Changes in Renal Blood Flow, Extraction of Inulin, Glomerular Filtration Rate, Tissue Pressure and Urine Flow With Acute Alterations of Renal Artery Blood Pressure

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IN A PREVIOUS publication (1) it was reported that acute stimulation of the renal nerves in the anesthetized dog caused reductions in renal blood flow (measured directly by a rotameter), the extraction of inulin (filtration fraction), glomerular filtration rate, extraction of Diodrast, and urine flow. The findings indicated that the effects of nerve stimulation were primarily those attributable to constriction of the blood vessels afferent to the glomeruli.

Since an effect, if any, on the efferent arteriole could not be determined or excluded, the present study was designed to demonstrate the hemodynamic effects of a constriction which was known to be exclusively afferent to the glomeruli. This was accomplished by progressive clamping of a tube carrying blood to the cannulated renal artery. In addition, the study was extended to include the experimental changes induced by the converse procedure of progressively increasing above normal the perfusion pressure to the kidney.

METHODS

Mongrel dogs, weighing from 12 to 19 (average 14) kg. were anesthetized with pentobarbital sodium and suspended over the animal board in the prone position by hooks placed through the dorsal skin. In this position an exceptionally good retroperitoneal exposure of the left renal pedicle could be made through the usual lumbar incision. The renal artery, a portion of the renal vein near the kidney, and the ureter were dissected free. The ureter was cut and catheterized with plastic tubing which was connected to a drop recorder. Renal venous blood samples were obtained through a small plastic tube attached to a fishhook-shaped hollow needle inserted into the renal vein close to the kidney. The ovarian or spermatic vein was ligated. In approximately one half of the experiments the renal nerves were dissected free, ligated and cut.

Blood from the left carotid artery was led to a closed 40-cc. reservoir half filled with air, from which the blood could be pumped at any desired perfusion pressure through a 50-cc. pulse dampening chamber half filled with air, to a large optical recording rotameter (2) and then to the renal artery cannula. Arrangement of the apparatus is shown semidiagrammatically in figure 1. The pump consisted of a segment
of 1-inch bore elastic tubing with a valve at each end, activated by a motor driven cam at a rate of 120 rpm. At the side of the cannula was a stopcock from which blood samples could be obtained; from another side tube, mean renal artery blood pressure could be recorded optically. The renal artery was clamped near the aorta, cannulation completed, and blood flow established within 20 to 60 seconds. Systemic blood pressure (BP) was recorded from a T-shaped cannula placed in the right carotid artery. Bypass connections (not shown in fig. 1) were provided for in situ calibration of the rotameter before, after and occasionally near the middle of the experiment. Paritol A² (initial dose 35 mg/kg., and 18 mg/kg. every hour thereafter) was used as the anticoagulant. Inulin was given in a priming dose and the blood level (approximately 20 mg. %) maintained by an intravenous infusion in saline at 2 to 3 cc/minute. Inulin was determined by a modification of the method of Corcoran and Page (3) in the early experiments, and by the method of Rolf, Surtshin and White (4) in the later ones.

The experimental procedure consisted of raising or lowering renal artery blood pressure (RABP) in successive steps, allowing 7 to 10 minutes to elapse after each pressure adjustment before the simultaneous drawing of renal arterial and venous blood samples. Pressures above systemic levels were maintained by adjustment of the stroke volume of the pump, while pressures below systemic levels were obtained by graduated clamping of the tube leading to the renal artery cannula. Hematocrit estimation and total protein with A/G ratio and blood sugar determinations were made after every 2 to 3 adjustments of the renal artery perfusion pressure.

Renal plasma flows (RPF) were calculated from the renal blood flow (RBF) (rotameter) and corresponding hematocrit values [RPF = RBF (rotameter) X 1 - Ht] and are expressed as cc/min/gm. of kidney. Glomerular filtration rate (GFR) was calculated as RPF X extraction percentage of inulin (E%in). Filtration fraction (FF) is expressed as being numerically equal to E %in.

