Hyperplasia of the Juxtaglomerular Complex with Hyperaldosteronism and Hypokalemic Alkalosis*

A New Syndrome

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with comments by

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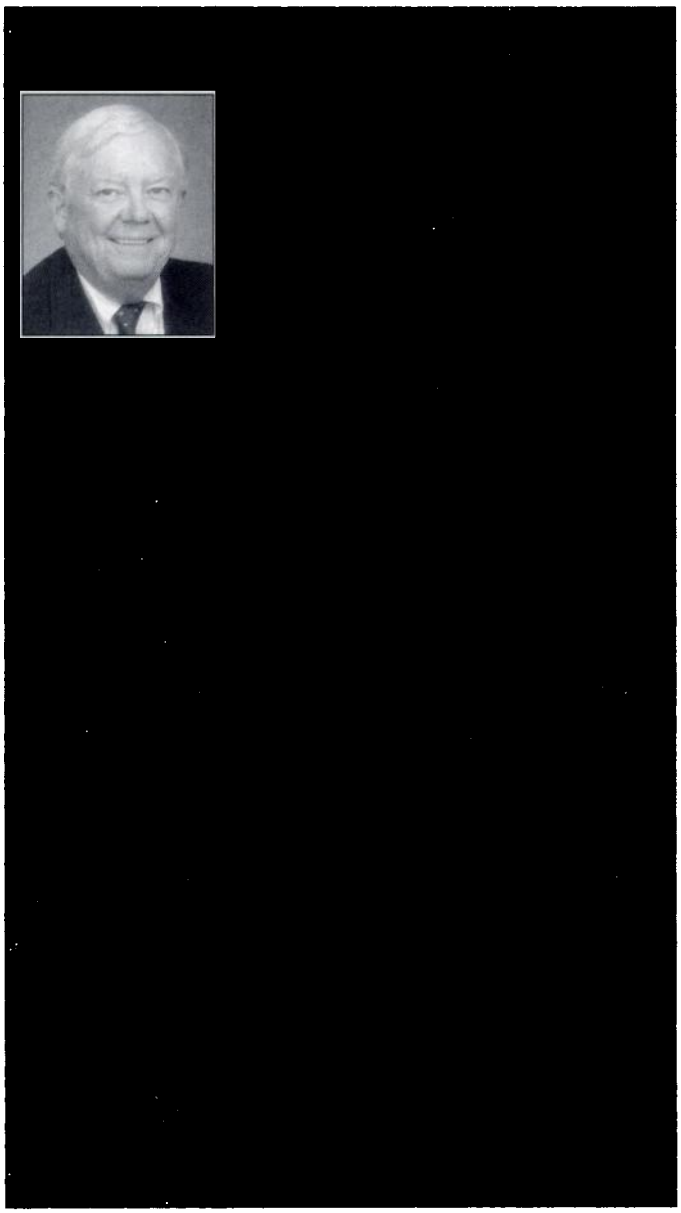

HYPERPLASIA and hypertrophy of the juxtaglomerular apparatus, and primary aldosteronism with hypokalemic alkalosis were found associated with normal blood pressure in two patients [1]. This coexistence of a histologic lesion not previously described with a rare disease entity suggests an association between them; the appearance of these disorders in two patients who have never had hypertension appears to represent a new syndrome.

We here report studies designed to elucidate the pathologic physiology of this syndrome. The findings provide strong support for the growing body of evidence that adrenal cortical secretion may be influenced by the renin-angiotensin system.

CASE REPORTS

Patient C. J. was a five year old Negro boy who had been admitted to Children's Hospital, Washington, D. C., with complaints of tetany and dwarfism, at the age of four years and ten months. He weighed 8 kg. and appeared dehydrated. Carpopedal and quadriceps femoris spasms were present, and Chvostek's sign was positive. The serum calcium was normal, but the tetany would respond temporarily to calcium gluconate therapy. Hypokalemia was found, and he was referred to the National Institutes of Health for further studies.†

Full term normal delivery had followed an uneventful pregnancy. His birth weight was 3 kg., his birth length 42 cm.

At the age of four months he was hospitalized because of fever of one week's duration, associated with vomiting, diarrhea, dehydration and generalized convulsions. A lumbar puncture, skull roentgenograms and gastrointestinal series revealed no abnormalities; slight albuminuria was present. A pneumoencephalogram showed slight dilation of the lateral ventricles. Since discharge he had been well except for retardation of growth and polydipsia which required him to drink 10 to 12 glasses of fluid daily. His family history was non-contributory.

†We are indebted to Dr. Joseph Lo Presti for referring this patient.

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Physical examination revealed a short (36 inches), proportionately dwarfed Negro boy (span = 37 inches) who weighed 8 kg. and appeared moderately dehydrated. The blood pressure ranged between 92/52 and 120/80 mm. Hg on numerous determinations with an average of 100/60 mm. Hg. Chvostek's sign was positive and carpopedal spasm readily appeared when he cried. Physical and neurologic examinations were otherwise non-contributory.

Numerous urine specimens showed a pH of 7 or above, specific gravity 1.010 or below, occasionally a trace of albumin, and no sugar.

Chemical analysis revealed the following serum values: Potassium 2.2 mEq. per L., carbon dioxide content 34 mEq. per L., chloride 75 mEq. per L., sodium 130 mEq. per L., and blood urea nitrogen 15 mg. per 100 ml. The result of a serologic test for syphilis was negative.

Repeated electroencephalograms all showed "diffuse dysrhythmia." Several records also showed "bilaterally synchronous paroxysmal activity of the atypical centrencephalic type." The electrocardiogram revealed a "T-vector of low magnitude, rotated posteriorly (juvenile pattern), S-T segment depression and prominent U waves in leads V_2 through V_4." Roentgenograms revealed a bone age of three years; and an intravenous pyelogram was normal.

Adrenal cortical function and reserve were normal as regards hydrocortisone and 17-ketosteroid excretion. Aldosterone excretion was elevated, as shown in numerous metabolism studies (*vide infra*).

Urinary concentrating ability was somewhat impaired: dehydration for twenty-four hours induced a loss of 2.5 per cent of body weight and a rise of urine osmolality to 717 mOsm per kg., not further increased by the administration of Pitressin® (aqueous Pitressin 10 units intramuscularly). Glomerular filtration rate (inulin) was 30 ml. per minute or 68 ml. per minute per 1.73 M^2. Twenty-four hour urinary amino acid nitrogen was 1.10 per cent of total nitrogen (normal, 1 to 2 per cent).

The patient had always been a poor eater and craved salt. He was constipated and required an enema every other day. Prior to correction of the hypokalemia he had several convulsive seizures which did not resemble carpopedal spasm; these did not recur while the hypokalemia was being successfully treated. Before operation this was accomplished with potassium chloride supplements (134 mEq. per day), a low sodium diet and human serum albumin given intravenously once a week. Appetite and growth rate increased with treatment and constipation disappeared. The electrocardiogram reverted to normal.

In view of the persistent aldosteronism, partial adrenalectomy and renal biopsy were performed when the patient was eight years old. The findings will be discussed subsequently. Muscle analysis showed a decrease of intracellular potassium and an increase of intracellular sodium. For the next fifteen months the patient remained symptom-free without sodium restriction or potassium supplements, but serum potassium remained low (2.3 to 2.8 mEq. per L.), and urinary aldosterone high (e.g., 38 μg. per day on a daily sodium intake of 110 mEq.).

Fifteen months after surgery, tetany returned, and the patient was readmitted. Serum potassium was 2.0 mEq. per L., serum carbon dioxide 29 mEq. per L., and serum sodium 139 mEq. per L. Urinary aldosterone was 5 μg. per day, and aldos-

To our surprise, the article occasioned a rapidly increasing number of publications reporting observations on the condition that was quickly coming to be known as Bartter's syndrome. It is of interest that Fred himself initially resisted this classification, preferring to refer to the syndrome as juxtaglomerular hyperplasia. After partially relenting to "so-called Bartter's syndrome," he eventually capitulated and joined the rest of the medical world.

