

THE ROLE OF ALDOSTERONE IN RENIN SECRETION<sup>1</sup>

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Numerous investigators have demonstrated that a primary excess or deficit of adrenal mineralcorticoids is generally associated with decreased or increased renin secretion, respectively, (1-12). Our present experiments were designed to study the nature of this interaction, specifically whether aldosterone exerts a direct effect on renin secretion independent of its indirect effects resulting from steroid-induced changes in body sodium balance. This question is of particular interest since it has recently been demonstrated that angiotensin inhibits renin secretion in dog (13) and man (14). Since angiotensin is a potent stimulator of aldosterone secretion, it is possible that the inhibitory action of angiotensin on renin secretion is mediated by aldosterone. Such an effect, however, could not be the result of steroid-induced changes in body sodium or tubular reabsorption because it is manifested within minutes after the administration of angiotensin (13).

Methods

The effects of aldosterone on renin secretion were studied in dogs over short periods of administration in acute, anesthetized preparations and over longer periods using trained, unanesthetized dogs.

Acute Experiments: These studies were performed on dogs anesthetized with pentobarbital, 30 mg/Kg, administered intravenously. Through a right flank incision, the right ureter was cannulated with polyethylene tubing.

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In many of these experiments, a clamp was positioned loosely around the aorta proximal to the origin of the renal arteries. By tightening this clamp, the mean pressure in the aorta below it could be adjusted within  $\pm 3$  mm Hg. Arterial blood pressures above and below the clamp were monitored from a carotid and a femoral artery using strain gauges recording on a polygraph. To obtain renal venous blood, a polyethylene catheter (2.4 mm, o.d.) was introduced into the left femoral vein, passed up the inferior vena cava, and guided manually into the right renal vein. Arterial blood was obtained from a second femoral artery catheter. Brachial veins were catheterized for delivery of fluids by constant infusion pumps. Renal excretory and hemodynamic data were obtained using standard clearance technique. Each animal was primed with creatinine and p-aminohippurate after which these substances were infused in isotonic saline at the constant rate of 0.2 ml/min. Arterial and renal venous blood samples were taken at the midpoint of each clearance period. Creatinine clearance was used as a measure of glomerular filtration rate (GFR), and total renal plasma flow (RPF) was determined by the Fick principle, using p-aminohippurate. At least 30 minutes after completion of all operative procedures and the administration of the creatinine and p-aminohippurate prime, control clearances were performed. The protocols then followed are described in the Results section.

Chronic Experiments: To observe aldosterone's affect on renin secretion over a longer time course in unanesthetized animals, dogs were trained to lie quietly on a table for a daily routine of tests. Catheters were chronically implanted in the aorta through the carotid artery, and in the superior vena cava through the jugular vein, and were brought subcutaneously out the back of the neck. After the operative procedures and training, the dogs were begun on a controlled diet of regulated sodium content. At the same time each day, arterial mean and pulsatile blood pressures were monitored, and arterial blood samples were drawn for determination of hematocrit, renin, sodium, and

potassium. Red blood cells were returned suspended in an equal volume of isotonic saline. Following a series of control days on a 60 mEq sodium diet, the sodium intake was reduced to 10 mEq per day. In addition, 2 of the 3 dogs were given mercurhydrin, 2 ml (80 mg Hg) intramuscularly, on the first day of dietary change. The mercurial was given to facilitate salt depletion and cause a more marked increase of plasma renin (15). After 3-5 days on the 10 mEq Na diet, d-aldosterone, 1 mg/24 hours, was administered intramuscularly twice daily in sesame oil for 3-5 days, the 10 mEq Na diet still being maintained.

A fourth animal was studied similarly after bilateral adrenalectomy. This dog was maintained on cortisone, 25 mg/day, and was given 120 mEq Na (instead of 60) during the control period.

Analytical Techniques: Urine and plasma electrolytes were determined by flame photometry. Creatinine was analysed by the method of Bonsnes and Taussky (16), PAH by the method of Smith et al. (17).

