SUMMARY  To generate quantitative data relating to the hypertensive activity of aldosterone, 9 μg/kg per day (4 times normal) aldosterone (4-ALDO) were infused chronically in both adrenalectomized and intact dogs until steady state conditions were achieved. Mean arterial pressure (MAP) was monitored continuously, 24 hours per day, and daily steady state values for MAP based on approximately 600 sample points per day were determined by employing computerized data analysis. In some studies, angiotensin II (A II) was also infused chronically (5 ng/kg per min) prior to and during 4-ALDO administration to maintain plasma levels of A II constant. 4-ALDO infusion in intact dogs maintained on 75 mEq sodium per day increased MAP by only 13 mm Hg compared to a greater than 30 mm Hg rise observed with A II infusion alone, even though plasma aldosterone concentration rose in these experiments only two-thirds as much as when 4-ALDO was infused. When A II was infused chronically, a fall in plasma A II concentration could not compensate for the hypertensive effects of superimposed 4-ALDO infusion; therefore, maximal aldosterone-induced hypertension was expected. However, in both adrenalectomized and intact dogs, the further addition of 4-ALDO failed to increase the degree of A II-induced hypertension and failed to promote sodium retention. Chronic A II infusion in intact dogs maintained on 75 and 190 mEq sodium per day produced a sustained increase in plasma aldosterone concentration (2.7 and 1.6 times control, respectively) as well as kaliuresis and hypokalemia. Infusion of A II in adrenalectomized dogs produced hyperkalemia, not hypokalemia. The data indicate that high plasma levels of aldosterone, at least over a period of several weeks, have only weak hypertensive effects in the dog, particularly in instances in which the aldosteronism is associated with a primary increase in A II.

ALTHOUGH aldosterone is considered by many to have an important role in the genesis and maintenance of hypertension associated with increased activity of the renin-angiotensin-aldosterone system, in fact, there are few quantitative data to distinguish the suspected hypertensive effects of the aldosterone from that of simultaneously formed angiotensin. The best example of the blood pressure-elevating effect of aldosterone is the syndrome of primary aldosteronism. Patients with this syndrome have hyperaldosteronemia and mild-to-severe hypertension. However, in both human subjects and experimental animals, chronic infusions of aldosterone which produce strong mineralocorticoid effects have been reported to produce little or no increase in arterial pressure. This is in contrast to the aldosteronism and severe hypertension which accompany chronic angiotensin II (A II) infusion in experimental animals, or the aldosteronism and severe hypertension observed in patients with primary reninism. Some of the factors that must be considered as possible contributors to the reported variability in the long-term blood pressure response to aldosterone include: (1) the plasma level of aldosterone, (2) the presence of other hypertensive agents such as A II, (3) sodium and potassium intake, (4) the presence or absence of steady state conditions, (5) the animal species studied, and (6) particularly in chronic studies or disease, whether or not there are aldosterone-induced renal vascular lesions.

In assessing quantitatively the effects of aldosterone on arterial pressure, an especially important consideration is the level of activity of the renin-angiotensin system. It is well known that plasma renin activity (PRA) decreases with aldosterone treatment, and in patients with primary aldosteronism, PRA is suppressed to low or undetectable levels. This suppression of the renin-angiotensin system might provide an important compensatory mechanism which attenuates or limits mineralocorticoid hypertension. One approach to answer this question would be to study the effects of aldosterone on arterial pressure under conditions in which plasma levels of A II are held constant, such as by infusion of A II. Additionally, this approach of fixing plasma levels of A II while varying plasma aldosterone concentration by infusion lends itself to evaluation of the relative quantitative importance of aldosterone vs. A II in the maintenance of experimental hypertension associated with A II infusion and hypertension observed clinically in conditions such as primary reninism. Finally, although circumstantial to the present study, prolonged infusions of A II under different experimental conditions might provide further insight into the role of the renin-
angiotensin system in the long-term control of aldosterone secretion, particularly since it has been reported that chronic infusion of A II fails to produce a sustained increase in plasma aldosterone concentration in sodium-replete dogs.11

Therefore, to generate quantitative data relating to the effects of excess aldosterone on arterial pressure, we infused aldosterone chronically into both adrenalectomized and intact dogs. Also, high levels of aldosterone were administered both in normal dogs and in dogs receiving A II given to suppress PRA and thus prevent changes in plasma levels of A II during the aldosterone infusion. It was reasoned that maximal aldosterone-induced hypertension might be observed under these conditions where the renin-angiotensin feedback suppression was prevented.