The advent of the prostaglandin era in the 1970s led to new insights about the pathogenesis of Bartter's syndrome and, more importantly, changed the focus from the vasculature to the renal tubule as the principal player in the etiology of the disorder.

Patients with Bartter's syndrome overproduced prostaglandins; when they were treated with a prostaglandin synthase inhibitor, their renin-angiotensin-aldosterone system returned to normal. Their blood pressure, unaffected by the treatment, then responded normally to angiotensin II. In contrast to the improvement noted above, hypokalemia persisted. These observations, together with the demonstration that experimental potassium depletion increased prostaglandin formation and reproduced most of the clinical features of the syndrome, led to the conclusion that potassium loss by the kidney, rather than a defect in the vasculature, was the cause of the syndrome.

With regard to the kidney, I cannot explain why the presence of nephrocalcinosis in patient M.W. was not further pursued initially. When we got around to measuring urinary calcium, it was high. In only two of the many other patients with the syndrome that we had an opportunity to study was urinary calcium high. In fact, in all of the other patients urinary calcium was either low or undetectable. The low urinary calcium was usually associated with a high urinary magnesium and hypomagnesemia. Patient C.J. had a low serum magnesium that would place him in this far larger group.

Thus, in retrospect, C.J. and M.W., although they share many of the same clinical features, probably do not have the same disorder. More likely, each has a distinct tubular transport abnormality that is different from the other.

The pioneering studies of Dr. Richard Lifton and his associates, using the powerful tools of molecular biology, represent a watershed period in the history of Bartter's syndrome. These scientists have been able to identify the defective renal transporters responsible for two varieties of Bartter's syndrome and the gene defects that give rise to them.

I am grateful for the publication of our article in the series "Milestones in Nephrology." It has provided a wonderful opportunity for me to reflect on a most fascinating journey from phenotype to genotype, in which so many have participated over the past four decades. Little did we realize that this uncommon condition would occasion so much interest and contribute as much information as it has.

terone secretion 750 μg. a day. An assay of serum angiotensin concentration gave a figure of 240 mμg. per 100 ml., an approximately eightfold increase above the normal figure.

Patient M. W. was a twenty-five year old Negro man with a history of enuresis, slow growth, weakness and fatigue which had prevented him from entering school until the age of twelve. He had been seen first at twelve years of age in the Pediatric Clinic, Duke University Medical Center, with a history of vomiting and pain in the abdomen and calves. At the age of sixteen he had been admitted to Duke University Medical Center in a semicomatose condition with a blood pressure of 104/70 mm. Hg. Chemical analysis revealed the following serum values: potassium 1.28 mEq. per L., chloride 41.3 mEq. per L., and carbon dioxide 58.6 mEq. per L. Proteinuria was present. The electrocardiogram was reported as showing changes of hypokalemia. Pyelography revealed dilation of the middle third of both ureters. The patient was treated with potassium chloride and rapidly recovered.

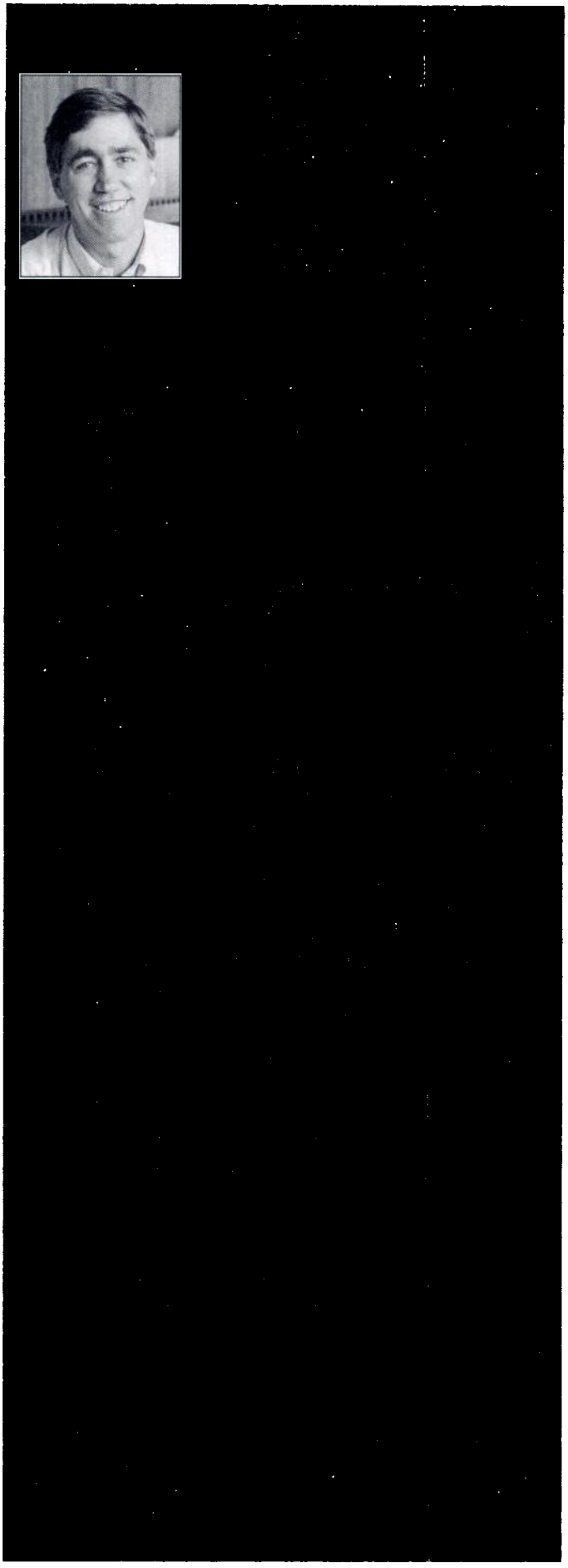
He returned to the clinic at the age of nineteen. He had not taken potassium chloride consistently, and weakness, polydipsia, polyuria, enuresis and episodic cramps in his hands and legs had persisted. Serum potassium and chloride were 1.8 and 82 mEq. per L., respectively. He was admitted to Duke University Medical Center for study. Chvostek's and Trousseau's signs were elicited. The urine again contained protein (2 plus). He excreted 65 per cent of a dose of phenolsulfonphthalein in two hours. Serum potassium and carbon dioxide were 2.0 and 30 mEq. per L., respectively, non-protein nitrogen was 30 mg. per 100 ml. Serum calcium, phosphorus, total protein and albumin and globulin were normal. Eosinophil count was 55 per cu. mm., with a fall to 5 per cu. mm. after an eight-hour infusion of ACTH. He was then referred to the National Institutes of Health for further studies.

Physical examination revealed a thin Negro boy 65 inches tall who weighed 45.8 kg. Blood pressure ranged between 124/70 and 94/52 mm. Hg on numerous determinations, with an average of 105/75 mm. Hg. Chvostek's sign was positive. Physical and neurologic examinations were otherwise non-contributory.

Numerous urine specimens showed a pH of 7 or above, specific gravity 1.014 or below, trace to 1-plus albumin and no sugar. Chemical analysis revealed the following serum values: potassium 2.0 mEq. per L., carbon dioxide 37 mEq. per L., chloride 77 mEq. per L., sodium 132 mEq. per L., blood urea nitrogen 9 mg. per 100 ml. The result of a serologic test for syphilis was negative.

An electroencephalogram showed "diffuse dysrhythmia with no epileptiform discharges or focal abnormalities." Repeated electrocardiograms revealed "T-vector of low magnitude, S-T segment depression and prominent U waves in leads V_2 through V_4." A roentgenogram revealed that the epiphyses of the radius, ulna and iliac crest had not fused; calcific deposits were present in the region of the calyces in both kidneys. Urinary excretion of 17-hydroxycorticoids and 17-ketosteroids was 1.8 and 5.6 mg. per day, respectively. Aldosterone excretion was elevated (*vide infra*).

Maximal urinary osmolality after dehydration for four hours and Pitressin (aqueous Pitressin 200 milliunits intravenously and 500 milliunits subcutaneously every thirty minutes) was



only 254 mOsm per kg. Glomerular filtration rate (creatinine) was 105 ml. per minute. He has consistently refused adrenal exploration; tissue was obtained from the right kidney by percutaneous punch biopsy. An assay of serum angiotensin concentration gave a figure of about 70 mμg. per 100 ml., a clear increase above the normal figure.