Two methods were used for estimation of renin. That used for renin determination in the acute animals has been described previously in detail (18). Briefly, the renal venous plasma sample is incubated under standardized conditions for 31 minutes, during which time angiotensin II is formed in a quantity directly related to the renin concentration of the plasma. The generated angiotensin is then extracted into butanol, redissolved in saline, and assayed by comparing its pressor activity with that of standards of synthetic Val<sup>5</sup>-angiotensin II amide in anesthetized ganglion-blocked rats. Renin concentrations were expressed in terms of nanograms of angiotensin formed (Ang.-Equiv.) per ml of original plasma.

The method used for renin determinations in the chronic animals is the same as the preceding in theory, (i.e. angiotensin generation during plasma incubation and rat pressor bioassay), but is more sensitive. It is essentially that described by Boucher, et al. (19), the only difference being that, to

to facilitate final solvent sublimation, we employed butanol extraction. Renin concentrations are expressed in the same units as those described above for the method used in acute dogs, but the values are not comparable.

### Results

Acute Experiments: Protocol 1: Acute infusion of d-aldosterone into anesthetized animals secreting renin at near basal rates. Following the control samples, d-aldosterone was infused into animals at the rates of 1.2 to 2.0  $\mu\text{g}/\text{Kg hr}$ . From 0.5 to 6 hours after beginning the d-aldosterone infusion, further clearances were performed and renal venous blood drawn. At least 30 minutes after discontinuing d-aldosterone infusion, further sets of control

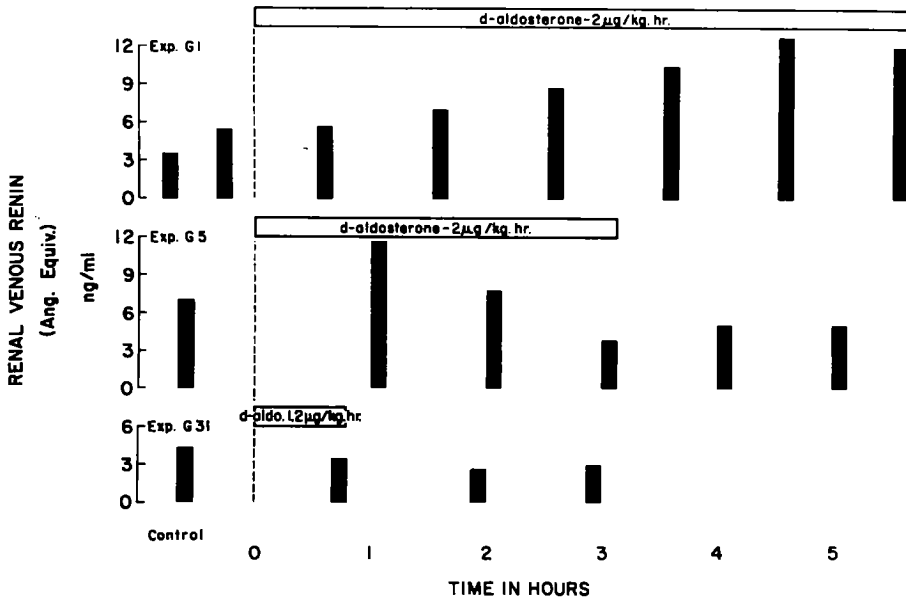


FIG. 1

Acute intravenous infusion of aqueous d-aldosterone in three anesthetized dogs with normal renal perfusion pressures.

clearances were collected. As seen in Figure 1, aldosterone had no consistent effect on renal venous renin. There was no significant changes in renal hemodynamics or plasma sodium or potassium.

Protocol 2: Acute infusion of d-aldosterone into anesthetized animals stimulated to high rates of renin release by lowered renal perfusion pressure.