**Methods**

Ten male dogs weighing an average of 20.3 ± 1.0 (se) kg were employed in this study. Chronic indwelling polyvinyl femoral artery and vein catheters were implanted surgically for continuous 24-hour measurement of MAP and infusion, respectively. Five of the 10 dogs were bilaterally adrenalectomized and maintained postoperatively until experimentation on methylprednisolone acetate (Depo-Medrol, Upjohn) and deoxycorticosterone acetate (DOCA, Organon), as previously described.12

Approximately 2 weeks after surgery, the dogs were placed in metabolic pens and fitted with a plaster of Paris backpack housing a pressure transducer at heart level. The dogs had relatively unrestricted movement about the pen and, after a few days, appeared quite relaxed in the apparatus. The dogs were given free access to water and maintained on a fixed daily diet of two 15.5-oz. cans of h/d and no supplemental NaCl. Two cans of h/d provide <5 mEq sodium and 45-50 mEq potassium.

During the entire experimental period, mean arterial blood pressure was recorded continuously, 24 hours per day, by employing a Grass polygraph (model 7D) and Statham arterial blood pressure transducers which were built into the backpacks.13 In addition, the dogs were continuously infused intravenously with 150-200 ml of sterile isotonic NaCl per day by means of a Sage tubing pump (model 375 A). When appropriate, daily hormonal supplements were added to the saline. A II (CBA) was prepared fresh daily in saline, methylprednisolone sodium succinate (Solu-Medrol, Upjohn) every other day in the Mix-o-vial (Upjohn), and d-aldoesterone (Ciba) monthly in absolute pure ethyl alcohol. A Millipore filter was connected in series with the infusion line to prevent passage of bacteria and other contaminants.

Infusions of saline, A II, and/or aldosterone were continued until the dogs achieved sodium and potassium balance and all measured variables were steady. To provide for accurate measurements of 24-hour urinary sodium and potassium excretion rates for the purpose of establishing steady state conditions, the urinary bladder was catheterized aseptically on the last few days of each different infusion period.

Body temperature was measured daily, and ampicillin and a trimethoprim-sulfamethoxazole combination (Septra, Burroughs Wellcome Co.) were given prophylactically.

**Specific Protocols**

Five-milliliter blood samples were taken periodically for measurement of plasma renin activity (PRA), plasma sodium and potassium concentration, and hematocrit. Also, an additional 7 ml of blood were withdrawn on the final day of each different infusion in four of the five intact dogs (series 2) for measurement of plasma aldosterone concentration. All blood samples were taken at 8-9 a.m., 18-20 hours after feeding. In series 2 (intact dogs), 24-hour urine collections were made daily, whereas, in series 1 (adrenalectomized dogs), urine was collected only during the last few days of the different infusion periods. Series 2 animals were also weighed at each steady state.

**Series 1: Adrenalectomized Dogs**

The five adrenalectomized dogs were maintained on two cans of h/d and no supplemental NaCl so that, including the 150- to 200-ml saline infusion per day, their daily sodium intake was 20-25 mEq. Throughout the entire experimental period, the dogs were infused with methylprednisolone, 1.5 mg/day. This dose of synthetic glucocorticoid has essentially no mineralocorticoid activity and is adequate to maintain the dogs in good health. During the control period of approximately 2 weeks, 2.1-2.4 μg/kg per day of aldosterone (ALDO) were required to achieve sodium and potassium balance and maintain plasma potassium concentration at about 4 mEq/liter. Following the control period, A II (5 ng/kg per min) was then infused for 2 weeks, along with ALDO, and a steady state was established. Subsequently, the infusion rate of ALDO was increased 4-fold (4-ALDO) while the rate of A II infusion remained unchanged, and the combination of 4-ALDO + A II was infused for 2 more weeks at which time steady state conditions had once again been achieved. Finally, A II was discontinued and the 4-ALDO infusion reduced back to normal ALDO infusion until recovery measurements were steady approximately 2 weeks later.

**Series 2: Intact Dogs**

The five dogs with intact adrenal glands were studied while maintained on daily dietary sodium intakes of both 75 and 190 mEq. At each sodium intake, all variables were measured under steady
state conditions following infusion of saline, A II at 5 ng/kg per min, aldosterone at 9 μg/kg per day (4-ALDO), and A II + 4-ALDO. Each different infusion was continued for 6–7 days, at the end of which time steady state conditions were observed. The sequence of infusions was as follows (infusions at 190 mEq sodium intake are italicized): (1) saline, (2) A II, (3) A II + 4-ALDO, (4) A II, (5) saline, (6) 4-ALDO, (7) 4-ALDO, (8) saline, (9) saline, (10) A II, (11) A II + 4-ALDO, (12) A II + 4-ALDO + KCl (diet supplemented with 150 mEq KCl, (13) saline.