METHODS

Metabolic balance studies were carried out to measure (1) the effects of changing sodium intake (see Fig. 1, 2 and 6), (2) the effects of changing potassium intake (Fig. 2 and 3), (3) the effects of acid, (4) the effects of albumin given intravenously (Fig. 3 and 11), and (5) the effects of spirolactones (Fig. 4 and 5). (These studies were performed prior to surgery in C. J.). The final studies were made to determine [6] the effects of angiotensin and of renin (after surgery in C. J.). The adrenal (C. J.) and renal (C. J. and M. W.) biopsy specimens were studied by several histochemical technics (Fig. 9 through 14). Many of the methods employed have been previously reported from this laboratory [2]. In addition, in some studies urinary aldosterone was determined by the method of Kliman and Peterson [3], urinary titratable-acidity-minus-bicarbonate was measured by a modification of the method of Dawson and associates [4], urinary ammonium by the method of Conway [5], urinary 17-ketosteroids by a modification of the method of Zimmerman [6], and urinary and plasma 17-hydroxycorticoids by the method of Peterson [7].

Effects of Changing Sodium Intake (Fig. 1, 2 and 6). On a high sodium intake (67 and 117 mEq. in C. J. and M. W., respectively) both patients showed hypokalemic alkalosis and excreted excessive amounts of aldosterone (20 to 40 μg. per day). When the sodium intake was

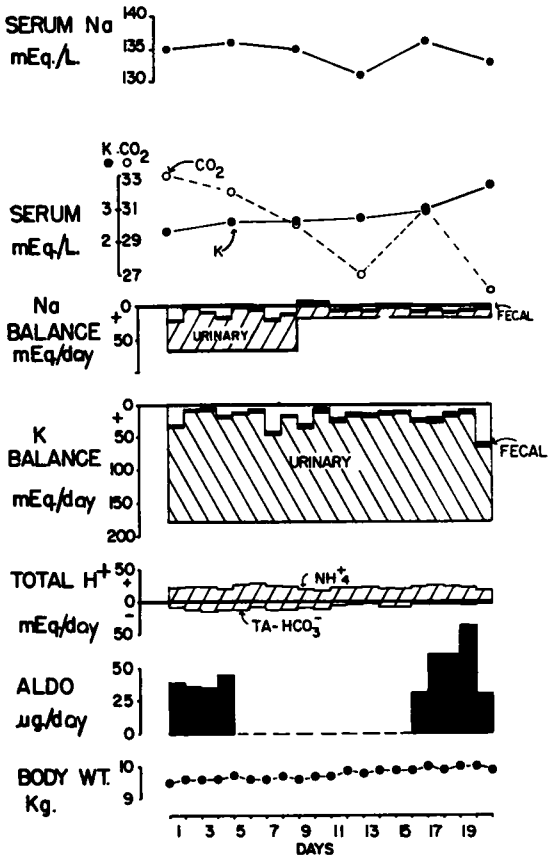


FIG. 1. Patient C. J. Effect of changing sodium intake on serum sodium, potassium and carbon dioxide concentration, sodium and potassium balance, urinary hydrogen ions and aldosterone and body weight.

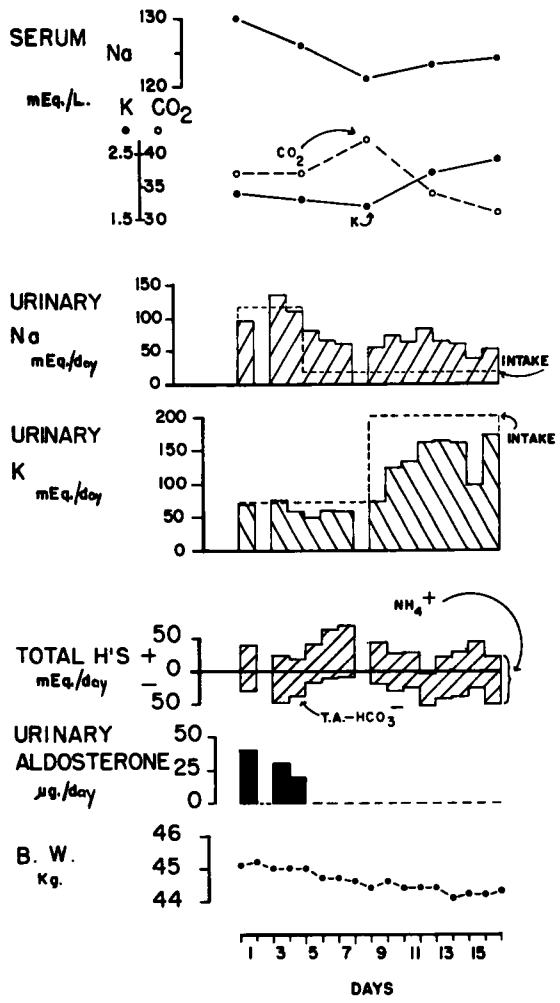


FIG. 2. Patient M. W. Effect of changing sodium intake on serum sodium, potassium and carbon dioxide concentration, urinary sodium, potassium, hydrogen ions and aldosterone and body weight.

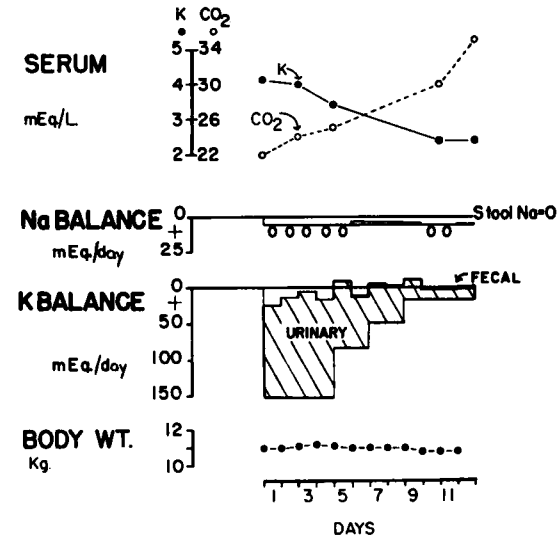


FIG. 3. Patient C. J. Effect of stepwise reduction of potassium intake on serum potassium and carbon dioxide concentration, sodium and potassium balance and body weight.

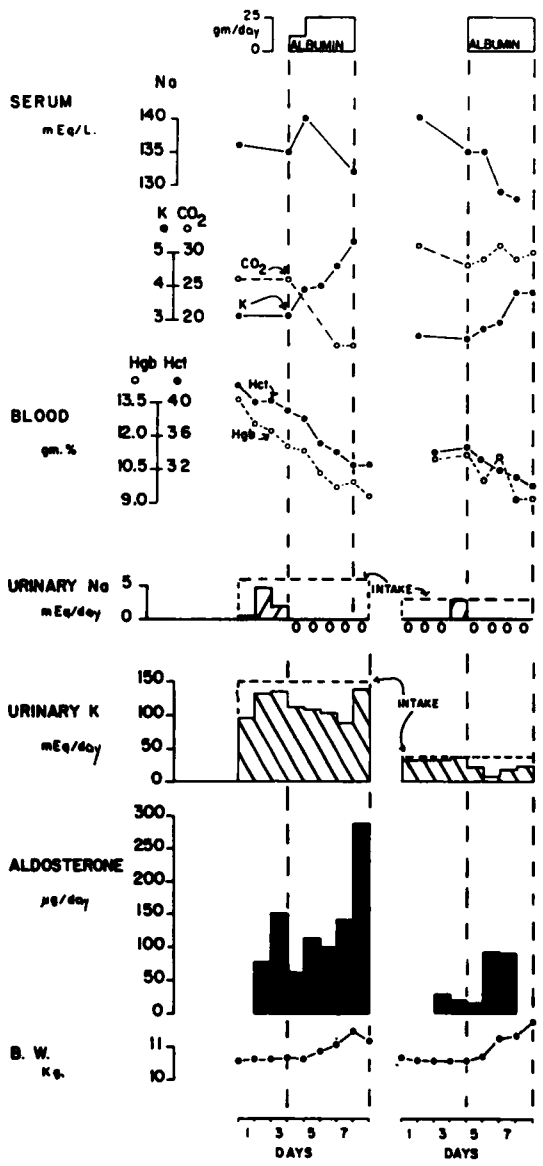


FIG. 4. Patient C. J. Effect of human serum albumin infusion on serum sodium, potassium and carbon dioxide concentration, blood hemoglobin and hematocrit, urinary sodium, potassium and aldosterone and body weight.

lowered to 17 mEq. a day, urinary sodium fell to intake values in C. J. (Fig. 1), serum potassium and carbon dioxide returned towards normal, and serum sodium did not change. Urinary potassium did not change.