In this series of animals, after the control periods, the aorta was constricted by tightening the adjustable clamp to the point where the arterial blood pressure distal to the clamp was controlled between 80 and 90 mm Hg. This lowered renal perfusion pressure has been shown to be consistent stimulus to renin release (18, 20). At least 30 minutes after aortic constriction, clearances were performed and a renal venous blood sample was collected before aldosterone infusion was begun. D-Aldosterone was then administered intravenously at the rate of 2  $\mu\text{g}/\text{Kg hr}$  for a period of from one to four hours, renal arterial pressure being still maintained at 80-90 mm Hg. As seen in Figure 2, the reduction of mean renal arterial pressure enhanced the release of renin. When aldosterone was infused, these higher levels were not depressed. GFR, RFF, and sodium excretion were decreased by the reduction in renal arterial pressure, and these hemodynamic and excretory parameters were not modified by the infusion of d-aldosterone.

Chronic Experiments: Figure 3 illustrates data from a typical experiment, the results being similar in all three dogs. Following the induction of negative sodium balance, there occurred a rapid renin rise, hemoconcentration, and weight loss. When the animal received repository doses of aldosterone, there was no significant change in plasma renin concentration. Reinstating 60 mEq Na diet reversed all changes.

Figure 4 illustrates the data from the adrenalectomized dog. Note that on successive days after dietary sodium depletion, he reduced his urinary sodium to 43 mEq/day, despite the absence of adrenals.

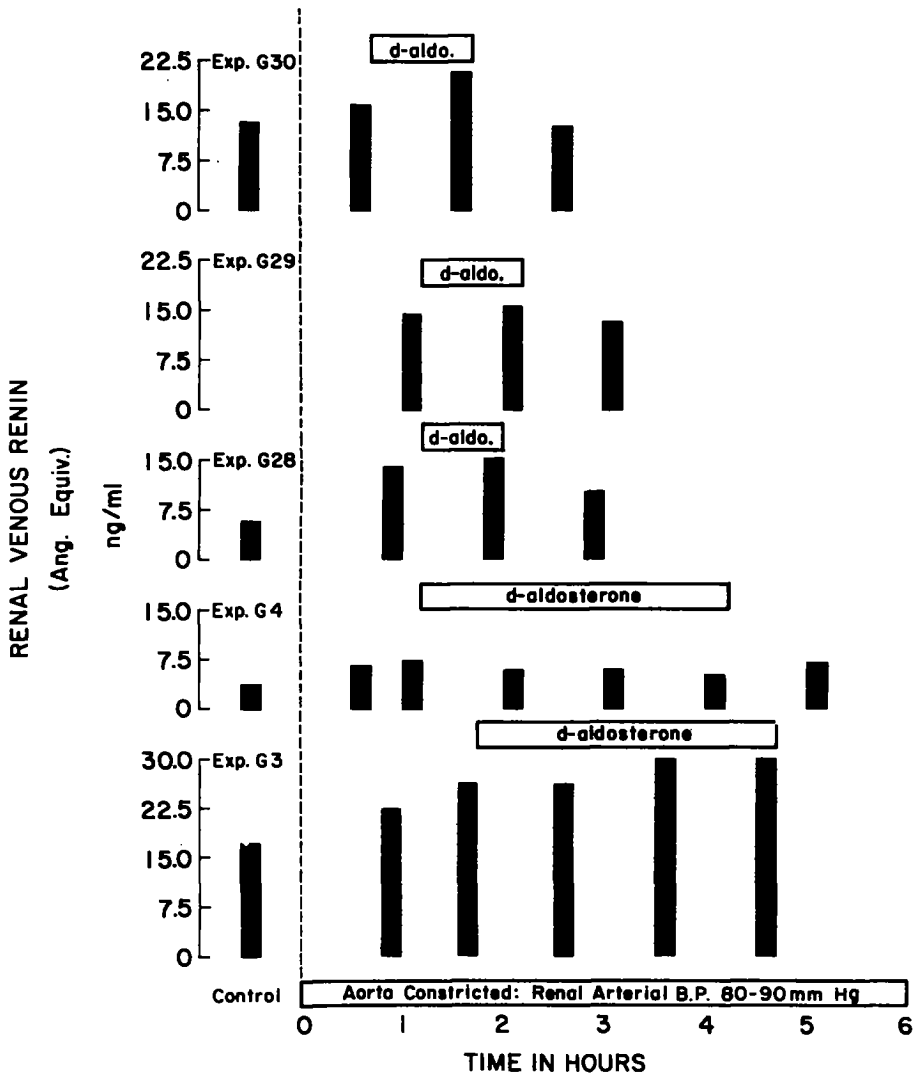


FIG. 2

Acute intravenous infusion of aqueous d-aldosterone, 2.0 micrograms/kg hr., in five anesthetized dogs with aortic constriction and reduced renal perfusion pressures.