Analytical Methods

PRA was measured by a radioimmunoassay procedure for angiotensin I (A I) (E. R. Squibb & Sons) and is expressed as nanograms of angiotensin I generated per milliliter of plasma per hour incubation (ng A I/ml per hr). Plasma aldosterone concentration was determined by the radioimmunoassay method of Bühler et al. Plasma and urine concentrations of sodium and potassium were determined by flame photometry (Instrumentation Laboratory, IL 343).

The automated techniques used in our laboratory for computerized data analysis have been previously described in detail. In brief, the continuous mean arterial blood pressure recordings from the Grass polygraph were converted to electrical signals for computer analysis by employing a four-channel analog curve-reading system with fiber optic scanning pens which generated analog voltages from the recorded ink tracings. The analog voltages from the curve tracer were fed into an analog-to-digital converter that changed them to digital signals for analysis by a PDP 11/70 computer. The digitized information was used by the computer to calculate hourly and daily values for mean arterial pressure based on approximately 30 sample points per hour.

All values presented are means ± SE. Student's t-test for paired observations was used to determine statistical significance. Statistical significance was considered to be $P < 0.05$.

Results

Series I: Adrenalectomized Dogs

The steady state values of the measured variables for the five adrenalectomized dogs studied on 25 mEq sodium/day are presented in Figure 1 and Table 1. The dogs were not considered to be in a steady state unless they were in sodium and potassium balance and all measured variables were steady for the last 3 consecutive days of each infusion period. That the basal maintenance doses of adrenal steroid hormones were appropriate is corroborated by the normal control values for concentrations of plasma electrolytes, mean arterial pressure, and PRA.

As expected, PRA fell to undetectable levels with A II infusion. Control mean arterial pressure averaged 101 ± 1 mm Hg and increased to a maximal level of 118 ± 5 mm Hg by day 3 and then waned over the next 48 hours. Thereafter, the 24-hour mean arterial pressure values were virtually constant at 107 ± 2 mm Hg from day 5 through day 14 of A II infusion.

Mean plasma potassium concentration increased significantly with A II infusion by as much as 0.5 mEq/liter by day 5; however, the mean elevation in plasma potassium concentration by day 14 of A II infusion was not statistically significant.

On day 14, when all measured variables were steady, the infusion rate of aldosterone was then increased to 4 times normal (4-ALDO) and infusion of both A II and 4-ALDO was maintained for another 14 days. The most prominent change and the only change in steady state values that was statistically significant was plasma potassium concentration which fell by an average of 1.3 ± 0.3 mEq/liter. Most important to this study, however, was the fact that there was no statistically significant change in the steady state value for mean arterial pressure with 4-ALDO administration; i.e., increasing the aldosterone infusion rate to 4 times normal actually reduced arterial pressure by an average of 2 mm Hg and this was not a statistically significant change. Thus, the high infusion rate of aldosterone did not influence the arterial pressure rise induced by A II infusion.
TABLE 1  Steady State Values of Variables for Adrenalectomized Dogs (Series 1)

<table>
<thead>
<tr>
<th></th>
<th>P_N. (mEq/liter)</th>
<th>P_K. (mEq/liter)</th>
<th>HCT (%)</th>
<th>U_N.V (mEq/day)</th>
<th>U_K.V (mEq/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>147 ± 1</td>
<td>4.2 ± 0.1</td>
<td>35 ± 1</td>
<td>23 ± 4</td>
<td>48 ± 2</td>
</tr>
<tr>
<td>All</td>
<td>145 ± 1</td>
<td>4.3 ± 0.1</td>
<td>37 ± 1</td>
<td>26 ± 4</td>
<td>42 ± 4</td>
</tr>
<tr>
<td>All + 4-ALDO</td>
<td>147 ± 1</td>
<td>3.0 ± 0.3*</td>
<td>26 ± 3</td>
<td>45 ± 3</td>
<td></td>
</tr>
<tr>
<td>Recovery</td>
<td>146 ± 1</td>
<td>4.2 ± 0.1</td>
<td>35 ± 1</td>
<td>22 ± 3</td>
<td>50 ± 6</td>
</tr>
</tbody>
</table>

Values are means ± SE. Abbreviations: All, angiotensin II (5 ng/kg per min); 4-ALDO, aldosterone (8.4-9.6 µg/kg per day); P_N., plasma sodium concentration; P_K, plasma potassium concentration; U_N.V, urinary sodium excretion; U_K.V, urinary potassium excretion.* P < 0.05 vs. control.