In another study (Fig. 6), the sodium intake was abruptly increased from 6 to 106 mEq. per day. This induced loss of potassium, a fall of serum potassium from 5.1 to 2.0 mEq. per L. and a rise of serum carbon dioxide from 25 to 34 mEq. per L.

In M. W., on the other hand (Fig. 2), when sodium intake was lowered to 17 mEq. per day, urinary sodium exceeded the sodium intake, serum potassium did not change, serum carbon dioxide rose and serum sodium fell. Urinary potassium did not change. After four days on this regimen (Fig. 2) symptoms of hypokalemia became worse, and potassium intake was increased as a therapeutic measure.

Effect of Changing Potassium Intake (Fig. 2 and 3). In C. J., when potassium intake was lowered stepwise (Fig. 3), potassium balance became slightly negative, urinary potassium not falling significantly

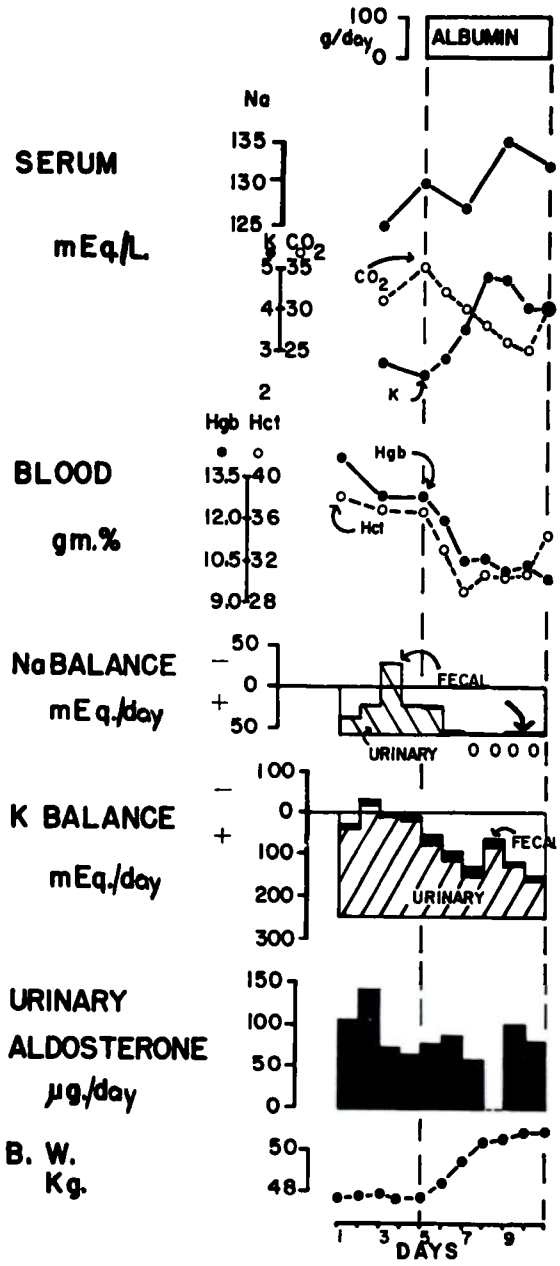


FIG. 5. Patient M. W. Effect of human serum albumin infusion on serum sodium, potassium and carbon dioxide concentration, blood hemoglobin and hematocrit, sodium and potassium balances, urinary aldosterone and body weight.

below intake despite restriction of sodium intake to 6 mEq. per day, and serum potassium fell while serum carbon dioxide rose. In another study (not shown) when potassium intake was lowered abruptly, the same results were obtained and urinary aldosterone fell progressively—presumably an effect of potassium depletion *per se* [8].

In M. W., on an intake of 70 mEq. of potassium a day (Fig. 2), urinary potassium remained close to the potassium intake despite restriction of sodium intake to 17 mEq. per day, serum potassium remained below 2 mEq. per L. and serum carbon dioxide rose. As already noted, the patient's clinical condition precluded a study with a lower potassium intake.

Effects of Albumin Given Intravenously (Fig. 4 and 5). When human serum albumin was given intravenously, both patients showed large decreases in hemoglobin concentration and hematocrit and increases in body weight. Urinary sodium fell from 2 (C. J.) and 36 (M. W.) mEq. per day, to zero in both patients. In C. J., who received 5 mEq. of sodium a day, serum sodium fell; while in M. W., who received 60 mEq. of sodium a day, it rose. In both patients there was retention of potassium and a rise of serum potassium. Urinary aldosterone rose in C. J., and did not change in M. W.

Effect of Spirolactones (Fig. 6 and 7). When aldosterone antagonists were given there was an increase in urinary sodium, with a negative sodium balance, in both patients. In both patients there was a fall of urinary potassium with potassium retention, as serum potassium rose sharply (from 2.6 to 6.8 mEq. per L.) in C. J. and slight (from 2.2 to 2.6 mEq. per L.) in M. W. Serum carbon dioxide had decreased by the first day after treatment in C. J., whereas it decreased throughout treatment in M. W. Urinary aldosterone rose in both patients.

Effects of Renin and of Angiotensin. During the studies with renin and angiotensin serum potassium ranged from 2.4 to 3.8 mEq. per L. in C. J., and from 1.6 to 2.4 mEq. per L. in M. W.

When angiotensin II* was given to C. J. intravenously at rates of 0.005, 0.01, 0.05 and 0.1 µg. per kg. per minute (weight 17.7 kg.), blood pressure increased from control levels (range: 90/55 to 96/60 mm. Hg) to sustained peak values of 92/64, 98/60, 106/80 and 128/98 mm. Hg for each of these doses, respectively; the pulse rate decreased from control levels (range: 80 to 88) by 6 to 12 beats per minute with each dose.

When angiotensin II was given to M. W. intravenously at rates of 0.05, 0.1, 0.2 and 0.4 µg. per kg. per minute (weight

* We are greatly indebted to Drs. Franz Gross and Robert Gaunt of Ciba and Co. for angiotensin I and II.

potent diuretic antihypertensive agents that do not have significant extrarenal effects.

During a period of explosive growth in our understanding of the molecular basis of inherited human disease, it is particularly appropriate to acknowledge the key contributions made by physicians performing detailed clinical investigation of patients with rare diseases. As the molecular bases of these diseases are uncovered, one anticipates that there will be important new opportunities for further investigation of the physiology of these and related diseases based on new molecular insights. Recognition of this opportunity, however, also raises the concern of whether a cadre of physicians will still exist with the training, time, and resources to conduct these studies with the same skill and care exemplified by this Milestone article.

