The Goldblatt Memorial Lecture

Part I: Experimental Renovascular Hypertension

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It was a great privilege and pleasure for me to deliver a memorial lecture for Dr. Harry Goldblatt. I had long admired his pioneering studies on renovascular hypertension but it was only in recent years that I came to know him personally. It was my good fortune, at one of the dinners of the Council for High Blood Pressure Research in Cleveland, to be seated with Dr. and Mrs. Goldblatt. During that delightful evening I was accepted into the extended Goldblatt scientific family when they discovered that we had several common bonds; both Mrs. Goldblatt and I had been affiliated with the Peter Bent Brigham Hospital in Boston, and second, Dr. Allan M. Butler, who was the first to show that hypertension of pyelonephritic origin was cured by removal of the diseased kidney, had been my teacher. When Dr. Goldblatt learned of this latter association, he spoke of one of Butler's patients, a young girl whose blood pressure fell postoperatively from 260/160 to 110/60. "She grew up," he said, with an infectious chuckle, "to be a typical American woman, — she had two children, and three husbands!" I was captivated by Dr. Goldblatt's charm, his wit, his erudition, his concern, his gentleness, and his humility. It was only later that I learned that he was also an authority on prints, etchings, and paintings, and that he had worked his way through McGill University as a narrator for French-Canadian movie audiences, orally translating English captions on old-fashioned silent films into French. He was truly a man for all seasons. And yet, despite his enormous contribution to medicine, Dr. Goldblatt did not live long enough to receive a well-deserved Nobel Prize. However, as early as December 5, 1939, Dr. Walter Bradford Cannon, Chairman of the Department of Physiology at Harvard Medical School, had proposed Dr. Goldblatt for the award. In his letter to the Nobel Prize Committee of the Royal Caroline Institute, Cannon wrote: "Another candidate whose investigations seem to be worthy of high esteem is Dr. Harry Goldblatt of Cleveland, Ohio. His production of hypertension by reducing the blood flow through the renal vessels has opened a significant new field of research. It has stimulated investigations in the United States and in a number of foreign countries, and it has offered an explanation of a long-standing mystery of human illness. He and his collaborators have made a valuable advance in medical knowledge."

What motivated Dr. Goldblatt to examine the role of the kidney in the pathogenesis of hypertension? Apparently, the original stimulus occurred many years before his experiments began. He wrote that "as a resident in surgery, in 1916, I witnessed a tragic event which did stimulate me to think about hypertension. On another service, a diagnosis was made of a tumor of the lower pole of the kidney, in a middle-aged individual, and I witnessed the nephrectomy. It proved to be an anomalous, half-horseshoe kidney; there was no tumor, and, unfortunately, no other kidney. The patient survived about 6 days, and I had the opportunity to follow the fatal course. I was eager to learn whether the blood pressure would rise as uremia developed. Although profound azotemia did occur, yet at no time during this period was there any elevation of blood pressure. I remember that this came as a surprise to me, because I had seen patients die in uremia as a result of severe inflammatory, or congenital cystic disease of both kidneys, and they usually had con-
siderable elevation of blood pressure. I may as well confess that this was the first time that I gave any consideration to the problem of the origin of hypertension in general, and all I really salted away from the experience was, no kidneys, no hypertension. It was only in 1928 after I had been a pathologist for a goodly number of years, and had performed the autopsies on a large number of hypertensive patients, that I began to give serious thought to the problem of the origin of hypertension . . . \(^9\)

Although Dr. Goldblatt began to give serious thought to the problem of the origin of hypertension in 1928, the first public presentation concerning his investigations was not made until 1933, and the first of a series of papers entitled “Studies of experimental hypertension” was published in 1934.\(^1\) Today, Dr. Goldblatt would have some difficulty explaining to a study section why only one paper appeared in 6 years. Months were required to prepare the carotid or van Leersum loops necessary for blood pressure measurements in the conscious dog, and in the training of these animals. The problem of the method for producing renal ischemia was solved with the development of the famous silver Goldblatt clamp. And then, four of the dogs had to be followed for 15 to 18 months after the production of renal artery stenosis. However, in 1934, he was able to state unequivocally that “these experiments indicate that in dogs, at least, ischemia localized to the kidneys is a sufficient condition for the production of persistently elevated systolic blood pressure.” He went on to say that “it is hoped that these investigations will afford a means of studying the pathogenesis of hypertension that is associated with renovascular disease.”