Within 12 days after cessation of A II + 4-ALDO, all variables returned to their control values.

Series 2: Intact Dogs

Figures 2-7 illustrate the data for three consecutive experiments performed in the five intact dogs.

Experiment 1: dietary sodium intake = 75 mEq/day; successive 6- to 7-day infusions of (1) saline, (2) A II, (3) A II + 4-ALDO, (4) A II, and (5) saline.

Figures 2 and 3 show the results obtained from intact dogs subjected to a protocol similar to that described for the adrenalectomized dogs. In contrast to the adrenalectomized dogs, there was a typical arterial blood pressure response to A II infusion: arterial blood pressure increased progressively for several days from a control value of 110 ± 4 mm Hg and plateaued on days 4-5 of A II infusion (at 142 ± 4 mm Hg). Although not measured in this study, we have observed many times in our laboratory that the degree of hypertension achieved by days 4-5 of A II infusion persists for up to weeks of A II administration (unpublished observations). Most significantly, however, as in the adrenalectomized dogs, there was no further significant change in MAP with subsequent infusion of large amounts of 4-ALDO. Finally, the recovery steady state value for mean arterial blood pressure measured 7 days after cessation of all hormonal infusions was not significantly different from control.

With A II infusion, there was a sustained 2- to 3-fold increase in plasma aldosterone concentration. The mean steady state values for plasma aldosterone concentration for the control period and for...
days 5-6 and for day 19 of A II infusion were 3.7 ± 0.8, 9.9 ± 1.3, and 8.5 ± 1.0 ng percent, respectively. The mean steady state value for plasma aldosterone concentration with infusion of exogenous aldosterone (A II + 4-ALDO) was 23.5 ± 3.9 or 6.4 times control.

In contrast to the adrenalectomized dogs in which plasma potassium concentration actually increased with A II infusion, in the intact dogs plasma potassium concentration decreased significantly with A II infusion and by day 5 was depressed by an average of 0.4 ± 0.1 mEq/liter (Fig. 2). With subsequent infusion of 4-ALDO in addition to the A II, mean plasma potassium concentration fell on additional 0.6 ± 0.2 mEq/liter. The hypokalemia observed during both A II and A II + 4-ALDO infusion was associated with a kaliuresis. During the periods of A II and A II + 4-ALDO infusion, potassium excretion exceeded potassium intake by a total of 24 ± 3 and 36 ± 12 mEq, respectively. During the subsequent periods of A II and saline infusion, urinary potassium excretion was equal to that observed during the control period. Mean plasma potassium concentration during the recovery period was elevated by 0.3 ± 0.1 mEq/liter, although this was not statistically significant.

During A II infusion, there was a significant net retention of sodium which averaged 64 ± 19 mEq (Fig. 3). During this period of sodium retention, hematocrit fell significantly although body weight was unchanged. With subsequent infusion of high amounts of aldosterone in addition to the A II, there was no further retention of sodium; in fact, during the total 7-day period of A II + 4-ALDO infusion, the dogs actually were in negative sodium balance by an average of 8 ± 6 mEq, although this was not statistically significant. Simultaneously, body weight was unaltered. The sodium retention depicted for day 7 in Figure 3 may have been more apparent than real, since the urinary bladder was not catheterized on this day. Sodium balance and body weight remained constant during days 14-19 of A II infusion; however, body weight increased significantly by an average of 0.5 ± 0.1 kg by the end of the recovery period although there was no further retention of sodium.

Finally, as expected, PRA was suppressed to undetectable levels during A II infusion and remained suppressed during the entire experiment. Unexpectedly, however, PRA did not return to control by the end of the recovery period.

Experiment 2: dietary sodium = 75 and 190 mEq/day; successive 6- to 7-day infusions of (1) saline, (2) 4-ALDO, and (3) saline.

The data for experiment 2 are presented in Figures 4 and 5. In this experiment, the same high level of aldosterone (9 µg/kg per day) was infused but in the absence of background infusion of A II. The increase in the steady state value for plasma aldosterone concentration to 16.4 ± 2.1 ng percent (3.7 times control) with 4-ALDO administration was significantly less than the increase observed in experiment 1 (6.4 times control) in which A II and 4-ALDO were infused simultaneously.