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2. Simon DB, Karet FE, Hamdan JH, Di Pietro A, Sanjad SA, Lifton RP: Bartter's syndrome, inherited hypokalemic alkalosis with hypercalciuria, is caused by mutations in the Na-K-2Cl cotransporter NKCC2. *Nat Genet* 13: 183–188, 1996
3. Simon DB, Karet FE, Rodriguez-Soriano J, Hamdan JH, DiPietro A, Trachtman H, Sanjad SA, Lifton RP: Genetic heterogeneity of Bartter's syndrome revealed by mutations in the K+ channel, ROMK. *Nat Genet* 14: 152–156, 1996
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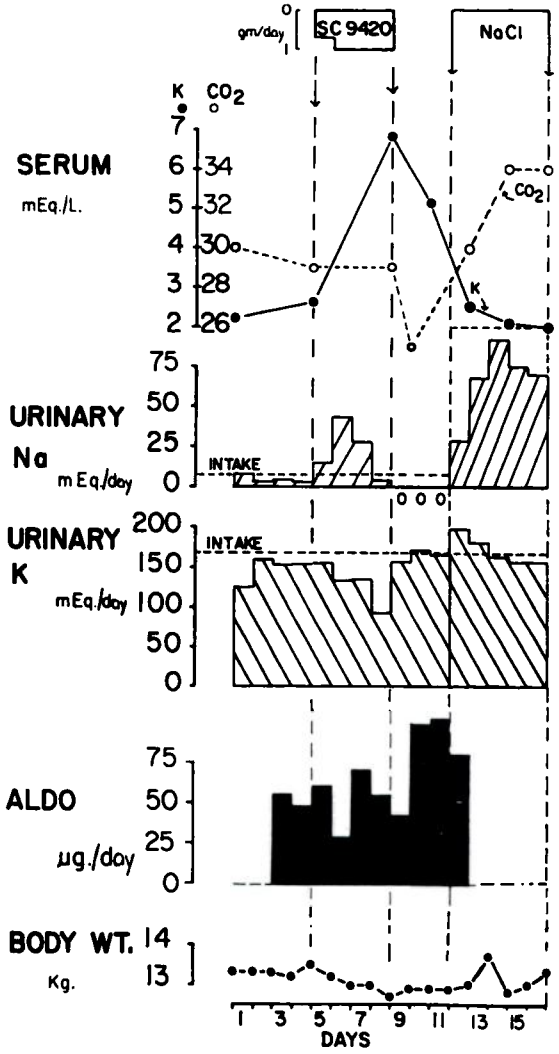

FIG. 6. Patient C. J. Effect of SC-9420 (Aldactone) and a low sodium intake on serum potassium and carbon dioxide concentration, urinary sodium, potassium and aldosterone and body weight. Also the effect of high sodium intake on these same measurements.

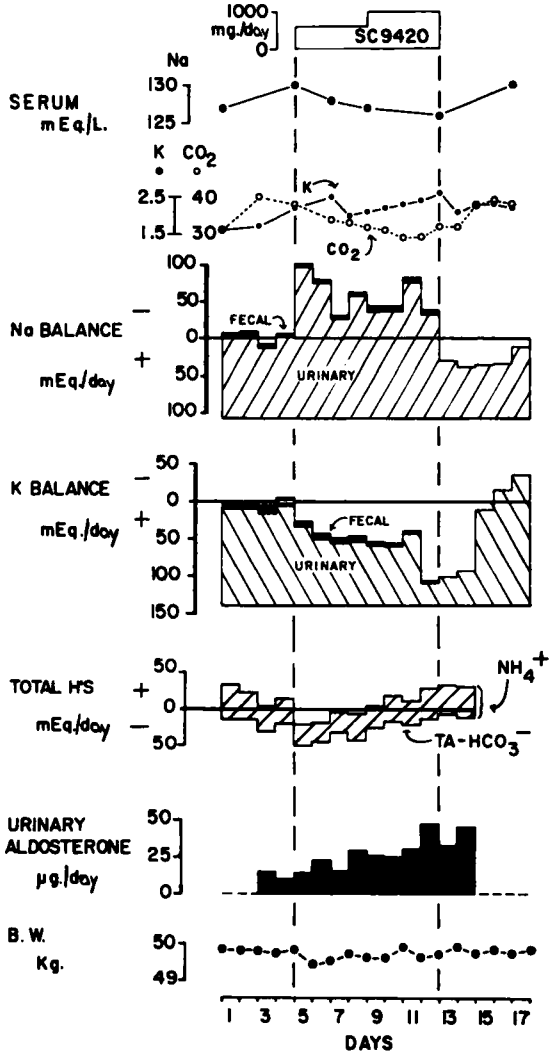

FIG. 7. Patient M. W. Effect of SC-9420 (Aldactone) on serum sodium, potassium and carbon dioxide concentration, sodium and potassium balances, urinary hydrogen ions and aldosterone and body weight. There were no fecal measurements during the last five days.

45.8 kg.), blood pressure increased from control levels (range: 104/66 to 114/76 mm. Hg) to peak values of 118/84, 124/88, 140/104 and 172/112 mm. Hg for each of these doses, respectively; the pulse rate decreased from control levels (range: 72 to 78) by 4 to 8 beats per minute with each dose. Blood pressure then declined from these peak values, ***despite continued infusion of angiotensin***, to pressures of 110/80, 112/82, 128/94 and 140/108 mm. Hg, respectively, with the doses given. When angiotensin I was infused at the rate of 5 µg. per minute the blood pressure increased from 106/70 to 114/80 mm. Hg. The effects of angiotensin II on blood pressure in these patients and in several normal subjects are shown graphically in Figure 8.

HISTOLOGIC STUDIES

Patient C. J.

The kidneys and adrenal glands in C. J. appeared to be normal upon gross examination at the time of operation. The left adrenal gland was excised *in toto* and weighed 2.7 gm. Eight-tenths of a gram of tissue was removed from the right adrenal gland, and a biopsy of the right kidney was performed.

Adrenal tissue was fixed in 4 per cent formaldehyde, embedded and cut in the usual way, and colored with hematoxylin and eosin. Histologically, the sections showed limited areas of hypertrophy of the zona glomerulosa which consisted of two distinct layers: (1) a compact outer layer of small cells with hyperchromatic nuclei, and (2) an inner layer of large, clear cells arranged in the form of blind tubules, abutting the peripheral cells, which did not seem to be a part of the zona fasciculata. The thickness of the zona glomerulosa, considered as consisting of both of these glomerulosa layers, was approximately twice that of the zona glomerulosa of a normal child of the same age. In some areas in which the peripheral layer was hyperplastic, the inner tubular layer was not discernible.

The kidney specimen consisted of a wedge about 10 mm. wide and 4 mm. thick. It contained about 60 glomeruli. It was cut in two, and one section was fixed for six hours in Zenker's formol solution (90 ml. of stock Zenker fluid and 10 ml. of 40 per cent formaldehyde). The other section was fixed and refrigerated for four days in Regaud's fluid (80 ml. of 3 per cent potassium bichromate and 20 ml. of 40 per cent formaldehyde) and mordanted in a refrigerator for eight days

in 3 per cent potassium bichromate for mitochondrial stains [9]. Sections were cut 5 and 6 μ thick and colored by ordinary hematoxylin and eosin, the Cowdry-Bensley modification of Altmann's acid-fuchsin-aniline blue, the periodic acid-Schiff reaction, or by Bowie's strain [10,11] for granules of the juxtaglomerular cells.

About 60 per cent of the uriniferous tubules showed hyperplasia of the juxtaglomerular apparatus, and the walls of the glomerular vessels of about 20 per cent of these were hyperplastic (Fig. 9 and 10). In these hyperplastic apparatuses the wall of the afferent arteriole was found to be 80 to 100 μ in thickness, whereas the wall of this vessel in the kidney of a normal child of the same age was only 30 μ in thickness. The juxtaglomerular cells of only two Malpighian bodies contained so-called JG granules, colorable with Bowie's stain. The cells of the juxtaglomerular perivascular collar (juxtaglomerular cells of Hartroft) were found to be long, thin and fibroblastic in nature, unlike the glandular type of cell in the normal apparatus.

The macula densa of the uriniferous tubule was enlarged to accommodate as many as 30 or 40 nuclei within the mural roof adjacent to the juxtaglomerular cells and the prominent pohlkissen body. The basement membrane was continuous throughout the length of the afferent arteriole, forming an abnormal membrane obstructing the space normally existing between the juxtaglomerular wall and the macula densa. (There is no such membrane in this position in a normal kidney.)

