original counting rate) 5% was subtracted from both the top and bottom. The time at the half-maximum value of the curve was noted. The interval between the time the counting rate declined from 95% of its initial value to one-half maximum value divided by two was called σ_1. Similar measurement of the tail of the curve from time at half-maximum value to the time the remaining radioactivity had declined to 5% of the initial counting rate divided by two was called σ_2 (Fig. 1). A measure of overall spread (σ) was calculated as

$$\sigma = \left(\frac{\sigma_1^2 + \sigma_2^2}{2}\right)^{1/2}.$$

This method of deriving sigmas very closely approximates numerical derivations of standard deviation that assume true Gaussian distributions. The degree of skewness of the curves was calculated by

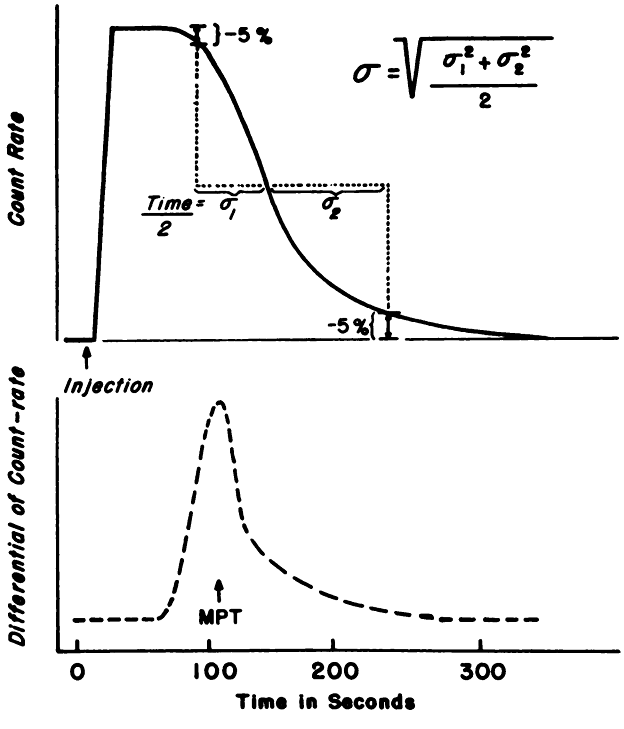

FIG. 1. Top: Times at which 95%, 50% and 5% of maximum counting rate remain are noted. Interval between 95% and 50% remaining divided by 2 equals σ_1. Similar measurement between 50% and 5% provides σ_2. Formula for overall sigma is also shown. Bottom: Modal passage time is determined by differentiating above curve.

$$\text{skew} = \left(\frac{\sigma_1}{\sigma_2}\right)^3.$$

In the studies to be presented, σ_1, σ_2, σ, the interval between injection and the disappearance of 50% of the radioactivity, and the interval between injection and total disappearance of radioactivity were separately correlated with the various physiological parameters. Since they all varied in the same direction and to approximately the same degree, only the values for σ and the modal passage time (MPT) are presented here. In all cases σ_2 was greater than σ_1. The skewness of the curves was statistically independent of the measured parameters and is not presented. The values for UFR, GFR and osmotic excretion are for the injected kidney only.

Semilogarithmic plots yielded no evidence of an exponential function except at low urine flow rate (0.1 ml/min or less) which produced data that fitted an exponential poorly.

Two studies were performed to evaluate the role of the renal pelvis in affecting these curves. In one, a double-lumen catheter was inserted per ureter into the pelvis and the pelvis "washed" with a stream of water controlled by a peristaltic pump. Radiohippuran was injected into the renal artery. In the second experiment a series of injections of radiohippuran was made into the renal pelvis at differing intrinsic urine flow rates.

To compare various parameters, regression lines were fitted by least mean squares and correlation coefficients obtained by standard statistical methods.

PAH and creatinine were determined by standard methods and osmolality determined by freezing-point depression.

Because the dogs studied differed considerably in size, the physiological parameters were divided by the body weight as a normalizing procedure.

RESULTS

Effect of pelvic retention. Injection of a bolus of ^{131}I-hippuran into the renal pelvis at a variety of urine flow rates showed almost complete disappearance in 16–32 sec at urine flow rates above 0.1 ml/min. During pelvic flushing with a stream of water through a double-lumen tube, curves of disappearance of radioactivity from the whole kidney, obtained after bolus injection into the renal artery, were similar to those obtained without flushing. It was concluded that at urine flow rates above 0.1 ml/min from a kidney, the renal pelvis contributed very little to the time of passage and that the phenomena described below reflect intratubular events.

