Steady State Aldosterone Dose-Response Relationships

DAVID B. YOUNG AND ARTHUR C. GUYTON

SUMMARY The steady state effects of different infusion rates of aldosterone on plasma concentrations of sodium and potassium, plasma renin activity, sodium space, and mean arterial pressure were determined. Measured amounts of aldosterone were infused continuously into adrenalectomized dogs for 13 weeks. Four rates of aldosterone administration were used: 16 ± 1 μg/day, 48 ± 3 μg/day (approximately the normal secretory rate for 22-kg dogs on a daily sodium intake of 27 mEq, potassium intake of 27 mEq), 91 ± 4 μg/day, and 219 ± 10 μg/day. Each rate of infusion was continued until the dogs were in sodium and potassium balance and measured variables were steady. Decreasing the rate of aldosterone administration below normal led to sharp decreases in plasma sodium concentration and sodium space, while raising the rate above normal had little effect. Plasma potassium concentration varied inversely and significantly with changes in aldosterone administration over the entire range of rates. Plasma renin activity rose extremely rapidly as the level of aldosterone fell below normal and went to zero at aldosterone infusion rates slightly above normal. Arterial pressure increased as aldosterone rose above normal but did not fall below normal at subnormal aldosterone levels, probably because of thepressor effects of simultaneously generated angiotensin II.

ALTHOUGH several decades of clinical observation and experimental investigation have proved qualitatively that aldosterone is an important participant in the control of extracellular electrolytes and volume, we have been unable to find in the literature data describing quantitatively the long-term dose-response effects of aldosterone. Therefore, the following study was undertaken to generate a series of steady state dose-response curves relating the rate of aldosterone infusion into adrenalectomized dogs with plasma concentrations of sodium and potassium, sodium space, mean arterial pressure, and plasma renin activity.

Methods

Long-term experiments were carried out in a group of five male mongrel dogs whose weights averaged 22.5 kg. Several weeks prior to the start of the study the dogs were adrenalectomized and cannulas were implanted into the aorta and vena cava through the femoral vessels. Following recovery a portable infusion pump designed in our laboratory was connected to the venous cannula, and aldosterone (CIBA) and methyl prednisolone (Solu-Medrol, Upjohn) were infused in distilled water continuously 24 hours/day, 7 days a week, for 13 weeks. The rate of flow from the pump was 15 ml/day throughout the study. The rate of aldosterone infusion was increased in four steps, each step lasting at least 2 weeks. Each infusion level was continued until the dogs had reached new sodium and potassium balances and all measured variables were steady. The first rate of aldosterone infusion was 16 ± 1 (SE) μg/day, followed successively by rates of 48 ± 3 μg/day, 91 ± 4 μg/day, and 219 ± 10 μg/day. One dog died after the completion of the second infusion level. At all four levels, 1 mg/day of methylprednisolone was infused along with aldosterone. This dose of synthetic glucocorticoid has essentially no mineralocorticoid effect and was adequate to keep the dogs in good health. Throughout the experiment the dogs were housed in metabolic cages and fed a diet containing 27 mEq/day each of sodium and potassium.

Between 9 a.m. and 12 noon each day during the experiment the dogs were brought into the laboratory in which they had been trained to lie quietly unrestrained. Arterial blood was drawn from the indwelling cannula into syringes containing several microliters of heparin (1,000 U/ml). Plasma for electrolyte analysis was obtained by immediately centrifuging the blood in a high speed centrifuge for a period of less than 5 minutes, then removing the plasma. All electrolyte determinations were made with an Instrumentation Laboratory model 343 flame photometer. For...
plasma measurements the flame photometer was calibrated using a standard solution with a viscosity of 1.8 centipoises (cp), the approximate viscosity of plasma. Plasma sodium and potassium concentrations were determined each day during the study; when these values reached a constant level and the dogs were in approximate sodium and potassium balance, the following measurements were made for each of the next 3 days: plasma and urine sodium-potassium concentration, plasma renin activity, mean arterial pressure, body weight, and on the last day of infusion of each level of aldosterone, \(^{22}\)Na space (used to estimate changes in extracellular fluid volume). Plasma renin activity was estimated by the technique of Haber et al. Briefly, arterial blood was placed in chilled test tubes containing ethylenediaminetetraacetic acid (EDTA)-sodium immediately after being drawn. The plasma was separated by centrifugation and stored frozen until the renin activity was determined by measuring radioimmunologically the rate of angiotensin I generation in the inhibitor-treated plasma. Mean arterial pressure was measured from the indwelling arterial cannula connected to a Statham model 23Ac pressure transducer, the output of which was recorded on a Grass polygraph. The transducer was calibrated using a mercury manometer before each measurement period. Sodium space was calculated from the dilution of 10 \(\mu\)Ci of \(^{22}\)Na injected intravenously 3 hours prior to sampling. The dogs were fed after the completion of daily data collection.