* Generously supplied through the kindness of Dr. J. Seifter, Wyeth, Inc.
RESULTS

Figure 2 shows a composite chart plotting RPF against mean RABP in 13 experiments. In general, the plots are concave to the pressure axis below 80 mm. Hg and convex to the pressure axis above 180 mm. Hg. In most of the experiments when RABP was altered between 80 and 180 mm. Hg a comparatively small or no change in RBF resulted. This tendency of the pressure-flow (P-F) curves to be sigmoid in shape was observed in each experiment and is regarded as characteristic of the relationship for the kidney under the conditions in which these observations were made.

In 2 of the experiments shown the sigmoid nature of the curve was less striking and RBF varied more nearly in direct proportion to the perfusion pressure. Even more linear renal P-F curves (data not shown) were obtained during physiologically poor states, e.g. when the animal's initial BP was only 70 to 90 mm. Hg, after a period of 10 to 15 minutes of renal ischemia, following prolonged perfusion of the kidney at very high pressures with resulting edema, or late in the experiment (after 4–5 hours). By inference, the sigmoid configuration of the P-F curve would appear to be the appropriate representation for the relationship which obtains in the more nearly normal physiological state.

Simultaneous with the P-F observations renal-venous and renal-arterial blood samples were drawn for the estimation of changes in the E%G in 10 experiments. The changes in and differences between arterial and venous blood sugar concentrations were not great enough to require a correction for the inulin determinations. Figure 3 shows a composite chart of the E%G data plotted against mean RABP. Beginning at
around 20 mm. Hg and extending to approximately 80 mm. Hg, E%IN increased rapidly and from 80 to 180 mm. Hg it remained relatively unchanged. A further increase in perfusion pressure above the 180 range was accompanied by a progressive decrease in extraction.

The several negative extraction values obtained at the very low perfusion pressures, and some of the irregularities observable in the curves (fig. 3) are believed to be attributable to random variations in analytic technic. Small errors (1 to 2%) in estimating the concentration in either or both blood samples would materially alter the extraction percentage, particularly when the A-V difference is very small.

![Fig. 3. Effect of altering renal artery BP on the extraction percentage of inulin in 10 experiments.](image)

By multiplying each E%IN by the corresponding value for RPF measured simultaneously, values for GFR were obtained. In figure 4 are shown in composite form the relationships of GFR to RABP in 10 experiments. In general, filtration began at a mean RABP of approximately 20 mm. Hg, increased rapidly and almost linearly with perfusion pressure up to about 80 mm. Hg, above which further increases in perfusion pressure had relatively little effect upon GFR.

In figure 5 are plotted the measurements of urine flow at different levels of RABP. The flow of urine began at an average of 60 mm. Hg and increased exponentially as RABP was elevated.

**DISCUSSION**

From the data presented it is possible to make further correlations which are helpful in understanding the role of several factors involved in the regulation of RBF,
intrarenal resistance, filtration fraction, GFR and rate of urine formation. However, the interpretations which are proposed are necessarily limited, and are possibly applicable only to the preparation and the conditions under which the experiments were performed. For convenience in presentation and to illustrate related trends in one chart, average (and therefore somewhat idealized) plots of the RPF, E\%\textsuperscript{IN}, and GFR data have been constructed from their respective curves (figs. 2–4) and are shown in figure 6.

**Blood Flow.** An intrarenal regulatory mechanism appears to be responsible for maintaining a relatively constant blood flow over a rather wide range of perfusion pressures. The renal nerves are not involved in the regulating mechanism since the P-F curves obtained with 6 denervated and 7 innervated kidneys were not detectably different. In the present experiments the contributing influences from all known extrarenal factors have been excluded and the autoregulation of blood flow is therefore presumed to occur exclusively in response to the mechanical alteration of blood pressure or blood flow to the kidney. The lower and upper limits of the pressure range within which RBF remains relatively unaltered are regarded as the points at which there is approximately maximal intrarenal vascular dilatation and constriction, respectively; these two points, which vary in different experiments (see fig. 2), have an average value of approximately 80 and 180 mm. Hg (fig. 6).