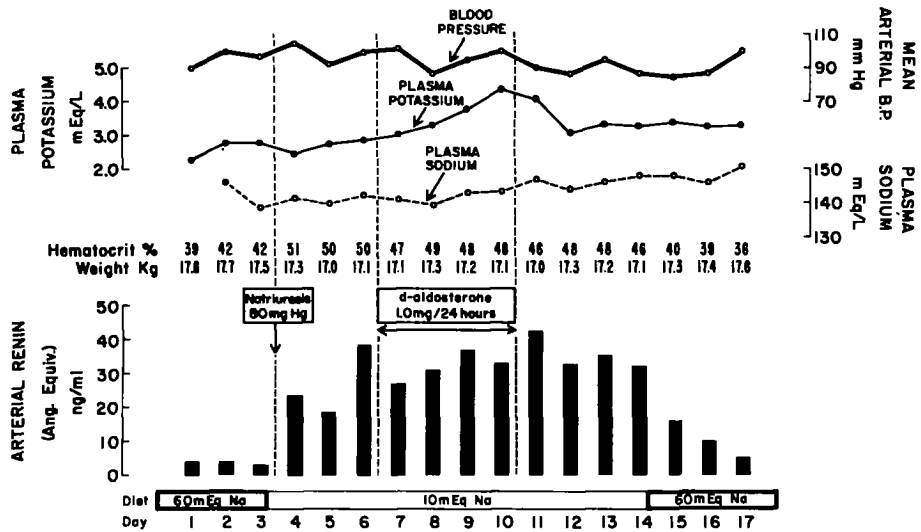


FIG. 3

Chronic repository administration of d-aldosterone in sesame oil to an unanesthetized dog during sodium depletion by mercurial natriuresis and diet control.

However, only while receiving aldosterone was he able to virtually clear his urine of sodium and maintain a new sodium balance at the 10 mEq Na intake. When aldosterone was withdrawn, urinary sodium excretion again increased and further negative sodium balance ensued. These changes are clearly reflected in the plasma renin concentrations which first increase, then plateau, then rapidly rise to very high values.

Discussion

These experiments have demonstrated that acute infusions of large quantities of aldosterone failed to decrease renal venous renin concentration in dogs under conditions of basal or elevated renin secretion. It is evident, therefore, that the ability of angiotensin invariably to decrease renin concentration under