The steady state values presented for the control period in experiment 2 (Figs. 4 and 5) are the same values presented for the recovery period of experiment 1 (Figs. 2 and 3) and, as mentioned above, PRA had not yet returned to normal levels. That PRA was in fact depressed at this time was a fortuitous occurrence because it provided yet another experimental condition to observe the hypertensive potency of aldosterone independent of compensatory changes in PRA. Nonetheless, in spite of the fact that maximal hypertensive activity of aldosterone was expected under these conditions, there was only a 13 mm Hg increase in mean arterial pressure with 4-ALDO, compared to the greater than 30 mm Hg rise observed with A II infusion alone in experiment 1. Increasing the sodium intake to high levels (190 mEq/day) resulted in a further increase in mean arterial pressure of only 6 mm Hg.

As in experiment 1, 4-ALDO treatment produced a dramatic fall in plasma potassium concentration and, in addition, the aldosterone-induced hypokalemia was even more severe in the presence of higher dietary sodium (190 mEq/day). In spite of
INTACT DOGS (N=5)

No intake:
• 75 mEq/day
• 190 mEq/day

MEAN ARTERIAL PRESSURE (mmHg)

URINARY SODIUM EXCRETION (mEq/day)

PLASMA SODIUM CONC. (mEq/l)

HEMATOCRIT (%)

DAYS

FIGURE 5 Effects of chronic infusion of high levels of aldosterone (4 times normal) on mean arterial pressure, 24-hour urinary sodium excretion, plasma sodium concentration, and hematocrit in intact dogs maintained on 75 and 190 mEq sodium/day.

the fact that there was a significantly smaller increase in plasma aldosterone concentration during the infusion of 4-ALDO in experiment 2 than in experiment 1, the average fall in plasma potassium concentration below control was significantly greater during 4-ALDO administration in experiment 2 (1.4 ± 0.1 mEq/liter) than in the previous experiment (1.1 ± 0.2 mEq/liter) in which A II was infused simultaneously with 4-ALDO. While the dogs were maintained on 75 and 190 mEq sodium/day and infused with 4-ALDO, the net potassium deficits for each period were 44 ± 16 and 72 ± 14 mEq, respectively.

In contrast to the sodium excretory response to infusion of 4-ALDO in the presence of high circulating levels of A II (experiment 1), 4-ALDO infusion in experiment 2 produced a typical aldosterone response: sodium retention for several days followed by "sodium escape." For the 7 days the dogs were maintained on 75 mEq sodium/day and infused with 4-ALDO, there was an average net retention of 49 ± 16 mEq sodium, which was statistically significant. Sodium and fluid retention with 4-ALDO treatment is reflected by the significant fall in hematocrit and increase in body weight (0.6 ± 0.2 kg) which was not observed in experiment 1 where 4-ALDO failed to produce sodium retention. When dietary sodium intake was increased to 190 mEq/day during 4-ALDO infusion, there was no further measurable sodium retention or increase in body weight. During the recovery period, sodium balance was maintained and body weight remained elevated at the 4-ALDO steady state level.

Again, PRA failed to return to normal levels during the recovery period, but the steady state values for all other variables were similar to and not significantly different from control.

Experiment 3: dietary sodium intake = 190 mEq/day; successive 6- to 7-day infusions of (1) saline, (2) A II, (3) A II + 4-ALDO, (4) A II + 4-ALDO with high dietary potassium, and (5) saline. Much of the protocol for experiment 3 was the same as that for experiment 1, the notable exception being that the dogs were maintained on a higher sodium intake (Figs. 6 and 7). The steady state control values presented in Figures 6 and 7 are the recovery values from the previous experiment (Figs. 4 and 5).

The results of experiment 3 were similar to those observed in experiment 1, and most notably: (1) 4-ALDO infusion again failed to produce sodium retention in the presence of A II infusion (and body weight was stable), and, accordingly, the degree of A II-induced hypertension was not influenced by subsequent infusion of large amounts of aldosterone. (2) Plasma potassium concentration decreased...
and urinary potassium excretion increased with A II infusion; however, on this higher dietary sodium intake, these changes were significantly greater than in experiment 1. Mean plasma potassium concentration decreased by 0.8 ± 0.2 mEq/liter and urinary potassium excretion exceeded intake by 81 ± 9 mEq (in experiment 1 these changes were 0.4 ± 0.1 mEq/liter and 24 ± 3 mEq, respectively). As in experiment 1, with subsequent infusion of 4-ALDO, the changes in plasma potassium concentration and urinary potassium excretion were even more exaggerated than with A II infusion alone, and, in addition, exceeded the corresponding effects produced in experiment 1. (3) Finally, A II infusion produced sustained increases in plasma aldosterone concentration in all but one dog in which plasma aldosterone concentration failed to increase; due to the response in this one dog, the average increase in plasma aldosterone concentration (63 ± 68% above control) was not statistically significant. The increases in aldosterone concentration with A II infusion were consistently smaller than those observed when the dogs were maintained on 75 mEq solution/day (experiment 1).