Selkurt (5) has previously reported renal P-F curves (for the normal and low pressure ranges) which were concave to the pressure axis, and also the presence of the
straight portion parallel to the pressure axis indicating an intrarenal flow-regulating mechanism. Denervation was also reported to have had an insignificant effect on the P-F curve. In Selkurt's study the P-F relationships observed in the 20 to 80 mm. Hg range were more curvilinear than those shown in figure 2, but this difference may be related to the fact that he maintained the reduction in RABP for ½ to ¾ minutes and then allowed a return to the control level, while in the present experiments RABP was reduced or increased in successive steps and maintained for periods of 7 to 10 minutes.

On the other hand, Batten et al. (6) found no evidence of autoregulation of flow in the kidney and in addition obtained P-F curves that were predominantly straight or convex to the pressure axis. It is not apparent why the latter results differ from those of Selkurt and from those reported here.

**Fig. 5A.** Composite plot of rate of urine flow with alteration of renal artery BP. Average glomerular filtration rate for all experiments indicated by interrupted line.

**Fig. 5B.** Plot of rate of urine flow as percentage of existing glomerular filtration rate at different levels of renal artery BP.

E% Inulin and GFR. An indication that regulation of blood flow was accomplished primarily by changes in the bore of the afferent vessels is the fact that the E%IN (or FF) was fairly constant over the same perfusion pressure range in which blood flow remained virtually unchanged. Any appreciable contribution to the regulation of flow by the efferent vessels would have caused corresponding changes in glomerular filtration pressure and consequent alterations in E%IN. From studies using clearance methods for measuring RBF and GFR, Forster and Maes (7), and Selkurt (8) have previously suggested that autonomous regulation of RBF may be accomplished primarily by afferent arteriolar constriction.

When RABP was elevated beyond the point of apparent maximum afferent constriction, the E%IN (and FF) was found to decrease progressively as blood flow became increased. The following explanation is suggested for the latter finding. The hydrostatic and osmotic forces which determine FF asymptotically approach, but
never completely come to, equilibrium within the glomerular capillaries by the time the blood leaves the glomeruli (see Lamport, 9). Under these conditions any increase in velocity of blood flow through the glomerular capillaries will allow correspondingly less time for the process of filtration and the attainment of equilibrium; as a consequence a smaller percentage of each cubic centimeter of plasma flowing through the capillaries would be filtered and E/\text{IN} and FF would therefore decrease. However, to increase the velocity of flow through the glomeruli the perfusion pressure must be elevated beyond the point at which the afferent arterioles are capable of further compensatory constriction. At this point some of the increase in perfusion pressure will be transmitted through to the glomerular capillaries, an increase in hydrostatic filtering pressure will result, and an increase in FF would therefore be expected. However, if as glomerular capillary pressure rises there were also to occur an elevation of pressure in the intracapsular space, the resultant differential filtration pressure might not be greatly altered and FF would remain similarly unaffected. Such an increase in intracapsular pressure could not have been caused by the formation of a larger quantity of glomerular filtrate since GFR remained essentially unchanged when the perfusion pressure was elevated to high levels. However, as will be discussed later, direct and indirect evidence was obtained indicating that renal tissue pressure became markedly elevated with progressive increases in perfusion pressure. Since renal tissue pressure is transmitted directly to the intracapsular space it is concluded that intracapsular pressure was correspondingly elevated. Hence, under the conditions existing at high RABP the hydrostatic filtration pressure gradient may have undergone relatively little change, but because of the increase in velocity of blood flow through the glomerular capillaries there was a shorter period of time available for filtration to take place and as a consequence the percentage of plasma filtered was diminished.

The relationship between the increase in RBF and the decrease in FF was such
that GFR (computed as the product of RPF and E\%DM) remained unchanged with successive increases in RABP (figs. 4 and 6).

