Regulation of Aldosterone in Anephric Man maintained with Regular Haemodialysis

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Of the four known humoral factors participating in the control of aldosterone secretion, the renin-angiotensin system, ACTH, sodium and potassium (Blair-West et al, 1963; Ganong et al, 1966; Davis et al, 1968), the renin-angiotensin system is generally considered to be of primary importance. It is of interest, therefore, to evaluate the regulation of aldosterone in a model in which renal renin secretion is no longer present, i.e., anephric man. This report includes data obtained from a group of bilaterally nephrectomised subjects and describes the effects of potassium administration, haemodialysis, and infusion of angiotensin II or ACTH on plasma aldosterone concentration in these patients.

MATERIAL AND METHODS

Eleven patients were evaluated who had undergone bilateral nephrectomy two weeks to 35 months prior to these studies. Blood pressure, body weight and plasma sodium, potassium, renin activity and cortisol were determined before and after: (a) oral administration of 60 mM of potassium (seven cases); (b) haemodialysis without ultrafiltration, using potassium-free dialysate (six of the potassium-loaded patients); (c) haemodialysis with ultrafiltration, using potassium-free or -poor (1 mM/l) dialysate (14 tests in seven patients); (d) haemodialysis with ultrafiltration, using dialysate containing 4.5 mM/l of potassium (seven patients); (e) infusion of a pressor dose of angiotensin (Kaplan & Silah, 1964) for one hour (ten patients); and (f) infusion of cosyntrin, a synthetic ACTH, at a rate of 0.1 mg per hour for two hours (nine patients). Patients were supine for at least one hour before blood samples were collected. The pre-test samples as well as an additional fifteen determinations obtained from the same group of patients under similar control conditions will be referred to as 'baseline' values. Plasma sodium and potassium concentrations were measured by Instrumentation Laboratory
flame photometer. Plasma renin activity (PRA) was determined by bioassay (Boucher et al, 1964), plasma aldosterone concentration by a radioimmunoassay method (Ito et al, 1972) and plasma cortisol by competitive protein binding (Mayes & Nugent, 1966). For statistical analysis of plasma aldosterone, the natural logarithm of the respective value was used.

RESULTS

PRA was consistently undetectable in ten patients; in the eleventh it ranged between 30 and 50 ng/100 ml (normal 30 to 600 ng/100 ml) and was unchanged following the different test procedures (Table I). Supine baseline plasma aldosterone levels ranged from less than 0.5 to 18 ng/100 ml. The majority of values were below 2 ng/100 ml, which is clearly subnormal. Statistical analysis showed significantly higher values for aldosterone, plasma potassium, body weight and blood pressure obtained on the third and fourth day following haemodialysis, as compared with those found on the first two days after dialysis (p < .05). There was a weak but statistically significant correlation between baseline plasma aldosterone concentration and the corresponding plasma potassium values (r = +.35; p < .01).

Table I. Plasma renin activity in 11 anephric patients

<table>
<thead>
<tr>
<th>Study condition</th>
<th>Total no of patients studied</th>
<th>Undetectable PRA</th>
<th>Detectable PRA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No of patients</td>
<td>No of patients</td>
<td>No of patients</td>
</tr>
<tr>
<td>Supine</td>
<td>11</td>
<td>10</td>
<td>1</td>
</tr>
<tr>
<td>Standing</td>
<td>11</td>
<td>10</td>
<td>1</td>
</tr>
<tr>
<td>After ultrafiltration</td>
<td>11</td>
<td>10</td>
<td>1</td>
</tr>
<tr>
<td>After angiotensin</td>
<td>10</td>
<td>9</td>
<td>1</td>
</tr>
<tr>
<td>After ACTH</td>
<td>6</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>After Heparin</td>
<td>6</td>
<td>5</td>
<td>1</td>
</tr>
</tbody>
</table>

* All samples with detectable PRA obtained from the same patient (34 year old female)

Measurements carried out two hours following oral potassium loading showed a mean increase in plasma potassium of 1.09 mM/1, associated with an acute increase in plasma aldosterone in six of seven tests (mean increase 2.85 ng/100 ml; p < .0025), but with no significant change in plasma sodium, body weight or blood pressure (Table II).