As Cannon had noted in his nomination of Dr. Goldblatt for the Nobel Prize, Dr. Goldblatt’s work stimulated investigation in the United States and in a number of foreign countries. Houssay, the Argentine Nobel Laureate, in his prologue to Braun-Menerdez’s book entitled Renal Hypertension,\(^4\) wrote: “The modern period is due entirely to Goldblatt. . . .” Braun-Menerdez and coworkers, in the first page of the text described the Cleveland research as follows: “In the year 1934, Goldblatt, Lynch, Hanzal, and Summerville made a brilliant discovery by showing that reduction in the amount of blood brought to the kidney gave rise to permanent hypertension in the dog. This discovery changed the orientation of investigations on hypertension. . . .”\(^4\) And from England, Sir George Pickering has written: “The greatest single advance in the experimental production of the condition resembling that of hypertension in man was a convincing demonstration by Goldblatt, Lynch, Hanzal, and Summerville, that persistent hypertension could be produced in dogs by constricting both renal arteries, or one if the other kidney had been removed.”\(^6\)

Goldblatt and his colleagues also demonstrated that ischemia of other organs did not lead to hypertension, that neural factors were not responsible since the excision of the entire thoracic portion of the splanchnic nerves and the lower four dorsal sympathetic ganglia, or section of the anterior spinal roots did not prevent the development of the elevated pressure following renal artery constriction, that hypertension could be induced in species other than the dog, that hypertension could be produced in the hypophysectomized animal, and that the adrenal medulla was not essential but that the adrenal cortex played a significant role.\(^8\) What then is responsible for the rise in pressure following renal artery constriction in the dog?

Goldblatt wrote that “the earliest indication that a humoral mechanism may be responsible for the elevated pressure in experimental renal hypertension was the effect of tying off the renal vein in dogs with the main renal artery constriction adequately to produce hypertension. All of these animals developed uremia, and died from 2 to 7 days, yet at no time did they show any elevation of blood pressure. . . .”\(^6\) Confirmation of the thesis that a pressor agent was present in the renal venous blood came from the renal transplantation studies of Houssay and Fasciolo.\(^7\) They reported that the grafting of an ischemic kidney into the neck of a nephrectomized dog raised the blood pressure of the recipient animal; transplanting a normal kidney into the neck did not induce hypertension. Houssay and Taquin\(^8\) then demonstrated that the renal venous pressor agent produced vasoconstriction in the perfused hindlimb of the toad, particularly if the blood was removed from dogs whose hypertension was only of a few days duration, — the first suggestion, I believe, that the ischemic kidney released more pressor agent during the initiation of renovascular hypertension than during the chronic stage of the syndrome.

The late thirties and early forties was an exciting period in the study of renovascular hypertension. Page and Helmer\(^9\) in this country, and Braun-Menerdez et al.\(^10\) in Argentina, had demonstrated the enzymatic nature of renin and identified the pressor agent, called angiotonin by Page and hypertensin by Braun-Menerdez. Since investigators are more likely to use each others’ toothbrushes than they are to use each others’ nomenclature, it was some time before a common terminology was agreed upon. The compromise was finally achieved, not by an international committee, but by Page and Braun-Menerdez at a cocktail party over a couple of martinis. Thus, the alcohol soluble pressor peptide was renamed angiotensin.