**Yield Pressure.** Extrapolations of the P-F curves of figure 2 intersect the pressure axis at 2 to 10 mm. Hg (average 7 mm. Hg). The pressure values obtained in this way have generally been regarded as the probable yield pressure or that required to start blood moving through the vascular bed (10, 11). Values of similar magnitude were obtained in another manner, i.e. by clamping the renal artery inflow tube and noting the lowest pressure recorded on the RABP manometer. However, when the manometer was disconnected from the renal artery cannula a small but persistent back bleeding from the kidney always resulted; when the manometer was reconnected the pressure rose from zero to 5 to 10 mm. Hg. Eckstein and Gregg (12) have reported comparable findings following acute clamping of the femoral and carotid arteries. The vascular communications through the renal capsule had not been disturbed in these experiments, and it is most likely that the renal artery pressure recorded at zero flow represented the mean lateral pressure in the renal artery system at the points of communication with the collateral capsular arteries through which a small amount of blood was still being supplied to the kidney.

Williams and Schroeder (13) have previously reported similarly low pressure values of less than 10 mm. Hg at which there was cessation of blood flow through the renal artery even though collateral arterial inflow to the kidney was not excluded. When collateral communications were excluded in the hind limb preparation, extrapolated extentsion of the P-F curves obtained by Pappenheimer and Maes (14) approached or passed through the origin, indicating that there was little or no yield pressure for blood flow in their preparation. Because of the known presence of a collateral blood supply in the experiments reported here the conventionally defined yield pressure, if such exists in relation to the kidney in vivo, could not have exceeded 5 to 10 mm. Hg.

**Urine Flow.** The consistently exponential increase in the flow of urine which was observed with increasing RABP (fig. 5A) was not a consequence of an increase in glomerular filtration (at RABP above 80 mm. Hg) since urine flow increased while GFR remained unchanged. A similar finding in rabbits has been reported by Forster and Maes (7). The relationship of the amount of urine excreted to the simultaneously measured GFR has been expressed as a percentage and is plotted against RABP in figure 5B. At pressures above 260 mm. Hg, as much as 25 to 75 per cent of the glomerular filtrate was excreted as urine.

The fact that the flow of urine increased when RABP was raised from 80 to 180 mm. Hg, in spite of essentially constant RBF, FF and GFR, indicated that some other factor associated with increasing perfusion pressure was responsible for the progressive diminution in reabsorption. Although no objective measurements were made, when the perfusion pressure was suddenly increased or decreased by 40 mm. Hg, palpation of the kidney in situ revealed a perceptible change in the firmness of the kidney which was particularly evident below 80 and above 180 mm. Hg. The palpable changes in the degree of tenseness were regarded as a crude but accurate indication of the corresponding directional changes in renal tissue pressure. According to the following reasoning it is suggested that the increase in renal tissue pressure (RTP) is the pri-
mary but not the only factor responsible for the observed augmentation of urine flow. The peritubular capillary and the fluid in the interstitial space are subjected to the same RTP, and no change in the ‘reabsorption gradient’ across the capillary wall would be expected because of a change in RTP alone. The tubule is also subjected to the same RTP, but it possesses a comparatively thick wall that would not be easily collapsed from the application of a concentric compressive force. Because of the physical structure of the tubule, RTP would not be transmitted without loss through the wall and thence to the contents of the lumen. If the RTP outside the tubule is materially greater than the pressure of the fluid within the lumen the centrifugal passage of water through the tubule wall must proceed against an unfavorable hydrostatic pressure gradient. The greater the RTP the more unfavorable the pressure gradient will become for water to leave the tubule. It would also be anticipated that with the rise in RTP, glomerular capsular pressure would increase, and as the volume of unreabsorbed filtrate and its velocity of flow along the tubule become greater the intratubular fluid pressure would also rise. However, the rate of increase and the extent to which the intraluminal pressure can become elevated is limited in that the unreabsorbed filtrate is constantly escaping from the kidney as urine and its maximum rate of flow presumably cannot exceed the existing GFR. It therefore appears that the intratubular fluid pressure would be increased indirectly with elevation of RABP but that the relatively greater increase in RTP surrounding the tubule would be the more influential factor in determining the slope of the hydrostatic pressure gradient across the tubule wall.

As a theoretical extrapolation, there exists the possibility that if RTP were to be increased to an extent that the hydrostatic pressure difference across the tubule wall exceeded the forces tending to cause fluid to pass from the lumen of the tubule to the interstitial space, a reversal in the direction of fluid transfer would be anticipated. Water (and other substances to which the peritubular capillaries are permeable) would move from capillary to interstitial space and replace that which had been forced into the tubule. Under these conditions the flow of urine could conceivably exceed the GFR. In the present experiments there is no significant evidence that would either support or argue against the possibility that such a mechanism could operate to increase urine flow above the existing GFR.