In the latter part of experiment 3, dietary potassium intake was increased 4-fold during A II + 4-ALDO infusion, and in spite of achieving partial restitution of plasma potassium concentration to control levels, there was no significant change in the steady state value for mean arterial pressure.

During this 7-day period of potassium supplementation, there was a net sodium deficit averaging 60 ± 15 mEq. On the final day of the infusions, a blood sample was taken from each dog for determination of complete blood chemistry by the University Chemical Laboratory. All 24 measured variables were within normal limits in all dogs.

**Discussion**

Attempts to produce hypertension by chronic administration of supranormal amounts of aldosterone in laboratory animals other than the salt-loaded rat have been relatively unsuccessful in spite of the fact that marked changes in electrolyte balance have been achieved. Similarly, human subjects treated with doses of aldosterone which produce strong electrolyte effects have either developed mild hypertension or remained normotensive. In the present study in dogs, we infused aldosterone continuously at approximately 4 times the endogenous secretion rate of the mineralocorticoid and measured changes in arterial pressure based upon approximately 800 data points per day. Mean arterial pressure increased by only 13 mm Hg with 4-ALDO infusion in dogs maintained on a liberal sodium intake, 75 mEq sodium per day (Figs. 4 and 5); in comparison, infusion of A II at a dose which would be expected to increase the plasma concentration of A II to only 2-3 times normal produced a steady state elevation in mean arterial pressure of greater than 30 mm Hg (Figs. 2 and 3). Further, since increasing the sodium intake from 75 to 190 mEq per day during 4-ALDO infusion produced a further increase in mean arterial pressure of only 6 mm Hg (Figs. 4 and 5), it is apparent that aldosterone-induced hypertension in the dog is fairly salt-insensitive at higher sodium intakes.

Since hypertension is present in patients with primary aldosteronism and since plasma aldosterone concentration also is elevated during chronic A II infusion and in primary reninism, it has been suspected that aldosterone contributes significantly to the hypertension observed in these high A II states. However, an interesting and consistent observation in this present study was the failure of high amounts of aldosterone to produce sodium retention or influence the magnitude of the A II-induced hypertension. This would indicate that the moderate increases in plasma aldosterone concentration which occur with A II infusion or in primary reninism probably have little influence on the severity of the hypertension, at least within an expance of time (a few weeks) in which there are no aldosterone-induced vascular or renal structural changes. The hypertension seems to be more a function of the peripheral vasoconstrictor effects of A II and the renal effects of A II which promote sodium retention.

In contrast to the data reported by McCaa et al., in the present study, chronic infusion of A II
in sodium-replete dogs produced a sustained increase in plasma aldosterone concentration in seven of eight observations. Further, in every instance there was a greater steady state increase in plasma aldosterone concentration during A II infusion when the dogs were maintained on 75 vs. 190 mEq sodium/day. In addition, in every intact animal, hypokalemia and kaliuresis accompanied A II infusion, and both were more severe at the higher sodium intake (Fig. 6) than at 75 mEq sodium/day (Fig. 2) when the aldosterone response was greater. In light of the well established potent effects of potassium metabolism on aldosterone secretion, the present data are consistent with the notion that at higher sodium intakes the increasingly greater degree of potassium depletion which accompanies A II infusion has a greater mitigating influence on the aldosterone response to A II infusion. The greater aldosterone response to A II infusion in experiment 1 (Fig. 2) may also have resulted from alterations in adrenocortical sensitivity unrelated to potassium metabolism, since the sensitivity of the zona glomerulosa to acute infusions of A II is enhanced at lower sodium intakes. Since chronic administration of renin or A II has a sensitizing effect on the adrenocortical zona glomerulosa, it is presumed that the renin-angiotensin system mediates changes in adrenocortical sensitivity during alterations in sodium intake, although other factors apparently are involved also. However, if changes in adrenocortical sensitivity do in fact account for any part of the differential aldosterone response observed in the present study, one might expect the sensitivity changes not to be mediated by the renin-angiotensin system since this feedback loop was fixed at the same level in both experiments by 7 days of A II infusion. On the other hand, the prolonged suppression of the renin-angiotensin system prior to infusion of A II in experiment 3 (Fig. 6) may have contributed to the smaller aldosterone response observed after 7 days of A II infusion at this higher level of sodium intake.