Effect of urine flow rate. Urine flow rates between 0.06 and 1.2 ml/min-kg were achieved by varying the osmotic and water loads to initially

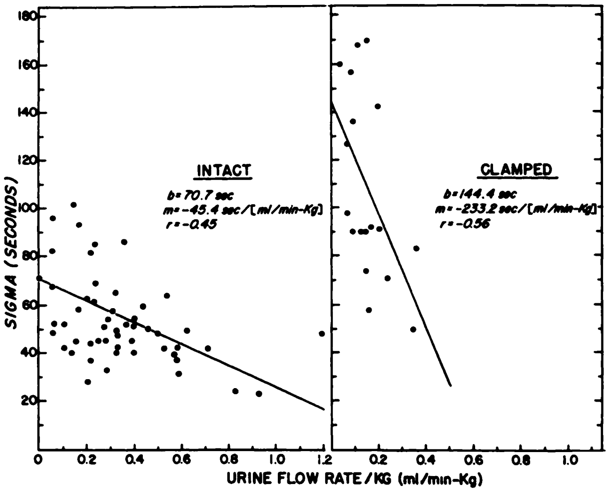

FIG. 2. Sigma as function of urine flow rate is shown in intact and clamped animals.

hydropenic dogs. Table 1 and Fig. 2 show the correlation of σ with urine flow rate/kg (UFR/kg) in dogs with intact and partially occluded renal vasculature. In intact dogs, sigma is inversely correlated with urine flow rate, but the slope of the relationship is shallow and the correlation coefficient is not high (r = —0.45). Kidneys with clamped renal arteries showed steeper relationship between σ and UFR, indicating that changes in UFR were associated with greater changes in σ than in dogs with intact renal circulation. Again the correlation was not high (r = —0.56). The modal passage time (MPT) was very poorly inversely correlated with UFR in intact animals but much more steeply and better correlated in dogs with clamped vessel (Table 1 and Fig. 3). The differences between the two groups was statistically significant ($p < 0.05$) for both σ and MPT.

Effect of altering glomerular filtration rate. Glomerular filtration rates were varied over about a 10-fold range. The slopes of the relationship between σ and GFR in intact and clamped artery animals were not significantly different (Table 1) and so the combined data were fitted with a single slope (Fig. 4). The correlation coefficient is —0.84. The modal passage time also was similar in the two groups (Table 1) and well correlated with filtration rate.

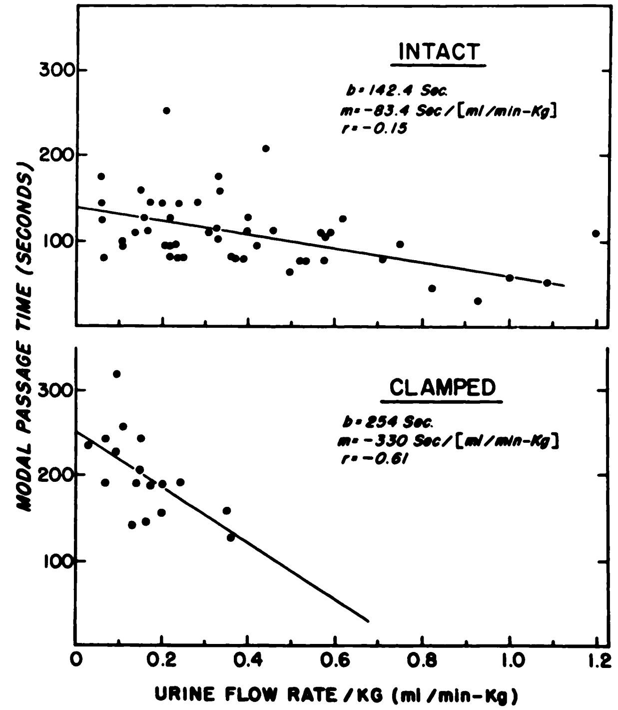

FIG. 3. Modal passage time as function of urine flow rate is shown in intact and clamped animals.

Effect of osmolar excretion. In dogs with intact renal circulation, osmolar excretions, varying over a six-fold range, were associated with slight variation in σ (Fig. 5) or modal passage time (Table 1), and the correlations were poor (r = —0.24 and —0.25, respectively).