In these potassium-loaded patients, subsequent haemodialysis without ultrafiltration using potassium-free dialysate resulted in a mean decrease in plasma potassium of 2.05 mM/1 and in a significant fall in mean plasma aldosterone of 2.90 ng/100 ml (p < .005), while plasma sodium, body weight and blood pressure did not change significantly (Table II).

Haemodialysis with ultrafiltration using potassium-free or -poor (1 mM/1) dialysate consistently caused a decrease in plasma potassium (mean decrease
Table II. Effect of oral potassium loading and haemodialysis on plasma aldosterone, potassium, sodium, weight, and blood pressure in anephric man

<table>
<thead>
<tr>
<th>Experimental procedure</th>
<th>N</th>
<th>Aldosterone (ng/100 ml) Mean ± SE Range</th>
<th>Potassium (mM/l) Mean ± SE Range</th>
<th>Sodium (mM/l) Mean ± SE Range</th>
<th>Weight (kg) Mean ± SE Range</th>
<th>Mean BP (mm Hg) Mean ± SE Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral potassium loading</td>
<td>7</td>
<td>-0.06 ± 0.66 to &lt;0.0025</td>
<td>+0.6 ± 0.16 to &lt;0.001</td>
<td>+0.71 ± 0.36 to NS</td>
<td>+0 ± 0 to -- NS</td>
<td>+1.0 ± 3.0 to -- NS</td>
</tr>
<tr>
<td>(60 mM)</td>
<td></td>
<td>+5.70</td>
<td>-1.6</td>
<td>+2</td>
<td>+18.0</td>
<td>-18.0</td>
</tr>
<tr>
<td>Haemodialysis</td>
<td>6</td>
<td>-0.06 ± 1.04 to &lt;0.005</td>
<td>-1.5 ± 0.29 to &lt;0.001</td>
<td>-1.00 ± 1.46 to NS</td>
<td>-0.65 ± 0.30 to NS</td>
<td>-1.70 ± 0.44 to NS</td>
</tr>
<tr>
<td>no ultrafiltration</td>
<td></td>
<td>-6.50</td>
<td>-3.4</td>
<td>-5</td>
<td>-1.90 ± 0.44 to NS</td>
<td>-34.7 ± 0.44 to NS</td>
</tr>
<tr>
<td>dialysate-K 0 mM/l</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haemodialysis</td>
<td>14</td>
<td>-3.11 ± 1.07 to &lt;0.005</td>
<td>-1.64 ± 0.22 to &lt;0.001</td>
<td>-3.71 ± 0.68 to &lt;0.001</td>
<td>-1.83 ± 0.43 to &lt;0.0125</td>
<td>-19.8 ± 2.1 to &lt;0.001</td>
</tr>
<tr>
<td>ultrafiltration</td>
<td></td>
<td>+1.00</td>
<td>+0.1</td>
<td>+0</td>
<td>-4.50 ± 0.43 to NS</td>
<td>-36.0 ± 0.44 to NS</td>
</tr>
<tr>
<td>dialysate-K 0-1 mM/l</td>
<td></td>
<td>-5.60</td>
<td>-3.6</td>
<td>-9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haemodialysis</td>
<td>7</td>
<td>+9.4</td>
<td>+0.5</td>
<td>+4</td>
<td>-0.5</td>
<td>+1.0</td>
</tr>
<tr>
<td>ultrafiltration</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dialysate-K 4.5 mM/l</td>
<td></td>
<td>+1.16 ± 1.66 to NS</td>
<td>-0.09 ± 0.36 to NS</td>
<td>-3.43 ± 1.49 to &lt;0.05</td>
<td>-2.33 ± 0.80 to &lt;0.0125</td>
<td>-18.5 ± 5.9 to &lt;0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td>-5.6</td>
<td>-1.2</td>
<td>-8</td>
<td>-5.60 ± 0.43 to NS</td>
<td>-36.0 ± 0.44 to NS</td>
</tr>
</tbody>
</table>
1.64 mM/l) and in eleven of fourteen instances a decrease in plasma aldosterone (mean decrease 3.11 ng/100 ml; p < .005); in addition, plasma sodium, body weight and blood pressure decreased significantly during this manoeuvre (p < .0125).