During the next decade, Goldblatt continued his investigations on the pathogenesis of renovascular hypertension, with particular emphasis on the role of renin. In 1943 he defined the unit of renin as the quantity which, when injected intravenously into a normal, unanesthetized dog, would cause a rise of direct mean pressure of 30 mm Hg in at least three dogs, — the well-known Goldblatt unit.\(^11\) He was keenly interested in the immunologic approach of Wakerlin and Johnson\(^12\) to the treatment of hypertension with crude renin, and began to purify the enzyme for his own studies of antirenin. The first extensive purification of renin was done by Lamfrom, Haas and Goldblatt in 1953.\(^13\) From that time on, one is impressed by the number of acknowledgments in publications from all over the world which state: “We are indebted to Drs. Harry Goldblatt and Erwin Haas for their generous
supply of standard renin."* The long and fruitful collaboration of Goldblatt and Haas continued to Goldblatt's retirement in 1976 from the directorship of the Louis D. Beaumont Memorial Research Laboratories at the Mt. Sinai Hospital in Cleveland.

The next major advances in the renin-angiotensin story also occurred here in Cleveland when Skeggs and his colleagues isolated and sequenced the decapeptide, angiotensin I, and purified "the hypertensin-conveting enzyme" which is responsible for the production of the vasoactive octapeptide, angiotensin II.* Angiotensin II was soon synthetized simultaneously in Cleveland by Bumpus and coworkers and in Basle by Schwyzer and his group. Finally, the development of a radioimmunooassay for measurement of plasma renin activity by Haber et al. the synthesis of angiotensin II antagonists by Marshall et al., Bumpus, Pals, and Poulsen, the demonstration by Bakhle that the venom peptides from Bothrops Jararaca were potent inhibitors of the angiotensin converting enzyme (CEI), the synthesis of the nonapeptide CEI by Ondetti et al., and the purification of renin from a number of species enabled a number of laboratories to reexamine more precisely the role of the renin-angiotensin system in the pathogenesis of renovascular hypertension.

In our laboratory my colleagues have been primarily interested in the simpler form of renovascular hypertension, the one-kidney Goldblatt model. Since the unanesthetized dog was Goldblatt's favorite experimental subject (fig. 1), it would seem appropriate to focus primarily on the studies in the trained, unanesthetized dog. In the operative preparation employed in our laboratory, one kidney is removed and catheters implanted in the aorta and inferior vena cava below the kidneys, and in the renal artery distal to an inflatable cuff secured around the renal artery close to the aorta. The proximal end of each catheter, exteriorized through a hollow needle, is tied to a plastic loop implanted in the skin. The catheters are flushed daily with saline, then filled with heparin and sealed with stainless steel obturators. Thus, in the conscious, trained dog, we are able to regulate renal perfusion pressure by monitoring the pressure drop across the stenosis induced by cuff inflation. The physiological changes observed in such an animal on normal salt intake (50-80 mEq/day) are schematically summarized in figure 2 from the experiments of Gutmann, and Miller and coworkers.* With reduction of renal arterial pressure, systemic plasma renin activity rose steadily, reaching a peak level in the first 24 hours, with a parallel doubling of plasma aldosterone concentration. Aortic blood pressure increased concurrently with the elevated plasma renin activity, and continued to rise more gradually over the next 24 to 48 hours. Marked sodium retention occurred during the first several days until a new steady state was achieved. Increased water intake was observed in the first hours, and plasma volume reached a higher steady-state value in 2 to 3 days. Although the renal perfusion pressure was maintained at a constant level, plasma renin activity and plasma aldosterone concentration returned to control values over several days. Follow-

*Although Dr. Goldblatt freely provided purified renin for other investigators, he refused co-authorship unless he actively participated in the research. His attitude regarding this delicate subject is well exemplified in the following letter to Dr. Phyllis Hartroft dated January 30, 1961:

"I am sure that you realize that I have supplied renin and antirenin to workers all over the world, but never have I permitted my name to be placed on a paper resulting from the work done without my active participation. The only thing I could possibly allow you to do, as I have permitted others to do, is to mention that the preparations were made and supplied to you by Dr. Haas and me. Even this is unnecessary, but you may do so, if you wish. You may rest assured that it is a pleasure to both Dr. Haas and me to be of help to other investigators in the field, and any and all materials which we have supplied have always been given without any strings attached. I really do hope that you will understand this is no reflection whatsoever upon the quality of your work. In fact, I am greatly impressed with the care with which this study was evidently carried out."