In dogs with clamped renal arteries, much steeper slopes of the relationship between σ or MPT and the

TABLE 1. RELATIONSHIP OF PHYSIOLOGICAL PARAMETERS WITH MODAL PASSAGE TIME AND SIGMA

	Modal Passage Time					Sigma				
	Intact		Clamped			Intact		Clamped		
	y intercept (sec)	Slope	y intercept (sec)	Slope	p of diff.	y intercept (sec)	Slope	y intercept (sec)	Slope	p of diff.
UFR/kg	142.4	—83.4	254	—330	<0.05	70.8	—45.5	146.6	—237.3	<0.05
(ml/min-kg)	r = —0.15		r = —0.61			r = —0.56		r = —0.45		
GFR/kg	210.3	—39.9	379.8	—124.6	>0.05	111.3	—23.5	172.5	—52.8	>0.05
(ml/min-kg)	r = —0.67		r = —0.71			r = —0.77		r = —0.88		
Osmol excr.	136.1	—170.6	354.2	—1631	<0.05	60.5	—68.7	173.5	—788	<0.01
((Osmol/min-kg)	r = —0.25		r = —0.51			r = —0.24		r = —0.76		

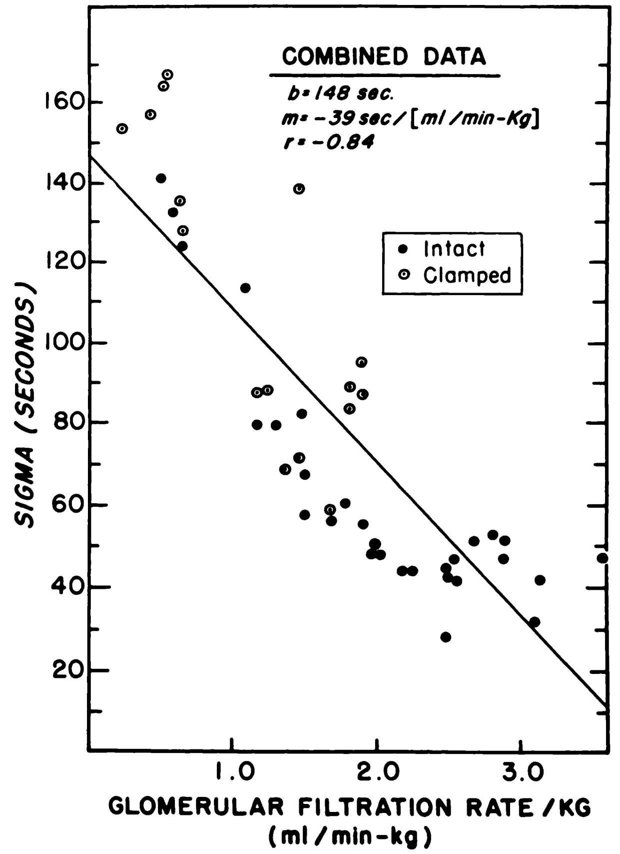

FIG. 4. Sigma is shown as function of glomerular filtration rate. Data for intact and clamped animals are combined because no statistically significant differences exist between groups.

osmotic excretion were obtained (Table 1 and Fig. 5) indicating that in this situation, for similar changes in osmotic excretion, much greater changes in σ and MPT resulted compared with the intact animals. The differences between the slopes of these two experimental conditions is statistically significant ($p < 0.01$ and < 0.05, respectively).

DISCUSSION

Introduction of a bolus of radioactive hippuran into the renal artery is presumed in these studies to pulse label the proximal tubular fluid. The factors that control the rate of fluid flow through the tubules, therefore, control the rate of movement of radioisotope. The interval to the first decline of radioactivity measures the shortest time of passage, and the steepest portion of the curve of declining radioactivity measures the time of passage through the greatest number of nephrons, i.e. the modal passage time. The spread in the data, σ, reflects the degree of disparity in flow rate among the several nephrons. In a steady state the main factors that control flow are the diameter of the tubule, the head of hydrostatic pressure and the viscosity of the fluid.

In dogs with intact renal circulation subjected to increasing osmotic loads, it may be presumed that viscosity is not changed significantly and that the head of pressure either rises or remains constant. Aortic pressure measurements showed no change with increasing osmotic loads. Under these circumstances, with markedly increased luminal bulk flow, the rate of linear flow should increase. The failure of the passage time and the measure of disparity to shorten significantly in these studies implies that tubular diameters increased sufficiently to keep the linear velocity almost constant. This observation conforms to the well known swelling of the kidney that is visible with administration of osmotic loads.

The sensitivity of the passage time and σ to decreases in glomerular filtration rate produced by aortic constriction must reflect an inability of the tubular lumen to contract sufficiently with diminished filtered loads to maintain linear velocity. The great responsiveness of passage time to osmotic loads in the clamped dogs with reduced GFR and continued low arterial pressure suggests that in the low-pressure kidney the tubules are indeed "underfilled" and increased bulk flow rate after osmotic loads produces more rapid linear flow.