In contrast, neither plasma potassium nor plasma aldosterone were altered significantly following haemodialysis with ultrafiltration using a higher dialysate potassium concentration of 4.5 mM/l, while plasma sodium, body weight and blood pressure again decreased significantly (p < .05) with the absolute decreases comparable to those during the previous ultrafiltration studies (Table II).

Regression analysis of the potassium loading and haemodialysis experiments revealed a significant correlation between the acute changes in plasma potassium and the corresponding alterations in aldosterone (r = .64; p < .001) as well as a weak positive correlation between aldosterone and plasma sodium (r = .46; p < .01). In contrast, no correlation was observed between the changes in aldosterone and body weight or blood pressure.

The response of plasma aldosterone to the stimulatory effect of a pressor dose of angiotensin II infused for one hour was blunted or absent in eight of

![Graph](image)

**Figure 1.** The effect of infusions of either angiotensin II or synthetic ACTH (cosyn-tropin) on plasma aldosterone concentration in anephric subjects. The shaded areas in the upper half of the figure indicate the normal means ± 1 SD from previously reported data (Horton, 1969; Michalakis & Horton, 1970)
the ten anephric subjects studied (Figure 1). Patients with the lower baseline aldosterone levels were usually less responsive and, in addition, tended to have lower baseline plasma potassium concentrations. Similar blunted responses were seen in several patients during and following an infusion of synthetic ACTH (Figure 1). Baseline potassium levels ranged from 2.7 to 4.0 mM/1 (mean 3.54 mM/1) before the angiotensin-infusion and from 2.5 to 4.6 mM/1 (mean 3.77 mM/1) before the ACTH test. Plasma electrolytes did not change significantly with the two infusions. Baseline plasma cortisol levels were normal and were not altered significantly following the administration of angiotensin, but increased two- to more than three-fold with the ACTH infusion.

DISCUSSION

Although several components may be important, the renin-angiotensin system is generally considered to play a major role in the control of aldosterone secretion (Davis et al., 1962; Ganong et al., 1966; Davis et al., 1968; Peart, 1970). In the present study, measurable plasma renin activity was present in only one of eleven anephric subjects, and in none of these patients was there any change in plasma renin following known stimuli of renin release, such as upright posture or acute reduction of body fluid by ultrafiltration. These results are consistent with recent findings by others (Bayard et al., 1971; Berman et al., 1972) and provided the necessary baseline condition for the evaluation of factors other than the renin-angiotensin system in the control of aldosterone.

Both the demonstration of a significant correlation between plasma aldosterone and potassium under baseline conditions ($r = .35; p < .01$) and the observation of significant parallel increases of plasma aldosterone and potassium on the third and fourth as compared to the first two days following haemodialysis ($p < .05$) are in agreement with the data recently reported by Bayard et al. (1971). It should be noted, however, that the latter authors found a much closer correlation between the two parameters during baseline conditions.

An additional finding resulting from the present study is the demonstration of acute changes in plasma aldosterone in anephric subjects in response to acute small alterations in plasma potassium, e.g., oral potassium loading was followed by an acute parallel increase of both plasma potassium and aldosterone; whereas both parameters decreased as a consequence of haemodialysis against potassium-free or -poor dialysate, or remained statistically unchanged when alteration in potassium during haemodialysis were minimised by the use of a dialysate containing 4.5 mM/1 of potassium. The significance of this relationship is underlined by the close correlation which was obtained between the acutely induced alterations in plasma potassium and the
accompanying changes in plasma aldosterone (r = .64; p < .001). These data indicate that in the absence of the renin-angiotensin system and its adreno- 
trophic effect, potassium is an important and effective factor in the control of aldosterone secretion.