†Goldblatt's reservations concerning experimental data obtained in anesthetized animals was expressed in the following excerpt from a letter written to Dr. Ephraim Shorr on November 21, 1955: "... We have been playing a lot with ganglionic blocking agents in the experimental animal, that is, the unanesthetized trained animal, just because so little work has been done on the unanesthetized animal and we have perfected this method. Much that applies to the anesthetized animal does not (apply) to the unanesthetized."
implying deflation of the cuff, renal arterial pressure rose to equal aortic pressure. A brisk natriuresis ensued over the first 24 hours, but blood pressure returned to baseline more slowly, reaching control level after several days. From these data we postulated that the elevated renin-angiotensin levels were responsible for the initial rise in blood pressure in the one-kidney Goldblatt dog, but that the sustained hypertension was due to other factors, such as the salt and water retention, and increased plasma volume.

The availability of the nonapeptide angiotensin converting enzyme inhibitor, SQ20,881 (Glu-Trp-Pro-Arg-Pro-Gln-Ile-Pro-Pro), which blocks the conversion of angiotensin I to the pressor octapeptide angiotensin II, enabled us to demonstrate that the initial rise in blood pressure was causally related to the rise in plasma renin activity and could be prevented by the drug. Furthermore, when pressure was allowed to rise with renal artery constriction, the converting enzyme inhibitor restored blood pressure to normal in the first hour or days. However, by the fifth to the seventh day of constriction, when plasma renin activity was again essentially normal, no hypotensive action of a single dose of the converting enzyme inhibitor was observed, findings which have been confirmed in other laboratories. The next logical step to delineate the role of the renin-angiotensin system in the initiation of renovascular hypertension was the blockade of the conversion of angiotensin I to angiotensin II by a constant infusion of the nonapeptide converting enzyme inhibitor over the first 4 days of renal artery constriction; blood pressure did not rise.\(^{31}\) Plasma bradykinin was not detectably elevated throughout the infusion. Further strong supporting evidence for the role of renin in the initiation of renovascular hypertension has been provided by Dzau et al.\(^{42}\) by the use of renin-specific antibodies obtained by immunization of goats with canine renal renin purified to homogeneity.

Riegger and coworkers\(^{58}\) have recently reported experiments in dogs in which systemic blood pressure was elevated by angiotensin II infusion or by renal artery constriction, and the levels of angiotensin II in the plasma measured. The data indicate that the rise in blood pressure following renal artery constriction can be quantitatively related to the rise in angiotensin II, and that no other factors appeared to play a significant or quantitative role. Thus, a considerable body of evidence indicates that initiation of renovascular hypertension is dependent on release of renin from the kidney and the generation of angiotensin II. However, in dogs on normal or high salt intake, plasma renin activity and angiotensin II levels return to normal within the first week despite the maintenance of a constant renal arterial pressure. During the latter period, the positive salt and water balance may play a significant role in the maintenance of sustained hypertension, a view championed by Guyton: “The most important principle of our views is that all conditions that cause hypertension do so by reducing the ratio of the renal excretory capability, on one hand, to the intake of water and electrolytes, on the other hand. This controls the degree of filling of the circulatory system with blood volume, which is paramount to the long-term control of arterial pressure and the genesis of hypertension.”\(^{57}\) Although, as I have indicated, our studies on renovascular hypertension in dogs on normal salt intake would appear to be in agreement with this thesis, we had not previously addressed specifically the question of a pathogenic role of salt and water retention. To examine this question, Rocchini and Barger\(^{35}\) have recently induced renovascular hypertension in salt-depleted animals maintained on low sodium-chloride intake.