The present study also provides information regarding the differential effects of A II and aldosterone on potassium metabolism. In the adrenalectomized dogs, A II infusion produced hyperkalemia for several days and, although the hyperkalemia did not persist at a statistically elevated level, plasma potassium concentration did not fall with A II infusion as it did in the dogs with intact adrenal glands. In addition, in the intact dogs, the hypokalemia and negative potassium balance that accompanied aldosterone infusion were smaller at each level of sodium intake when A II was infused simultaneously with aldosterone (A II + 4-ALDO) than when aldosterone was infused alone (4-ALDO), in spite of the fact that plasma aldosterone concentration reached a higher level under the former conditions. The data, therefore, are consistent with that generated from acute intrarenal infusions of A II or A II antagonist, demonstrating that A II acts intrarenally to promote potassium retention. Thus, the hypokalemia and kaliuresis which accompany A II infusion in intact dogs, in contrast to the effect in adrenalectomized dogs, are likely secondary to aldosterone stimulation. Similarly, Urquhart et al. reported a kaliuresis and fall in potassium concentration in intact but not adrenalectomized dogs infused with A II. Finally, in accord with the observations of others, the mineralocorticoid-induced hypokalemia and potassium depletion observed with 4-ALDO or A II infusion were more severe at the higher level of sodium intake.

It has been reported that chronic potassium deficiency in both man and experimental animals is associated with an increase in PRA and several hemodynamic changes which may or may not include a reduction in arterial pressure. Therefore, since the effects of chronic potassium depletion on arterial pressure are still unresolved, we thought it of interest to determine whether the negative potassium balance which accompanies A II infusion and particularly aldosterone administration might have influence on the degree of hypertension achieved with these agents. The present data, however, do not indicate an effect. Although partial restitution of plasma potassium concentration to control levels was achieved with dietary potassium supplementation during A II + 4-ALDO infusion (Fig. 6), the severity of the hypertension was not affected.

In all three experiments in the intact dogs, the failure of PRA to return to the initial control level observed in experiment 1 (Fig. 2) during the week after cessation of hormonal infusions was unexpected but, in retrospect, not surprising. Although we usually observe restitution of PRA to control levels within 48 hours after terminating a chronic intravenous infusion of angiotensin II (unpublished observations), studies following removal of an aldosterone-secreting adenoma have shown that the return to normal of PRA, sodium and fluid balance, and aldosterone secretion may be delayed for days to weeks, and in the case of aldosterone secretion, even months. The time interval for return of PRA varies considerably from patient to patient and is probably influenced by the degree of initial renin suppression as well as by the level of sodium intake postoperatively. In the present experiment in intact dogs, PRA had been suppressed for several weeks during the infusion of both A II and 4-ALDO, and the liberal intakes of 75 and 190 mEq sodium/day which were maintained during the recovery periods apparently were sufficient to delay recovery of PRA. Indeed, during the 7-day recovery periods in all three experiments in the intact dogs, little or none of the sodium and water retained during the prior hormonal infusions was lost. Similarly, in spite of the fact that renin and aldosterone secretion rates are suppressed following
removal of an aldosterone-producing adenoma, during the first postoperative week natriuresis may be delayed and body weight and fluid balance maintained.29

The normalization of plasma aldosterone concentration during the recovery periods, in spite of suppressed PRA, may have been a function of plasma potassium concentration which increased modestly but consistently from recovery period to recovery period. Thus, although within normal limits, plasma aldosterone concentration during the recovery periods was inappropriately low for the concurrent concentration of plasma potassium.31 Hyperkalemia is also occasionally observed clinically during the period of suppressed aldosterone secretion which follows removal of an aldosterone-secreting tumor.29,32