The process of hyperplasia of the juxtaglomerular apparatus appeared from fixed preparations to comprise four stages

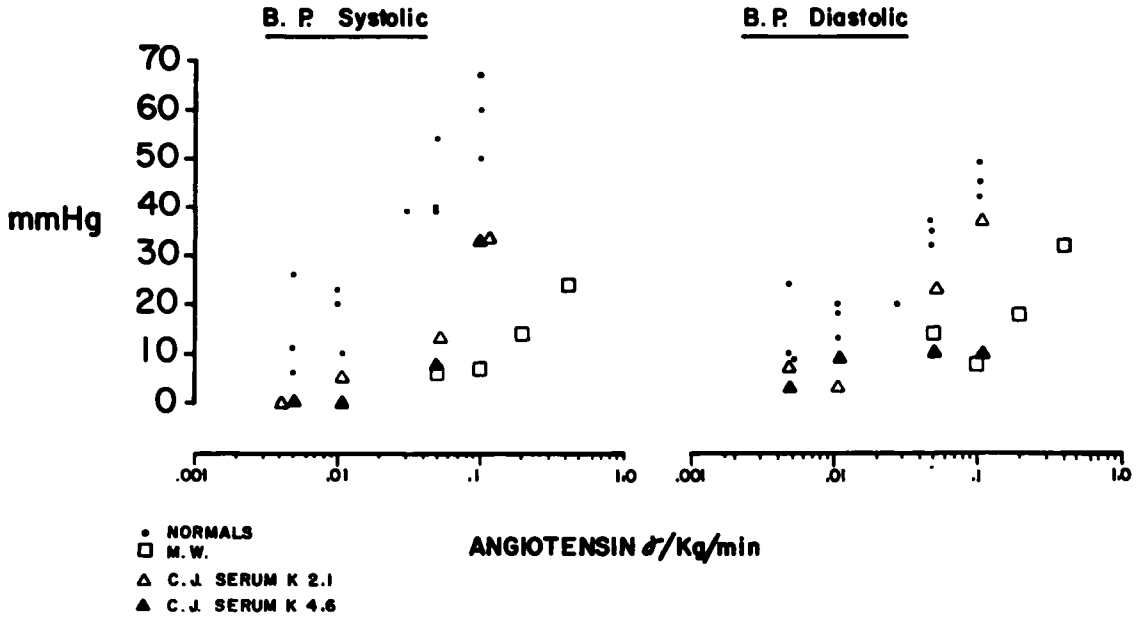

FIG. 8. Effects of angiotensin infused intravenously on systolic and diastolic blood pressure in normal subjects and in C. J. and M. W. Results shown represent increments above control pressure. Note that C. J. and M. W. showed less response at all dose levels.

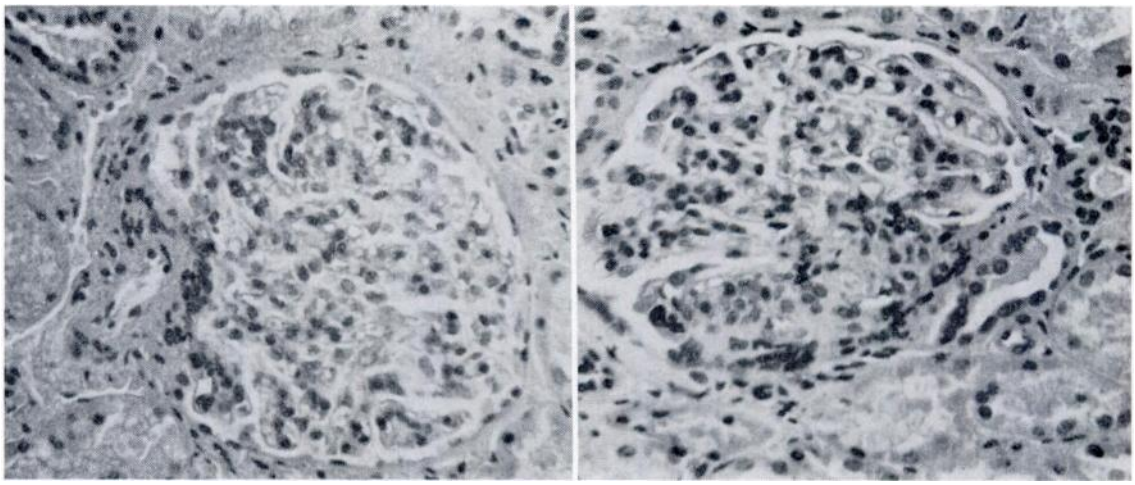

FIG. 9. Malpighian body of biopsy specimen of human kidney (C. J.) showing hyperplasia of juxtaglomerular collar of afferent arteriole. Regaud. Hematoxylin and eosin stain, original magnification × 210.

FIG. 10. Malpighian body (C. J.) showing enlargement of macula densa and hyperchromatism of portion of glomerular vessels (lower center). Regaud. Hematoxylin and eosin stain, original magnification × 210.

(Fig. 11), which may be reconstructed as follows; (1) hypertrophy of macula densa, (2) thickening of juxtaglomerular cells and hyperplasia of juxtaglomerular apparatus, (3) hyperchromatism of the vascular wall of part of the glomerulus, and (4) atrophy of the glomerulus.

Hypertrophy. The macula densa was larger than normal, containing about 30 to 40 nuclei arranged in two or three rows. Studied in serial sections, the macula densa constituted the canopy (the vascular roof) of the juxta-arteriolar section of the distal convoluted tubule (Fig. 10). The cytoplasmic mass of the macula densa was free of granules, but rich in filamentous and globular mitochondria.

Thickening. The muscular collar of the afferent arteriole was increased in thickness and length so that more of the muscular cells than normal appeared to have transformed into juxtaglomerular cells having large round nuclei in place of the original elongated nuclei typical of muscle fibers. Many of the juxtaglomerular apparatuses appeared to have two rows of juxtaglomerular (so-called glandibular) cells rather than one.

Hyperchromatism. The basement membrane of a portion of the wall of the afferent arteriole was thicker than normal, and this thickened membrane appeared to have involved about one-third to one-half of the glomerular capillaries, whereas in uninvolved glomeruli of the same kidney the glomerular vessels appeared to have little or no basement membrane. The hyperchromatism of the glomerular capillaries, which appeared to form a syncytium, was apparently a result of this thickening of the basement membrane. The abnormal syncytium and the thickened basement membrane involved the entire glomerulus in some instances. That the hyperplasia of the perivascular collar of the afferent arteriole (JG cells) also involved the vessels of the glomerular tuft is evident by the presence of juxtaglomerular granules (demonstrable by Bowie's strain) within the walls of the glomerular vessels.

Atrophy. Approximately 40 per cent of the glomeruli showed marked evidence of atrophy. The small atrophic glomerulus was always associated with a greatly enlarged juxtaglomerular apparatus (Fig. 12). Although it is possible that the atrophy of the glomerulus may have been a result of its failure to grow and mature in the first place, the gradual involvement of the glomerulus by the thickening basement membrane, and the alteration of the endothelial cells appear to show a relationship between dysfunction of the glomerulus on the one hand and the apparent atrophy on the other.

Patient M. W.

A percutaneous needle biopsy specimen of the right kidney was obtained. It was less than 1 mm. in width and 3 mm. in length, and contained 12 glomeruli. It was put in Zenker-formalin (Maximow's 10 per cent). The sections were colored by the periodic acid-Schiff method and by Bowie's strain for granules of the juxtaglomerular apparatus and by Altmann's acid-fuchsin methyl green for batonets and general cytoplasmic preservation.

Most of the glomeruli were about twice the size of normal, and approximately half of the juxtaglomerular apparatuses were hyperplastic. (In some of the sections seven of twelve, and in others, five of ten were hyperplastic.) One juxtaglomerular apparatus involved the entire hilar portion of the glomerulus,

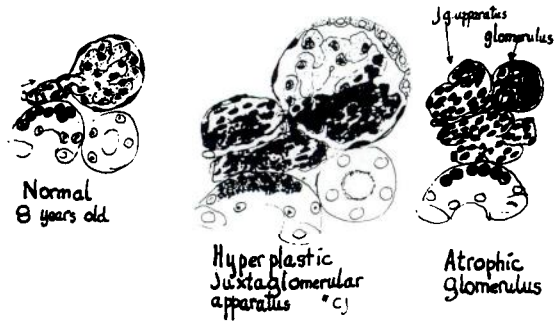

FIG. 11. Freehand drawings of different types of hyperplastic juxtaglomerular apparatuses of the patient (C. J.) compared with the normal apparatus to show the possible stages of development of the lesion.