Another electrolyte which may influence aldosterone secretion in aneph- 
ric man is the sodium ion. Marked reduction in the sodium level of adrenal 
perfsusate and sodium depletion in nephrectomised or angiotensin-immunised 
animals have been reported to directly stimulate aldosterone secretion 
(Blair-West et al, 1963; Davis et al, 1963; Palmore et al, 1969; Lowenstein 
et al, 1971). In the present study, the acute decreases in plasma sodium 
induced by haemodialysis were usually rather small (always less than 9 mM/1) 
(Table II) and there was no apparent stimulatory effect of these alterations on 
plasma aldosterone.* In another study in man removal of 170 to 330 mM of 
sodium by peritoneal dialysis failed to induce an increase in plasma aldoste- one (Peart, 1970). These findings do certainly not exclude participation of 
sodium in the regulation of aldosterone in anephric man; however, they 
indicate that its effect on adrenal function in these patients may not be more 
than a minor one.

No relationship could be demonstrated between alterations in plasma 
aldosterone and changes in body weight or blood pressure. This finding is 
consistent with the concept that the stimulatory effect of volume depletion or 
a decrease in blood pressure on aldosterone secretion is mediated by the 
renin-angiotensin system (Davis et al, 1962; Brown et al, 1966; Newsome et 
al, 1968). In contrast to the report by McCaa et al (1971), there were no 
marked increases in plasma aldosterone following ultrafiltration haemodialy- 
sis with acute moderate weight loss (0.5 to 5.6 kg) in our anephric patients. 
It should be noted, however, that increases in plasma aldosterone seen 
following volume depletion may not necessarily reflect increased secretion; 
such changes would also be consistent with the effect of a diminished liver 
blood flow and, consecutively, a fall in the metabolic clearance rate of 
aldosterone (Davis et al, 1965).

A major determinant of aldosterone metabolism is the adrenal responsi- 
siveness to the action of the various aldosterone-stimulating factors. In the 
present study, a normal increase in plasma aldosterone following adminis- 
tration of angiotensin II or ACTH was observed in a minority of patients 
only, while a diminished or absent response was seen in most instances. 
Furthermore, the absence of apparent alterations in cortisol metabolism 
indicates the presence of an 'isolated' type of hypoaldosteronism (Vagnucci,

* The weakly positive correlation demonstrated by regression analyses be- 
tween changes in plasma aldosterone and sodium in this study (see above) is 
probably biologically meaningless. Under the specific experimental condi-
tions used there was often an interdependence between changes in plasma 
potassium and sodium, both moving in parallel directions.
1969) in some of these patients. In anephric man, both the low baseline aldosterone levels and reduced sensitivity of the aldosterone secreting mechanism could be at least partly due to the absence of circulating angiotensin and its adrenotrophic effect (Weidmann et al., 1972). In contrast to our results and those of Peart (1970), Mitra et al. (1972) reported normal aldosterone responsiveness to ACTH in their nephrectomised patients. A key factor in this apparent discrepancy may be differences in plasma potassium between the groups of patients reported. Mitra's patients were hyperkalaemic, with a mean serum potassium of 6.2 mM/1, whereas our patients had baseline potassium levels ranging from 2.7 to 4.0 mM/1 before the angiotensin infusion, and from 2.5 to 4.6 mM/1 before the ACTH test. In addition to the direct influence of plasma potassium concentration on baseline aldosterone levels, increased potassium intake has also been shown to potentiate the aldosterone response to ACTH (Williams et al., 1970) and aldosterone production in rat adrenal tissue in response to various stimuli was diminished during potassium deficit (Müller et al., 1970).

Another factor which might play a role in the altered aldosterone metabolism in haemodialysis-treated nephrectomised patients is heparin, a known inhibitor of aldosterone secretion (Schlatmann et al., 1964). The mechanism by which heparin and some related polysulphated polysaccharides exert their effect on adrenal function is not yet clear. However, Bayard and co-workers (1971), also studying anephric subjects were unable to lower plasma aldosterone with an infusion of heparin, and the results obtained in the present study from six patients are consistent with this finding. These latter data make it unlikely that the changes in plasma aldosterone during dialysis and the low levels of aldosterone seen on the first day following dialysis are the result of a heparin effect. However, a long term effect of chronic intermittent heparin therapy on aldosterone metabolism in anephric man has not been excluded, and more detailed studies are necessary to further investigate the exact role of this anticoagulant on adrenal function.