Figure 3 illustrates the response of such salt-depleted, trained dogs to the same degree of renal artery hypotension as in the previous experiments. Blood pressure rose more rapidly than in the salt-replete animals, reached a peak within the first hour, and remained at this elevated level for the duration of the experiments; plasma renin activity also was maximally elevated in the first hour and plasma aldosterone concentration was closely correlated with renin activity. Unlike the response of the salt-replete animal, plasma renin activity remained elevated throughout the 14-day experiment. Water intake rose rapidly, followed by a comparable increase in urine output. Urinary sodium loss exceeded intake. On the
first day after constriction, plasma volume may have been slightly elevated but gradually and significantly decreased below control value by the fifth day as a result of increased sodium and water excretion; body weight also decreased. Although serum sodium concentration fell by the third day, it returned to control value by the seventh day; serum potassium was unchanged. Following the release of the constricting cuff, systemic pressure declined by 10 mm Hg at 1 hour and was nearly back to control level by 6 hours, a striking difference from the pattern observed in the salt-replete animal. To provide further evidence that the renin-angiotensin system was still responsible for the elevated blood pressure to the very end of the experiment, converting enzyme inhibitor was given simultaneously with the release of the cuff on the 14th day (fig. 4). Instead of the progressive decline in arterial pressure observed over the first 6 hours in the control experiments, a 54 mm Hg drop in pressure occurred within 5 minutes of the drug administration; prior to the constriction, the inhibitor produced only a 15 mm Hg drop in pressure. Thus, in the salt-depleted dog, in which salt and water retention do not occur, the renin-angiotensin system appears to play a major role in both the initiation and maintenance of renovascular hypertension. Gavras and his colleagues have previously shown that salt depletion made the one-kidney Goldblatt hypertensive rat responsive to the hypotensive action of Saralasin.

With this new information we can reexamine the mechanism by which the elevated blood pressure is maintained in the salt-replete one-kidney Goldblatt dog. Whether the sustained hypertension is due to the salt and water retention and increase in plasma volume, or to the reduction in concentration of vasodilator agents, such as bradykinin or prostaglandins, or to the release of vasoconstricting agents other than renin as proposed by Grollman, and Krishman-Amerty by Skeggs et al. and by Boucher et al. is not yet established. Riegger and the other members of the Glasgow group, however, have proposed that angiotensin II is responsible for the initial rise in systemic pressure and for the sustained hypertension in sodium-replete dogs. They have confirmed the earlier observations of Dickinson and Lawrence and McCubbin and co-workers that slow, chronic infusion of subpressor doses of angiotensin II leads to a progressive rise in blood pressure after a delay of some days. In addition, they have demonstrated that the dose-response curves for angiotensin II superimposed on long-term infusion are shifted to the left as pressure...
rises, that is, a given plasma level of angiotensin II is associated with a greater change in blood pressure. They conclude that the early renal hypertension results from acute vasoconstriction produced by angiotensin II, and that a second, slower developing pressor mechanism of angiotensin II becomes progressively more important as renal hypertension ensues. Thus, in the latter phase, a normal or only slightly elevated concentration of angiotensin II is able to maintain the elevated pressure. They suggest that among the factors that may be responsible for the slow developing pressor mechanism may be the positive salt and water balance. Thus, sodium balance would appear to be a major factor determining not only the responsiveness of the renin-containing cells in various physiological and pathophysiological states as proposed by Gross et al. many years ago, but also the responsiveness of arteriolar smooth muscle cells to angiotensin II. Moreover, we must also keep in mind, as DeChamplain and co-workers have emphasized, that sodium chloride balance also modifies the activity of the sympathetic nervous system. Therefore, let us examine the relative effectiveness of the renin-angiotensin system and the carotid sinus reflex in the regulation of blood pressure in low- and high-salt states.