**Results**

Figure 1 shows the levels of urinary sodium and potassium excretion at the end of the four levels of aldosterone infusion. The group means ± 1 SE are indicated. The shaded area labeled “normal range” is the mean ± 1 SD urinary excretion rate of a group of eight intact dogs eating the same sodium and potassium intake as the dogs in the present experiment. At each level of aldosterone infusion the average excretion rate is within or very close to the normal range, indicating the dogs were in a steady state of sodium and potassium balance. Differences between the rates of excretion at the different infusion levels probably are the results of errors in urine collection rather than actual differences in the steady state excretion rates.

**RELATIONSHIP BETWEEN ALDOSTERONE AND PLASMA SODIUM CONCENTRATION**

Figure 2 is a plot of the relationship between aldosterone levels and plasma sodium concentration. The normal level of plasma sodium concentration for dogs in our laboratory is between 140 and 143 mEq/liter, a level that, according to the plot of Figure 2, could be achieved by infusion of between 48 and 80 \(\mu\)g/day of aldosterone. Aldosterone levels appeared to have a potent influence on plasma sodium concentration at the low levels of infusion but very little effect at the infusion rates above the normal range.

**EFFECTS OF ALDOSTERONE ON PLASMA POTASSIUM CONCENTRATION**

Changes in aldosterone infusion rates produced very consistent changes in plasma potassium concentration, as can be seen in Figure 3. Again, the effect of the hormone was greatest at the lower levels of infusion. Normal plasma potassium concentration in the dogs in our laboratory is between 3.75 and 4.00 mEq/liter. According to the plot of Figure 3, this level could be achieved by infusion of between 40 and 60 \(\mu\)g/day in dogs of this size on this particular diet.

**EFFECTS OF ALDOSTERONE ON PLASMA RENIN ACTIVITY**

Plasma renin activity of dogs in our laboratory eating a diet containing 27 mEq/day each of sodium and potassium...
per day is normally between 0.7 and 1.5 ng/ml per hour. According to the plot of Figure 4, this level would be achieved by aldosterone infusions of approximately 40–60 μg/day. As one can see in Figure 4, plasma renin activity levels rise at a rapid rate as aldosterone levels fall below normal and, conversely, drop to zero at rates of aldosterone administration slightly above normal. This dramatic relationship between aldosterone and renin probably is involved in producing the complex relationship between aldosterone, on the one hand, and sodium space and mean arterial pressure, on the other hand, shown in Figures 5 and 6.

RELATIONSHIP BETWEEN ALDOSTERONE AND SODIUM SPACE AND BODY WEIGHT

Sodium space increased with increasing aldosterone at the two lower levels of aldosterone infusion, and then appeared to plateau as the rate of infusion increased above the normal rate (Fig. 5). The mean value actually decreased slightly as the infusion rate was more than doubled from level III, 91 μg/24 hours, to level IV, 219 μg/24 hours. Body weight increased progressively as the level of aldosterone infusion was increased. Going from level I to IV the weights (in kilograms) averaged 22.56 ± 0.63, 23.11 ± 0.62, 24.86 ± 0.85, and 25.16 ± 0.80.