CONCLUSIONS

Plasma aldosterone concentration in eleven anephric subjects maintained on chronic haemodialysis, in ten of whom no plasma renin activity could be detected, was reduced in most instances. Plasma potassium and aldosterone levels correlated significantly under baseline conditions, and acute changes in plasma potassium caused parallel changes in plasma aldosterone. The data suggest that plasma potassium concentration is an important but not the sole factor controlling aldosterone secretion in the absence of the renin-angiotensin system. Plasma aldosterone concentration was unaffected by acute alterations in plasma sodium concentration, body weight or blood pressure. The responsiveness of aldosterone to the stimulatory action of
angiotensin II or ACTH was diminished in most anephric subjects, patients
with lower baseline aldosterone levels being less responsive. In these patients,
plasma cortisol levels were normal and responded appropriately to ACTH.
Factors which may contribute to the 'isolated' type of hypoaldosteronism seen
in some anephric subjects include, (a) the absence of circulating angiotensin,
(b) hypokalemia which may develop with maintenance haemodialysis. Chronic
intermittent therapy with heparin, a known inhibitor of aldosterone secretion,
may also play a role, although in the present study there were no acute
changes in plasma aldosterone following heparin infusions.

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OPEN DISCUSSION

M FERNANDEZ (Ghent): You mentioned that other factors may also play a role in modifying the blood aldosterone. Well, another factor that is present throughout the study is heparin. Have you made any studies to quantitate the amount of heparin you have given?

WEIDMANN: Yes, in fact we have studied six of these patients and I did not have time to show the slides. With heparin infusion for four hours using doses of heparin equal to the requirement during a routine dialysis, there was no decrease in plasma aldosterone. This point has been very carefully studied by Bayard et al (Bayard, F. et al (1972) Journal of Clinical Investigation, 50, 1585); they found that not only during the acute phase of a heparin infusion lasting several hours, but also on the following morning, there was no change in plasma aldosterone.

L E R GUERRA (Oporto): You have not mentioned the potassium levels before and after dialysis. Also, I would like to know if you have any idea of the variation of the potassium concentration inside the red cells?

WEIDMANN: To answer the first part of the question the mean pre-dialysis levels of potassium were around 4.9 mM/l and the post-dialysis levels were about 3.8 to 4.0 mM/l. We do not have very hyperkalaemic patients. To your second question, we have not measured any erythrocyte potassiuems.

GUERRA: We have some observations on this and levels of potassium inside red blood cells seem to change very much. Have you measured the variation
of phosphate, because phosphate not only in plasma but mainly inside red blood cells changes quite importantly, as we have observed. Have you any idea if you can in some way correlate your observations with phosphate metabolism?

WEIDMANN: Well, we can only try it. I have many suggestions on the benefit of doing it, but we do not actually see any relation between phosphate metabolism and aldosterone but there could be, of course.

B GOLDBERG (Johannesburg): Was there any correlation between those who showed a response of aldosterone secretion on posture, as compared with those who did not, when compared with the failure to respond to angiotensin?

WEIDMANN: Well, there is one common line throughout the whole study. We have the impression (although on this small number of patients we cannot substantiate it statistically) that the patients with lower plasma potassium levels not only respond less to ACTH and angiotensin, but they also respond certainly less in terms of vascular activity to angiotensin. In fact, we have measured aldosterone metabolic clearance rate and secretion rates in three of those who have responded in the upright posture, and found an increase in secretion rate of aldosterone in two patients and no change in one. Again, the one with the higher plasma potassium levels seemed to have a better response than the other two.

W SCHOEPPE (Frankfurt am Main): Have you any observations on the correlation between the increase in plasma potassium concentration and aldosterone, and the responsiveness of blood pressure during dialysis or in the period between?

WEIDMANN: Could you refer to Table II. The upper line shows the potassium loading experiments, and the middle two lines the different forms of haemodialysis with or without ultrafiltration. As you see, with the changes in the serum potassium and blood pressure in the different experiments carried out here, there is always a parallel change in potassium and aldosterone. In contrast, weight and sodium showed a different pattern, and the statistical correlation of these changes of weight and blood pressure against aldosterone was not significant.