Although RTP will theoretically affect the transport of water through the tubule at all levels of RABP, two additional factors may reasonably operate to cause a diminution of water reabsorption at high levels of perfusion pressure. If at approximately 180 mm. Hg the afferent arterioles become maximally constricted and the RABP is increased further, the added increment of pressure can no longer be dissipated by further increase in resistance from constriction of the afferent vessel; the increase in perfusion pressure will then be transmitted, with progressive diminution, on through the vascular bed. The fact that blood flow through the kidney was increased at perfusion pressures above 180 mm. Hg indicates that there must have existed an elevated hydrostatic pressure, exceeding RTP throughout all portions of the renal vascular system, including the peritubular capillaries. Also to be considered is the fact that as RBF increased, FF (E%ON) decreased (fig. 3) and as a consequence the oncotic pressure of the blood flowing through the capillaries must have
been diminished. Because of the increased hydrostatic pressure and the decreased oncotic pressure of the plasma, the capacity for capillary reabsorption of water at the high perfusion pressures would be correspondingly decreased and a greater flow of urine would be anticipated.

Renal Tissue Pressure. The influence of changes in RTP on several functions pertinent to this study have already been considered under the sections on filtration and urine flow. The RTP appears to be derived from the hydrostatic pressure of the arterial blood supplied to the kidney. Each of the renal blood vessels is distended by an intravascular pressure, ranging in magnitude from RABP to renal venous pressure. By virtue of their expansibility the various blood vessels, if they were to be removed from the surrounding kidney tissue, would exhibit an increase in volume with increasing intravascular pressure according to their respective pressure-volume (distensibility) characteristics. However, since the renal vascular system in situ is surrounded by semifluid tissue elements and the whole enclosed by the renal capsule, distention of the blood vessels can occur only when an equivalent volume of kidney substance is displaced with consequent distention of the surrounding renal capsule. The kidney tissue and interstitial fluid which is interposed between the renal vessels and the renal capsule is therefore subjected to a pressure (RTP) which is determined primarily by the volume-distensibility characteristics of the renal capsule.

In the present experiments RBF, E%IN, and GFR remained essentially unchanged when RABP was raised from 80 to 180 mm. Hg (fig. 6), while urine flow showed a progressive increase (fig. 5A). If the observed relative constancy of RBF and FF is attributable to regulation by afferent arteriolar constriction, then the entire arterial vascular tree proximal to the afferent arteriole must have been increasingly distended as perfusion pressure was raised. The resulting increase in volume of the vessels would cause an equivalent displacement of kidney substance, an increased distention of the renal capsule, and an increase in RTP. Through such a mechanism a rise in RTP could result from an elevation of RABP and cause an increased urine flow without concomitant changes in RBF, FF or GFR.

Intrarenal Resistance. An attempt was made to estimate the changes in resistance which occurred afferent and efferent to the glomeruli following alteration of RABP. By applying Lamport's formulae (11) or their modification (8) the computed resistance values were not unreasonable for the pressure range between 80 and 180 mm. Hg. Progressive increases in afferent arteriolar resistance with increasing RABP were invariably found. However, above 180 and below 80 mm. Hg the formulae were found not to be applicable in that improbable or negative resistances were obtained. It is suspected that some of the factors included in the resistance calculations were changing variables and could not be correctly expressed as constants. In addition, it appears probable that there do not exist at the high and low perfusion pressures the necessary conditions for hydrostatic and oncotic equilibrium upon which are based the calculation and separation of afferent and efferent resistances.