In summary, increases in plasma aldosterone concentration comparable to those expected in many patients with primary aldosteronism16-18 were achieved by continuous infusion of exogenous aldosterone. Although the high infusion rate of aldosterone produced changes in sodium and potassium metabolism similar to those found in patients with primary aldosteronism, at best only slight hypertension was achieved (+13 mm Hg and +19 mm Hg at high and very high sodium intakes, respectively). Therefore, in the dog, the arterial blood pressure response to infusion of high levels of aldosterone is similar to that reported for human subjects subjected to prolonged intramuscular injections,5 or infusions of supranormal amounts of aldosterone. Thus, instances of severe hypertension in patients with primary aldosteronism may reflect more chronic effects of aldosterone not observed in the present study, such as the occurrence of renal vascular lesions.9 Accordingly, in spite of the fact that aldosterone secretion is normalized and arterial pressure reduced after removal of an aldosterone-secreting adenoma, it is not uncommon for hypertension to persist.5,4,31 Finally, the role of hyperaldosteronemia in the maintenance of hypertension associated with a primary increase in A II in the present experiments was insignificant indeed, and this may also apply to the secondary aldosteronism which is observed in the high renin states such as renovascular or malignant hypertension. In the present study, high levels of aldosterone failed to produce sodium retention or alter arterial blood pressure when hypertension was induced by A II infusion. Apparently, in hypertensive states associated with increased activity of the renin-angiotensin system, the ensuing hypertension is far more dependent on the renal sodium-retaining effects of A II than on those of aldosterone.

Acknowledgments

We are grateful to Dr. James O. Davis, Department of Physiology, The University of Missouri School of Medicine, for as-

saying the plasma for aldosterone concentration. Depo-Medrol and Solu-Medrol were generously supplied by Upjohn Company.

References

Adenosine Production in the Ischemic Kidney

WAYNE L. MILLER, ROSEMARY A. THOMAS, ROBERT M. BERNE, AND RAFAEL RUBIO

SUMMARY We conducted experiments to determine (1) tissue, blood, and urine levels of adenosine produced by the ischemic kidney under conditions of renal artery occlusion, and (2) the site(s) of production and release of adenosine by the kidney. Concentrations of adenosine, inosine, and hypoxanthine in the dog urine were found to increase after 2 minutes of renal artery occlusion as were concentrations of these metabolites in renal tissue after 10 minutes of renal artery occlusion. Renal venous plasma levels of inosine and hypoxanthine also were elevated after 3 minutes of arterial occlusion. In modified stop-flow experiments, adenosine appeared in the urine in a peak that corresponded most closely with proximal tubule flow. 5'-Nucleotidase, the enzyme which catalyzes the dephosphorylation of 5'-AMP or 5'-IMP to adenosine or inosine, respectively, was found to be located primarily on the external membranes and mitochondria of proximal tubule cells, but not in distal tubule or collecting duct cells. Since adenosine has been demonstrated to elicit renal vasoconstriction and is produced by the ischemic kidney, it is suggested that adenosine may be involved in the mediation of postocclusion renal ischemia.

ADENOSINE has been proposed as a mediator in the control of blood flow in heart muscle, skeletal muscle, and brain and has been shown to be formed by the dephosphorylation of AMP, a reaction catalyzed by the enzyme 5'-nucleotidase. 5'-Nucleotidase activity has been demonstrated in a variety of tissues, including isolated membranes from rat kidney, and indirect evidence suggests that adenosine may play a role in the regulation of renal blood flow. For example, in heart and skeletal muscle, adenine nucleotides and adenosine produce vasodilation; however, in the dog kidney, ATP and ADP elicit vasodilation, whereas AMP and adenosine elicit vasoconstriction. Furthermore, adenosine-induced renal vasoconstriction in the rat was observed after salt restriction and, hence, activation of the renin-angiotensin system, but not after salt loading. Considering these findings, it is of interest that Scott et al. observed that renal venous blood collected after release of occlusion of the renal artery produced dilation of the vessels of the perfused hindlimb but constriction of the vessels of the contralateral kidney. Also of interest are the observations that reactive ischemia occurred following a 2-minute period of renal artery occlusion in 52% of experiments completed on dogs and that AMP was found in the renal venous blood of ischemic kidneys. In addition, dipyridamole, a compound that inhibits adenosine uptake by red cells and tissues, greatly potentiated the vasoconstrictor response to bolus injections of adenosine and AMP, and also enhanced reactive ischemia in the dog kidney. Dipyridamole also has been shown to restore the renal autoregulatory response after spontaneous renal autoregulatory failure. Thus, it may be postulated that adenosine accumulation is partially responsible for the phenomenon of postocclusive renal reactive ischemia and that adenosine...
Failure of chronic aldosterone infusion to increase arterial pressure in dogs with angiotensin-induced hypertension.

T E Lohmeier, A W Cowley, Jr, J W DeClue and A C Guyton

doi: 10.1161/01.RES.43.3.381

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1978 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/43/3/381.citation

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Circulation Research_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Circulation Research_ is online at:
http://circres.ahajournals.org/subscriptions/