Fray et al. examined the release of renin induced by the same reduction in renal artery pressure in the conscious dog maintained successively with high-, normal-, and low-sodium intake. In the dogs with high-salt intake, 45 minutes of renal artery constriction produced little or no increase in plasma renin activity; aortic pressure did not rise. In the dogs with normal-salt intake, systemic renin activity rose from 0.6 to 5.5 ng • ml⁻¹ • hr⁻¹, while blood pressure gradually increased by 21 mm Hg. The blood pressure response in the salt-depleted dog was unexpected. Aortic pressure rose more rapidly than in the salt-replete animals, as plasma renin activity rose to 50 ng • ml⁻¹ • hr⁻¹. Thus, in the sodium-depleted animal, the enhanced ability to release renin makes the renin-angiotensin system a very effective pressure regulator during volume depletion; in high-salt states it plays little role in regulation of pressure. In contrast to the above observations, Rocchini et al. have shown that the blood pressure response to a similar degree of hypotension in the carotid sinus (25 mm Hg) produced by constricting van Leersum loops in the trained dog is higher in the salt-replete animal, and is attenuated by 50% in the salt-depleted dog. Figure 5 provides a quantitative comparison of the effectiveness of the renin-angiotensin system and the carotid sinus reflex in the elevation of blood pressure in the sodium-depleted animal (left) and sodium-replete dogs (right). For a given drop in pressure, the renin-angiotensin system is three times as effective as the sympathetic nervous system in raising blood pressure in the salt-depleted state. Conventional wisdom might suggest that the carotid sinus reflex would be more important in the regulation of blood pressure during volume depletion, but, as DeChamplain et al. have reported, the turnover of norepinephrine is markedly depressed by sodium depletion even though tissue norepinephrine content is greater than in the sodium-replete animal. Hence, it is not surprising that, in the volume-depleted dog, Rocchini and co-workers found that the blood pressure dose-response curve to tyramine, a drug that releases norepinephrine from nerve terminals, was shifted to the right of that obtained in the sodium-replete dog; that is, a higher concentration of tyramine was required to produce an equivalent rise in blood pressure. The vascular responsiveness to norepinephrine was unchanged by varying sodium chloride intake. The observations of reduced responsiveness of the sympathetic nervous system during volume depletion also raises the interesting question whether one of the important long-term effects of diuretic agents in hypertensive patients may not be mediated, in part, by inhibition of sympathetic function induced by sodium chloride loss.

Although four decades have passed since Dr. Goldblatt reported his first studies on experimental renovascular hypertension and the release of a humoral pressor agent into the circulation, our knowledge of the cellular events in the juxtaglomerular apparatus that control the release of renin is only fragmentary. For the future exploration of this important facet of the problem of hypertension, newer methodology must be used or developed specifically for a more precise description of the cell-to-cell interactions and the intracellular events. This will necessitate that the training of future investigators in the field of hypertension be broadened to enable our students to apply the methodology of cellular biology, of immunology, of neurobiology, etc., and to utilize the newer physical tools such as nuclear magnetic resonance and proton- or electron-probe microanalyses. Then we may be able to understand how the