Some of the possible reasons why the conventional calculations are believed to be inapplicable to the present experiments are given briefly as follows. At the high range of RABP when the velocity of blood flow through the glomerular capillaries became increased there may have been insufficient time for the hydrostatic and osmotic forces,
which determine the fraction of plasma filtered, to come to or even approach equilibrium, as suggested by the observed decrease in FF. If a filtration equilibrium is not attained, the calculated oncotic pressure will underestimate by an unknown amount the hydrostatic pressure existing in the glomerular capillaries, and, as a result, a corresponding error will be introduced in computing both afferent and efferent resistances. Similarly, because of the same increase in velocity of flow, a 'reabsorption' equilibrium may not have occurred by the time blood reached the venular ends of the peritubular capillaries and as a consequence there would not exist a terminal point of reference for calculating efferent resistance. At very low perfusion pressures (of less than approximately 20 mm. Hg when there was no filtration) the amount by which intraglomerular capillary pressure was less than oncotic pressure cannot be determined. At pressures between 20 and approximately 60 mm. Hg, when there was filtration but no urine formation, it is presumed that oncotic pressure exceeded hydrostatic pressure in the peritubular capillaries, but by an amount which is not determinable.

Because of the marked changes in RTP which occur with alterations of RABP, values for RTP, to be appropriate for the calculation of resistance, would need to be measured by a reasonably accurate method simultaneously with the recording of RABP, RBF etc.

Another factor involved in the calculations is the yield pressure, the value for which is unknown. In the present experiments it was not demonstrated that there existed a minimum pressure required to start blood flowing through the kidney, and consequently values for yield pressure could not be assigned.

Because of capsular collateral vessels through which there may be a transmission

Fig. 7. Plots of intrarenal resistance computed as mean renal artery BP minus extrapolated pressure at zero flow, divided by renal blood flow.
of pressure and flow of blood to or from the renal artery system, a variable inaccuracy will be inherent in estimating pressure-flow relationships when using the measurements of RABP and RBF through the renal artery. Without attempting to introduce a variable factor which would tend to correct for the influence of collateral communications, total intrarenal resistance was plotted for each experiment as mean RABP minus the extrapolated pressure value at zero flow, divided by the measured RBF (see fig. 7). Expressed in this manner the average resistance for all experiments may be compared with the average RBF in figure 8. It is reasonable that there would be an increase in intrarenal resistance if, while RABP was being elevated (between 80 and 180 mm. Hg) there was no appreciable increase in RBF. Also, above 180 and below 80 mm. Hg there was little alteration in total resistance with changes in RABP. However, due to the fact that an unrecorded amount of blood was supplied to the kidney through collaterals at low RABP, and escaped from the kidney at high RABP, more specific interpretations of intrarenal resistance computed in this way would appear to be unwarranted.

**Summary**

In anesthetized dogs one renal artery was cannulated *in situ* and the blood pressure to the kidney mechanically altered and maintained for periods of 7 to 10 minutes at levels from 20 to over 320 mm. Hg. Simultaneously measured were renal blood flow (RBF) (by recording rotameter) and the extraction of inulin (E%IN) from which were computed renal plasma flow and glomerular filtration rate (GFR). Urine flow was also measured. From perfusion pressures of approximately 20 mm. Hg, RBF, E%IN and GFR increased rapidly up to approximately 80 mm. Hg, above which all three functions remained relatively constant until perfusion pressure reached approximately 180 mm. Hg. Above 180, RBF increased rapidly, E%IN decreased, and the resulting GFR remained essentially unchanged. Flow of urine began at an average of
60 mm. Hg and increased exponentially, until at very high perfusion pressures the flow of urine was 25 to 75 per cent of the existing GFR.

The relative constancy of RBF and E%\text{on} between 80 and 180 mm. Hg is believed to be an indication of autonomous regulation, effected by changes in tone of the afferent arterioles.

Evidence suggests that distention of the renal blood vessels resulting from increased intravascular pressure caused an increased renal tissue pressure which was primarily responsible for a diminished transfer of filtrate through the tubular wall and the consequent increase in urine flow.

The presence of a collateral blood supply through the capsular arteries prevented the measurement, or determination of the existence, of a 'yield pressure' for the renal vascular bed.

Indications that certain factors including yield pressure, renal tissue pressure, filtration equilibrium, and reabsorption equilibrium were indeterminable, variable, or nonexistent, suggested an explanation for the many improbable values obtained for afferent and efferent arteriolar resistances as calculated from the usual formulae.

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REFERENCES