RELATIONSHIP BETWEEN ALDOSTERONE AND MEAN ARTERIAL PRESSURE

The relationship between aldosterone and arterial pressure (Fig. 6) was complex. Normal arterial pressure in conscious dogs in our laboratory averages about 100 mm Hg. Readings close to this level were found both at the second level of infusion, which is close to the normal aldosterone secretion rate, and at the first level of infusion, probably 1/4 to 1/2 the normal secretion rate. At these two lower rates of aldosterone administration there was no positive correlation between aldosterone and pressure. But as aldosterone infusion increased above the normal level, up to 91 and 219 μg/24 hours, the pressure did rise, reaching a level of 119 ± 2 mm Hg at the highest aldosterone level.
Discussion

By infusing measured amounts of aldosterone into adrenalectomized dogs for several weeks at each of four rates, we were able to obtain steady state dose-response relationships between aldosterone and five variables. Plasma sodium concentration and sodium space both declined sharply as aldosterone infusion rates fell below normal, but these variables increased only slightly above normal as aldosterone infusion rates were increased to approximately 4 times normal. This type of relationship suggests that aldosterone is not the prime controller of sodium concentration but, rather, a minimum level of aldosterone must be maintained for other mechanisms to be effective in its actual control. This conclusion is supported by two studies recently completed in this laboratory. In the first,6-7 feedback control of aldosterone secretion was blocked in dogs and then sodium intake was increased from a low level (10 mEq/day) to a high level (200 mEq/day). Feedback control of aldosterone secretion was blocked by adrenalectomy several weeks prior to the study and then maintaining the days on a fixed amount of infused aldosterone (100 μg/day) throughout both low and high sodium intake periods. In that study, the 20-fold increase in sodium intake resulted in a 2% change in plasma sodium concentration, exactly the same change seen when the same 20-fold increase in sodium intake was given to a group of intact dogs in which the level of aldosterone secretion could change in response to the change in intake. Although both the group without feedback control of aldosterone secretion and the intact group possessed the same ability to control plasma sodium concentration precisely, control of sodium balance was greatly impaired in the group lacking functional aldosterone control; in response to the increase in sodium intake the control group showed a 2% increase in 22Na space, whereas the experimental group had a 12% increase in sodium space. From those data it was concluded that the aldosterone feedback control system is not important in control of extracellular sodium concentration although it is of significant importance in regulating sodium balance.

A second group of experiments sought to determine how sodium concentration was controlled if aldosterone was not involved. In this study of degree of involvement of the antidiuretic hormone (ADH)-thirst feedback control system in regulating extracellular sodium concentration was assessed.8-9 The same 20-fold increase in sodium intake was again given, first to a group of intact dogs, then to a group with blocked feedback control of ADH secretion and water intake. Feedback control was blocked by continuously infusing ADH at a rate which maintained the dogs' urine osmolality at a maximal level (greater than 1,500 mOsmol/liter) during the low and the high intake period, and by giving the dogs a mandatory water intake of 700 ml/day during both low and high intake periods. Again, the 20-fold increase in sodium intake resulted in a 2% increase in sodium concentration in the intact group, but now a 12% increase in sodium concentration was measured in the group with blocked feedback control of the ADH-thirst system. From this study it was concluded that the ADH-thirst feedback control system was the primary controller of extracellular sodium concentration.

Although no ADH measurements were made during the study described in the present paper, it is reasonable to propose that the fall in sodium concentration observed at the lowest level of aldosterone infusion did result from a stimulation of the ADH system due to sodium depletion and hypovolemia.10 This contention is supported by data from severe Addisonian patients, showing that such patients have high levels of ADH associated with hyponatremia and volume depletion.11

Plasma potassium concentration appears to be controlled mainly by the aldosterone feedback mechanism, as evidenced by the consistent changes in plasma potassium levels resulting from altering the aldosterone infusion rate from the lowest to the highest rate.