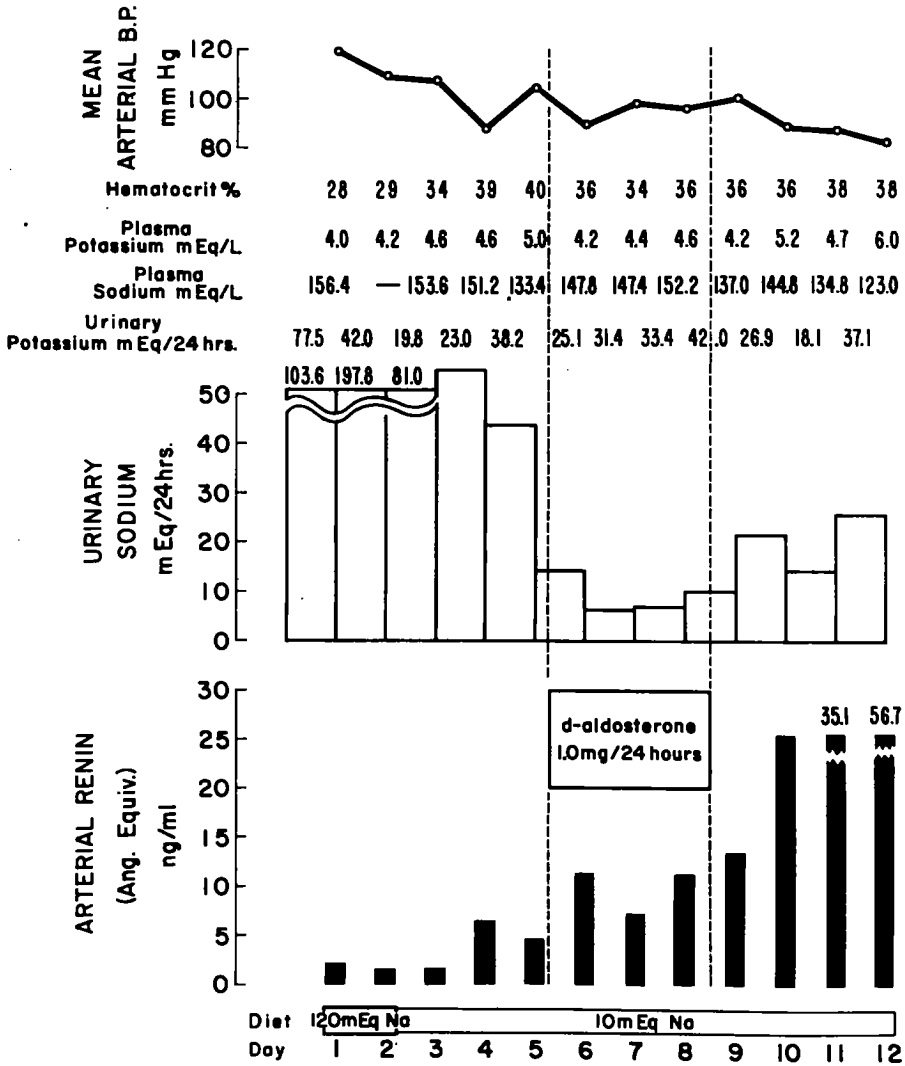


FIG. 4  
 Chronic repository administration of d-aldosterone in sesame oil during dietary sodium depletion in an unanesthetized adrenalectomized dog.



identical conditions (13) cannot be mediated by aldosterone. Similarly, while our experiments were in progress, Genest, et al. (14) reported that acute administration of angiotensin but not aldosterone lowered plasma renin concentration in humans on a low sodium diet.

In 1953, Hartroft and Hartroft (2) postulated that the effects of mineralcorticoids on juxtaglomerular granulation were due primarily to steroid-induced changes in body sodium. Gross, et al. (3), in studies of renal renin content, were the first to postulate a similar conclusion regarding the relationship of renal renin and adrenal steroids. Since these initial reports, similar studies have been reported by others (1,4-7). The arterial renin data of our present chronic experiments add further support to this concept of the indirect nature of steroid action upon renin secretion. In all experiments, the administration of aldosterone to dogs maintained on a low sodium diet (and thus prevented from retaining sodium) produced no reduction of arterial renin concentration. The pattern demonstrated by the adrenalectomized dog well illustrates these relationships. On a high sodium diet, the animal was able to maintain balance and had a very low arterial renin despite the absence of aldosterone. Upon institution of the low sodium diet, negative sodium balance ensued and arterial renin increased. The administration of aldosterone prevented further sodium loss and, thereby, stabilized but did not reverse the increased renin secretion. Upon cessation of the aldosterone, further sodium depletion occurred and renin secretion increased.

The conclusion of these experiments should point out a clinical caution, since a key element in the diagnosis of primary aldosteronism (Conn's Disease) is an abnormally low value for plasma renin (8-12). However a patient pretreated with diuretics for hypertension may fail to present with such a low renin value, since, according to our interpretation, the plasma renin is a function of sodium balance rather than a reflection of high aldosterone level per se.

### Summary

Aldosterone was administered acutely and chronically to dogs under conditions of basal or elevated renin secretion. When steroid-induced changes in sodium balance were prevented by dietary sodium restriction, aldosterone produced no change in arterial renin concentration. We conclude that the effects of aldosterone on renin secretion are not direct, but are secondary to steroid-induced changes in body sodium.

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