**Figure 5.** Comparison of the rise in blood pressure induced by carotid sinus (solid line) and renal arterial (broken line) hypotension in dogs on low-, normal-, and high-sodium intake (mean ± se) (Reproduced by permission from Rocchini AP, Cant JR, Barger AC: Carotid sinus reflex in dogs with low- to high-sodium intake. Am J Physiol 233: H196, 1977)
various neural, hormonal and humoral mechanisms interact at the juxtaglomerular apparatus of the kidney to control the release of renin. When the cellular processes are more clearly defined we shall be able to appreciate how the baro- or stretch-receptor function of the afferent arteriole, first postulated by Tobian, is modified by the sympathetic nervous system. And perhaps we shall be able to resolve the conflicting views of Vander and Miller regarding the role of the sodium chloride load at the macula densa of the distal tubule. Recently Taugner and co-workers, using freeze fracture techniques, have demonstrated gap junctions between most of the cellular elements of the juxtaglomerular apparatus, hydrophilic channels which allow the passage of ions and small molecules from cell to cell without significant leak into the extracellular space. Their observation that is particularly surprising is the absence of gap junctions between the macula densa cells and the other cells of the region, a finding to which I shall return in a moment. Hartroft first presented evidence that renin granules were present in the afferent arterioles of the glomeruli. The renin-containing cells can be separated from other elements of the afferent arteriole and identified by staining the renin granules with neutral red (fig. 6). Staining of the renin granules with neutral red has enabled Fishman to identify the myoepithelial cells in isolated afferent arterioles and to measure their transmembrane potential with microelectrodes. The stable resting potential fell into two distinct populations: a low-potential group, with a mean of —35 millivolts, and a higher potential group, with a mean of —70 millivolts. Surprisingly, epinephrine, which has been shown to increase renin release from kidney slices, hyperpolarized the cells instead of producing the depolarization seen in other secretory systems, while a high concentration of potassium chloride, which decreases renin release, depolarized the cells. Both the epinephrine- and potassium-induced potential changes are similar to those observed in smooth muscle cells from which juxtaglomerular cells are derived. One additional feature of the smooth muscle membrane potential is its variation with stretch. Fishman hypothesized: "In view of the baroreceptor theory of renin secretion, it is tempting to postulate a similar mechanical sensitivity for the juxtaglomerular cell membrane. Thus, renin secretion could be partially controlled by changes in arteriolar or interstitial pressures near the juxtaglomerular cell, as signalled by changes in its membrane potential." 

Another seemingly anomalous response of the juxtaglomerular cells is the reaction to changes in calcium concentration. Whereas calcium appears to be necessary for release of hormones from many secretory cells, Fray has demonstrated that lowering calcium concentration in fluid perfusing isolated rat kidneys induces a marked increase in renin release. Baumbach and Leyssac reports similar data in isolated, superfused glomeruli and have concluded that calcium has a direct action on the juxtaglomerular cells influencing the release of renin by a mechanism entirely different from exocytosis, and more likely related to cell volume regulation. Although many anatomical studies have suggested that the myoepithelial cells of the afferent arteriole was a major site of renin synthesis, Kresiberg et al. have been able to dissociate cells from isolated glomeruli and to clone mesangial cells which also appear to release a renin-like material. Thus, several types of cells in the juxtaglomerular region, with connecting gap junctions, may release renin. As I have already indicated, the macula densa cells do not appear to have gap junctions with other cell types in this region. What, then, is the role of the macula densa cells of the distal tubule? The macula densa is presumed to be the receptor through which changes in distal fluid composition may alter renin release. The tubuloglomerular feedback mechanism is thought to involve transepithelial ion movement at the macula densa. Although the transport properties of the adjacent thick ascending limb and distal convoluted tubule have been established, the transport properties of the macula densa cells remain unknown. Because of the association between sodium-potassium activated ATPase and active ion transport, quantitative data concerning the activity of this enzyme system in the macula densa would aid in the analysis of the function of these cells. Guth and Albers have developed a histochemical method for localization of sodium-potassium ATPase by means of the closely associated potassium-dependent, ouabain-sensitive, para-ni-
trophenyl phosphatase activity. Beeuwkes and Rosen have modified the technique for quantitative analysis of the enzyme system using electron-probe microanalysis. They have shown, both histochemically and by electron-probe microanalysis that in kidneys from dogs, rats, and rabbits, the highest levels of sodium-potassium ATPase activity are found in the thick ascending limb and distal convoluted tubule. In striking contrast to the high activity observed in the adjacent nephron segments, little or no reaction product was found within macula densa cells regardless of history of dietary sodium intake. If the ability to establish high transepithelial sodium-chloride gradients is required for transduction, then these results are inconsistent with the transducer role for the macula densa cells. It is clear that although these studies show a relative lack of the sodium-potassium ATPase system, they do not define the ac-

References

25. Slater EE, Cohn R, Dzau VJ, Haber E: Complete purification of human renin. Circulation 58 (suppl II): 11-249, [969], 1978
26. Dzau VJ, Slater EE, Haber E: Pure dog renin. Circulation 58 (suppl II): 11-249 [971], 1978