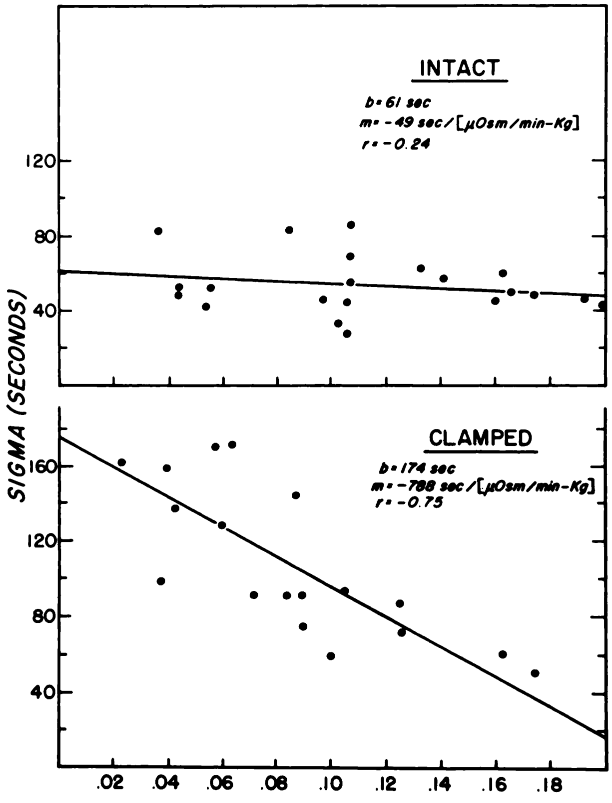

FIG. 5. Sigma is presented as function of osmolar excretion in intact and clamped animals.

In all the situations studied both σ and MPT varied in the same direction and approximately to the same degree (Table 1). Furthermore, the values σ_1 and σ_2 also varied similarly, and the skewness of the curves was independent of the experimental condition. Doubling of the modal passage time is associated with approximate doubling through the entire gamut of passage times. The observations indicate that the maneuvers studied do not preferentially affect one nephron population more than another.

Disparity of passage times could primarily reflect the differences in nephron lengths that exist with relative uniformity of the ratio of GFR to tubular diameter in each nephron. Alternatively, varying ratios of GFR to tubular diameters could produce disparity in passage time. However, if such variation exists to any significant degree, nephrons with large ratios should respond to the experimental stimuli differently than those with small ratios. The similar response of all the measured parameters to the experimental alterations of pressure and bulk flow favors the former hypothesis of a relatively uniform ratio of filtration to tubular diameter in the bulk of the nephrons.

Whereas urine flow rate correlated with σ and MPT in a fashion similar to osmotic excretion, the correlation is poorer. We presume the wide spread in urine flow rates simply reflects the range of osmotic loads and excretions achieved in these studies. Previous studies in man indicate that alterations in urine flow rate produced by water administration do not affect the renogram above a threshold value of 1.5 ml/min urine flow rate (*5*).

These observations clarify the previously reported renographic findings that in patients with renal artery stenosis, prolonged retention of radioisotope in the affected kidney is the most constant abnormality and that reduction in retention is produced by mannitol loads (*1*).

SUMMARY

The modal passage time and the spread of passage times through the renal tubules were determined in dogs following the intrarenal arterial injection of ^{131}I-hippuran. These parameters were related to GFR, osmotic excretion and urine flow rates. An inverse relationship between GFR and passage time was present under all circumstances. In intact animals, passage time was relatively independent of osmotic excretion but in animals with suprarenal aortic constriction, passage time was significantly inversely correlated with osmotic excretion. These data suggest that in intact animals, tubular diameter increases with increasing bulk flow produced by osmotic loads to maintain fairly constant linear velocity. In the "underfilled" state accompanying aortic constriction, increasing bulk flow shortens passage time. The data clarify previous renographic observations that in renal arterial stenosis, mannitol loads preferentially reduce retention of radioisotope in the affected kidney.

REFERENCES

1. Farmelant, M. H., Lipetz, C. A., Bikerman, V. and Burrows, B. A.: Radioisotopic renal function studies and surgical findings in 102 hypertensive patients. *Am. J. Surg.* **107**:50, 1964.

2. Burbank, M. K., Hunt, J. C., Tauxe, W. N. and Maher, F. T.: Radioisotopic renography. Diagnosis of renal arterial disease in hypertensive patients. *Circulation* **27**:328, 1963.

3. Farmelant, M. H., Sachs, C. E., Genna, S. and Burrows, B. A.: Physiological basis of radioisotopic renal function studies. *Clin. Res.* **12**:251, 1964.

4. Farmelant, M. H., Sachs, C. E., Genna, S. and Burrows, B. A.: A physiological model for renal excretion of labeled compounds. *J. Nucl. Med.* To be published.

5. Farmelant, M. H., Dukstein, W. G. and Burrows, B. A.: The effect of varied water loads on the ^{131}I-hippuran renogram. (Unpublished data.)