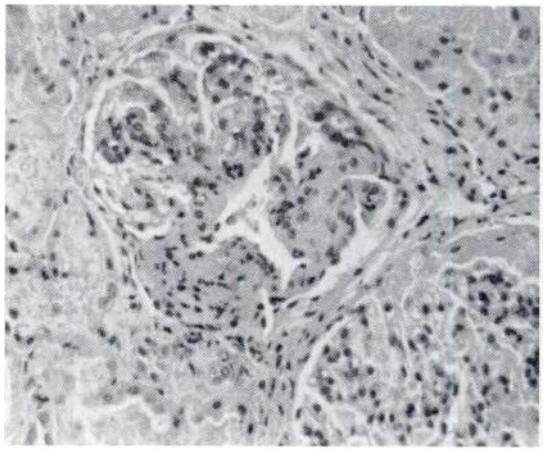

FIG. 12. Partially atrophic Malpighian body (C. J.) with extremely hyperplastic juxtaglomerular apparatus which was larger than the glomerular body itself. Compare with Figure 22. The glomerulus appeared to be a remnant of an immature glomerulus. Another large glomerular body may be seen below. Regaud. Hematoxylin and eosin stain, original magnification × 210.

and three involved about a third of the glomerulus. Four of the hyperplastic juxtaglomerular apparatuses contained granules. In one, the macula densa was about three times larger than normal; its multinucleated mass extended over the entire surface of the hyperplastic juxtaglomerular apparatus, forming a sheath of densely packed nuclei. There was a thin basement membrane between the macula densa and the hyperplastic juxtaglomerular apparatus. Two glomeruli were atrophic (Fig. 13 and 14).

COMMENTS

There is one striking clinical difference between these two patients: one (C. J.) is clearly dwarfed, whereas the other (M. W.), although he grew slowly, is of normal stature. It may be that potassium depletion occurred earlier and was more severe in C. J., and that this in turn led to dwarfism, but such a sequence of events cannot be established from the available evidence. Despite this important difference, the mass of evidence suggests that both patients suffered from essentially the same syndrome.

The clinical abnormalities in patient M. W. closely resembled those in C. J. in four important aspects. Both had

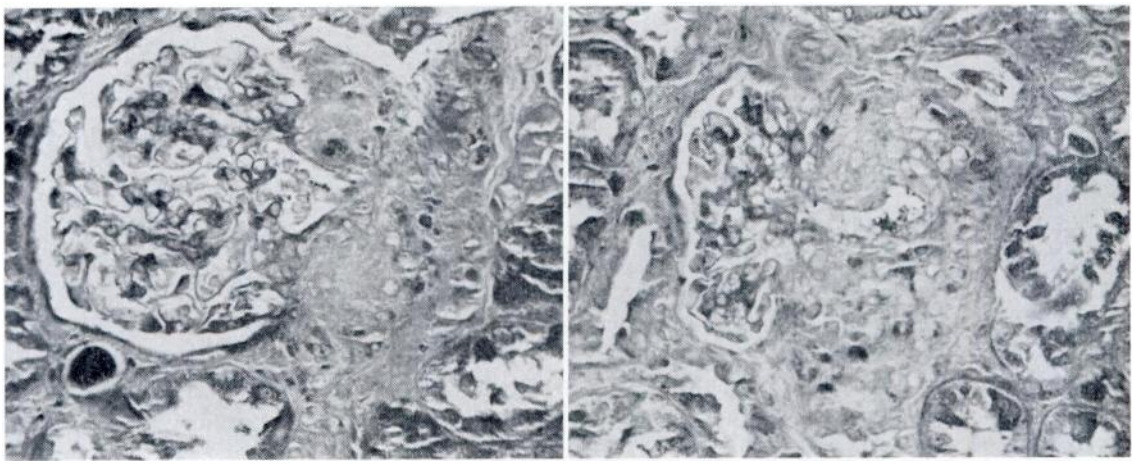

FIG. 13. Malpighian body (M. W.) showing hyperplastic juxtaglomerular apparatus. Bowie stain. original magnification × 430.

FIG. 14. Malpighian body (M. W.) showing hyperplasia and marked hypergranulation of juxtaglomerular apparatus and atrophy of glomerulus. Bowie stain, original magnification × 430.

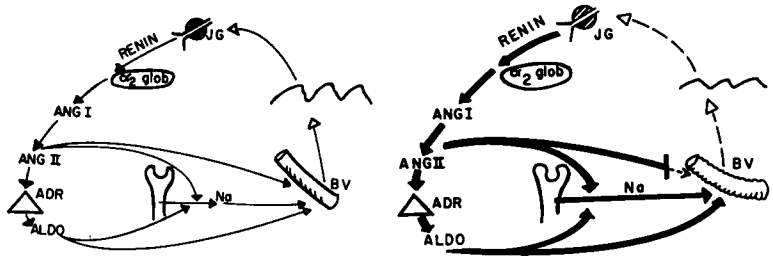

FIG. 15. A, schema representing the physiologic role of the renin-angiotensin system. Arrows with solid heads represent stimulation, those with hollow heads, inhibition. Renin is shown liberating angiotensin I from an α 2 globulin; three functions are shown for angiotensin II, direct effect on blood pressure, retention of sodium by an effect on the tubule (or glomerulus) of the kidney and stimulation of aldosterone secretion. Aldosterone is shown stimulating blood pressure directly, and via retention of sodium. Some function of blood pressure, or pulse pressure, is shown inhibiting renin production and thus operating a "feedback." B, schema to illustrate the defects in patients C. J. and M. W. A block in the ability of angiotensin II to elevate blood pressure results in decreased inhibition of renin production, increased production of renin, angiotensin I and angiotensin II, and in increase in aldosterone secretion. The blood levels of angiotensin II are elevated.

hypokalemic alkalosis, aldosteronism, normal blood pressure and a unique lesion of the juxtaglomerular apparatus. Both patients appeared to be mentally retarded. Whereas the results are equivocal in regards the effects of sodium deprivation, it appears likely (*vide infra*) that the responses of the two patients to this procedure are not qualitatively different.

Both patients showed persistent hypokalemic alkalosis; one (C. J.) showed depletion of muscle potassium on direct analysis. In both patients the development of hypokalemia was associated with urinary loss of potassium in excess of intake. Urinary potassium loss (*vide infra*) was decreased but not prevented by restriction of dietary sodium; in both patients it was prevented by infusion of human serum albumin (which lowered urinary sodium to zero) (Fig. 4 and 5), and by treatment with aldosterone antagonists. The excessive loss of potassium could thus be explained as accelerated distal tubular exchange of sodium for potassium under the influence of abnormally large amounts of aldosterone.

In both patients excretion of aldosterone was higher than that of normal subjects on comparable intakes of sodium; in C. J., it was shown to result not from tumor but from hyperplasia of the zona glomerulosa. The urinary aldosterone was not lowered by effective expansion of intravascular volume with human serum albumin; this is taken as evidence that the secretion of aldosterone in these patients is autonomous as regards hemodynamic factors [16]. (The *rise* in urinary aldosterone in C. J. with albumin may well be a result of the increase in body and serum potassium that it produced.)

In both patients the blood pressure was normal. It remained normal even when intravascular spaces were expanded with albumin, and thus the absence of hypertension cannot reasonably be attributed to hypovolemia. Hypertension might have been anticipated in these patients for two reasons. In the first place, they had aldosteronism: in the "classic" syndrome associated with adrenal cortical adenoma, hypertension is the rule, and is not prevented by potassium depletion comparable to that in our subjects. (Indeed, there is evidence other than that from this study to suggest that even in the "classic" syndrome the hypertension may not result from aldosterone alone. Aldosterone alone given to normal subjects even in large doses produces little or no hypertension [17,18], and patients with nephrosis and cirrhosis, secreting and excreting aldosterone in quantities larger than those reported for primary aldosteronism, generally have normal blood pressures.)