The complex relationship between aldosterone and arterial pressure may be explained by the participation of angiotensin in control of arterial pressure and sodium and volume balance. At the normal aldosterone level (48 μg/24 hours) fluid volume, plasma renin activity, and arterial pressure were at or near normal values. At the lower aldosterone level (16 μg/24 hours) pressure was still normal while sodium space was decreased by an average of 952 ml, or 13%. It is possible that this large decrease in fluid volume did not cause a change in arterial pressure because of the concomitant very marked rise in renin activity levels. The difference in plasma renin activity between the two levels was 457%, 9.6 vs. 2.1 ng/ml per hour. Under several conditions of reduced volume, such as sodium depletion12,13 and hemorrhage,14,15 increased renin secretion has been shown to be a major factor in maintaining normal arterial pressure. Therefore, it is reasonable to suggest that the pressor action of the high level of angiotensin may have been responsible in the experiments for maintaining arterial pressure during the reduced volume state. As aldosterone levels were increased above the normal level and arterial pressure began to rise, plasma renin activity fell abruptly to zero. With renin levels maximally suppressed, further increases in fluid volume could no longer be compensated for by further reducing the pressor contribution of angiotensin; this possibly explains the rise in arterial pressure when aldosterone levels and fluid volume were increased above normal even though the measured fluid volume increase was minimal.

The highest average arterial pressure recorded in this study was 119 ± 2 mm Hg. This pressure was the steady state result of aldosterone infused at rates 4½ times normal. In this experimental situation it would not have been possible to increase arterial pressure much higher by giving additional aldosterone, since pilot studies showed that slightly greater mineralocorticoid activity would lead to debilitating or fatal potassium depletion. In the pilot experiment, two adrenalectomized dogs eating the same diet used in the present study were given 300 μg/day of aldosterone for 2 weeks, after which the study had to be discontinued because the dogs' plasma potassium levels had dropped below 2.00 mEq/liter. Therefore, unless potassium intake is radically increased, the effects of mineralocorticoids alone probably cannot raise arterial pressure more than about 30 mm Hg in the dog. If these results
have relevance to human physiology, they also suggest that the very high arterial pressures occasionally seen in primary aldosteronism may be only partially due to the effects of the excessive mineralocorticoid levels. An important additional factor contributing to the marked hypertension in these cases may be vascular lesions, especially in the renal preglomerular bed, resulting from prolonged increased pressure, particularly in view of the fact that the arterial pressure often fails to return to normal following removal of the causative adrenal tissue.  

These results also suggest that the renin-angiotensin system may have a direct effect on sodium balance control. The dogs were in a steady state of sodium balance at infusion levels I and II even though at level I only 33% as much aldosterone was being given as at level II. Apparently the natriuretic effect of the reduction in aldosterone administration was partially balanced by an increase in the level of other antinatriuretic agents or factors. Probably one factor involved was angiotensin II, because in acute experiments in several laboratories, intravenous renal and arterial angiotensin II infusions at high physiological rates have produced a fall in sodium excretion. Therefore, it is likely that the 4 1/2-fold increase in plasma renin activity (and presumed resultant increase in angiotensin II) accounted for a portion of the compensatory sodium reabsorption in the present experiment. Apparently as well as affecting sodium balance indirectly through its stimulation of aldosterone, the renin-angiotensin system also plays an important parallel role in sodium control by directly affecting sodium reabsorption by the kidney.

In summary, the results of this study describe the quantitative steady state relationships between aldosterone and plasma sodium concentration, potassium concentration, plasma renin activity, sodium space, and arterial pressure. Decreasing aldosterone levels below normal strongly affects plasma sodium concentration and sodium space, whereas raising aldosterone above normal has little effect on these variables. Plasma potassium concentration varies in a highly consistent manner with changes in aldosterone levels. Plasma renin activity rises rapidly as aldosterone falls below normal and drops to zero at aldosterone levels slightly above normal. Arterial pressure increases as aldosterone rises above normal, but does not fall below normal at subnormal aldosterone levels, probably because of the pressor effects of angiotensin II.

References
8. Young DB, Guyton AC: Control of plasma sodium concentration by the ADH-thirst mechanism. Physiologist 17: 364, 1974
9. Young DB, Pan YJ, Guyton AC: Control of extracellular sodium concentration by the ADH-thirst feedback mechanism. Am J Physiol (accepted for publication)
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