In the second place, both patients had hyperplasia and hypertrophy of the juxtaglomerular apparatus, histologic

lesions thus far unique. This suggests hypersecretion of renin, which should also lead to hypertension. In addition, both patients showed, on bioassay, elevated quantities of a pressor agent resembling angiotensin in the circulating blood. As will be discussed, the normal blood pressure suggests a pathophysiologic explanation for the syndrome.

There is little reason to suspect that aldosteronism or the resulting potassium depletion produced the renal lesion. Similar lesions have not been found in renal biopsy specimens from patients with primary or secondary aldosteronism [19,20] or in the kidneys of potassium-depleted rats [21]. Administration of aldosterone or desoxycorticosterone to rats or cats receiving diets of average or high sodium content results in a *decrease* of prominence of the juxtaglomerular apparatus and (*vide infra*) decrease of renin content of the kidney [22–26].

It is possible that the aldosteronism and the lesion of the juxtaglomerular apparatus both resulted from an unidentified common cause. There is, however, much to suggest that the renal lesion may have led to the adrenal one.

Extracts of renal tissue have been shown to increase the size of the zona glomerulosa in rats [27,28]. A considerable body of experimental evidence relates granulation of the juxtaglomerular apparatus, renal renin content and the action of sodium-retaining steroids. Thus, constriction of one renal artery leads to ipsilateral increase of juxtaglomerular granulation and renal renin content, and to contralateral decrease of both variables [26,29]. The zona glomerulosa of the adrenal cortex shows hypertrophy [27,30]. The decrease does not occur in the absence of the adrenals, but can be produced (even without constriction of the artery) by giving desoxycorticosterone and large quantities of sodium [23,24]. These findings are consistent with the hypothesis that the renin released from the ischemic kidney stimulates the adrenal cortex to release aldosterone, which in turn lowers the renin content of the contralateral kidney. This is supported by the observations (1) that renin and angiotensin II are potent stimuli to aldosterone secretion in the hypophysectomized nephrectomized dog [31–33], and (2) that angiotensin II stimulates secretion of aldosterone by slices of beef adrenals [34]. Angiotensin II has also been shown to stimulate aldosterone excretion [35] and secretion [36] in normal human subjects.

Whereas the aldosteronism may thus result from the lesion of the juxtaglomerular apparatus, we have no explanation for the lesion itself. The presence of atrophic glomeruli in C. J., if they are not in fact secondary to the juxtaglomerular lesion, suggests a congenital etiology for the renal disease, and thus, perhaps, for the juxtaglomerular hyperplasia. Elaut [37] observed hyperplasia of the afibrillar cells (juxtaglomerular apparatus of Goormaghtigh) in dogs made hypertensive by section of baroceptor nerves, and Goormaghtigh [38] found the juxtaglomerular apparatus to be hypertrophied in dogs made hypertensive by the Goldblatt technic, and he and Kaufmann [39] described hypertrophy and hyperplasia of the afibrillar cells associated with hypertension in man. These lesions, all of which occurred in hypertensive subjects, showed no resemblance to those in these two patients (C. J. and M. W.).

C. J. could effectively lower urinary sodium until it was no higher than the sodium intake (Fig. 1). Body weight was maintained. In M. W., however, urinary sodium continued to exceed sodium intake over a thirteen-day period (Fig. 2) of sodium deprivation. Despite these apparent differences, the results suggest that (1) the patients did not differ essentially as regards renal transport of sodium and (2) this was not normal in either patient. In C. J., sodium deprivation could not prevent the development of hypokalemia upon potassium restriction even when the urinary sodium had fallen to zero (Fig. 3). Even with a potassium intake of 170 mEq. per day, serum potassium rose only slightly in this patient during twelve days of sodium deprivation. This is an abnormal response. In patients with primary aldosteronism, restoration of body potassium is readily accomplished when the dietary, and thus urinary sodium, is rigorously restricted [40]. The response suggests that a disproportionate amount of the filtered sodium is reabsorbed at distal sites in C. J., and that excretion of potassium and hydrogen ions by cation exchange is thereby facilitated. In M. W., the continued urinary loss of sodium with restriction of dietary sodium shown in Figure 2 was associated with a marked increase of dietary potassium, a procedure required for clinical reasons. In this study (Fig. 3) body weight did not continue to fall despite the continued loss of sodium, and serum sodium concentration actually rose during the last eight days of the study. The serum potassium rose by only 0.7 mEq. per L. despite a potassium intake of 200 mEq. for eight days. Accordingly, it is reasonable to attribute the continued urinary sodium loss to replacement of intracellular sodium with potassium. This is supported by the observation (Fig. 3) that urinary potassium remained substantially below the potassium intake throughout the studies. An "obligatory" renal sodium-losing lesion is ruled out by the finding that this patient could lower urinary sodium to zero (Fig. 5).

Renin splits angiotensin I from a circulating $\alpha 2$ globulin, and this is further split by circulating converting enzyme to angiotensin II, which elevates the blood pressure. These relationships are shown diagrammatically in Figure 15A. If the kidney in our patients were indeed producing excessive quantities of renin, as suggested from the histologic appearance of the juxtaglomerular apparatuses, there must be a block in the production or effectiveness of one or more of these agents to explain the absence of hypertension. Renin-containing extracts of human kidney were shown to induce hypertension in both patients. It was thus clear that there was not a complete block at any point in the sequence. The magnitude of the response, however (Fig. 8), supported the proposition that a partial block did indeed exist. The response to angiotensin I again indicated the presence of converting enzyme in both subjects. Finally, hypertension developed in both patients with angiotensin II. However, the response in both patients was quantitatively clearly less than that of normal subjects. The initial response of systolic pressure was less than that obtained in the least responsive of the normal subjects studied by Bock and associates [41], and by ourselves [42] (see Fig. 8); moreover, with continued infusion the pressure fell in M. W. to much lower figures: a fall in pressure does not occur with continued infusions of comparable duration in normal subjects. Although the blood pressure response may indeed have been affected by potassium deficiency, which has been reported to lower the pressor response to angiotensin in rats [43]. This is unlikely, since the response in C. J. was no greater after serum potassium had been returned to normal.

It is not clear what controls the production of renin by the kidney, or whether it may be released except under extreme conditions involving renal ischemia. Some results suggest, however, that the renal arterial blood pressure or pulse pressure may serve to control renin secretion [44]. Whereas available evidence does not allow a definitive explanation for the syndrome in these patients, a hypothetical sequence of events may be suggested as a working hypothesis which fits best with the facts at hand. We suggest that these patients C. J. and M. W. may have, for reasons unknown, a primary impairment of the vascular response to angiotensin. This might lead by a lack of inhibitory (pressure or pulse pressure) impulses to activation of the juxtaglomerular apparatuses, with increased production of renin, leading in turn to overproduction of angiotensin I and angiotensin II. This is shown diagrammatically in Figure 15B. Whereas the angiotensin II would be unable to induce hypertension because of the primary defect, it might still stimulate the adrenal cortex to overproduction of aldosterone. (Whereas angiotensin II stimulates secretion not only of aldosterone but also of hydrocortisone in the nephrectomized, hypophysectomized dog, a number of results suggest that it may stimulate secretion of aldosterone selectively in normal man [45,46]). In a patient with such a disease pattern, one should find overproduction of aldosterone, relatively independent of hemodynamic stimuli, and increased quantities of circulating angiotensin. Both were present in these patients.

SUMMARY

A new syndrome, characterized by hypertrophy and hyperplasia of the juxtaglomerular apparatus of the kidneys, aldosteronism resulting from adrenal cortical hyperplasia, and persistently normal blood pressure is described in two patients. Overproduction of aldosterone could not be prevented by sodium loading or by administration of albumin intravenously; it was associated with hypokalemic alkalosis and Pitressin-resistant impairment of urinary concentrating ability. In both subjects, increased amounts of circulating angiotensin were demonstrated; infusion of angiotensin II produced rises of blood pressure in both subjects considerably less than the rises induced by comparable doses in normal subjects.

The sequence of events, (1) primary resistance to the pressor action of angiotensin, (2) compensatory overproduction of renin and thus of angiotensin, and (3) stimulation of adrenal cortex by angiotensin is consistent with all the information available about the syndrome.

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