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THE INTERACTION OF ACTH AND ANGIOTENSIN II IN THE CONTROL OF CORTICOSTEROID PLASMA CONCENTRATION IN MAN

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SUMMARY

Angiotensin II infusion into normal human subjects increases the plasma concentration of aldosterone and 18-hydroxycorticosterone but leaves those of other corticosteroids unaffected. This contrasts with the response of adrenocortical cells *in vitro*. However, if a low, constant rate infusion of ACTH is maintained throughout increasing rates of angiotensin II infusion, the concentrations of cortisol, corticosterone and 18-hydroxy-11-deoxycorticosterone also correlate positively and significantly with the prevailing angiotensin II concentration. Plasma 11-deoxycorticosterone levels also rise. A possible explanation of this finding is that the presence of ACTH is necessary for a full corticosteroid response to angiotensin II and that infused angiotensin II inhibits ACTH secretion.

INTRODUCTION

Among the factors influencing adrenal secretion of corticosteroids in man, ACTH, angiotensin II, potassium and sodium are probably the most important. Many studies have demonstrated that depletion of sodium enhances the aldosterone response both to angiotensin II [1, 2] and ACTH [3, 4]. However, the interaction of angiotensin II and ACTH has not been as fully examined although it is known that angiotensin II infused into normal human subjects, while raising the plasma concentrations of aldosterone and 18-hydroxycorticosterone, does not affect those of 11-deoxycorticosterone (DOC), corticosterone and cortisol [5, 6]. This may also be the case with 18-hydroxy DOC, although Tuck, Chandler and Mayes [7] have recently published evidence to the contrary. Aldosterone level, compared with other plasma corticosteroid concentrations, is relatively unresponsive to ACTH administration in subjects on a normal sodium intake [8, 9].

In the study described here we have examined the effect of ACTH on the response of plasma corticosteroids to angiotensin II infused into normal human subjects. A preliminary report of these studies has already been made [10].

SUBJECTS AND METHODS

Five normal male volunteers, aged between 22 and 29 years, participated in the study. They were on a normal diet and were not receiving medication of any kind. All experiments began between 0800 and 0900 with the subjects recumbent and fasting. Endogenous ACTH secretion was suppressed by dexamethasone administration, 2 mg nine hours before and 2 mg 1 h before the experiment.

Three subjects were infused on two occasions at least two weeks apart and the sequence of the infusions was varied. On one occasion 1-Asp-NH_2-5-Val-angiotensin II (Hypertensin, CIBA) in 5% dextrose solution was infused into an arm vein at rates of 0, 2, 4 and 8 ng Kg^{-1} min^{-1}, each rate being continued for 1 h as previously described [1]. On the other occasion, a solution of ACTH (Synacthen, CIBA) in 5% dextrose was infused at a constant rate of 0.6 ng Kg^{-1} min^{-1}, for the 4 h of the experiment, and the graded infusion of angiotensin II was superimposed on this. Plastic apparatus was used throughout.

Blood samples were withdrawn via a plastic indwelling cannula in the contralateral arm at the end of each infusion period. The plasma was separated immediately and stored at $-20°$. Angiotensin II was measured by radioimmunoassay [11] and corticosteroids by gas–liquid chromatography [12–14]. Plasma electrolytes were measured by flame photometry.

The remaining two subjects acted as controls and received infusions of ACTH (0.6 ng Kg^{-1} min^{-1}) for 2 h. Blood samples were taken at 20 minute intervals.

RESULTS

1. *Effect of ACTH infusion alone*

ACTH produced a rise in the concentration of all steroids, compared to the suppressed basal levels, up to 1 h after commencement of the infusion (Fig. 1). This was steepest for the steroids produced predominantly by the zona fasciculata of the adrenal cortex: DOC, corticosterone, 18-hydroxy DOC and cortisol. No further rises were seen after one hour; a steady state had probably been achieved.

2. *Effect of angiotensin II infusion alone*

Graded infusion of angiotensin II into dexametha-

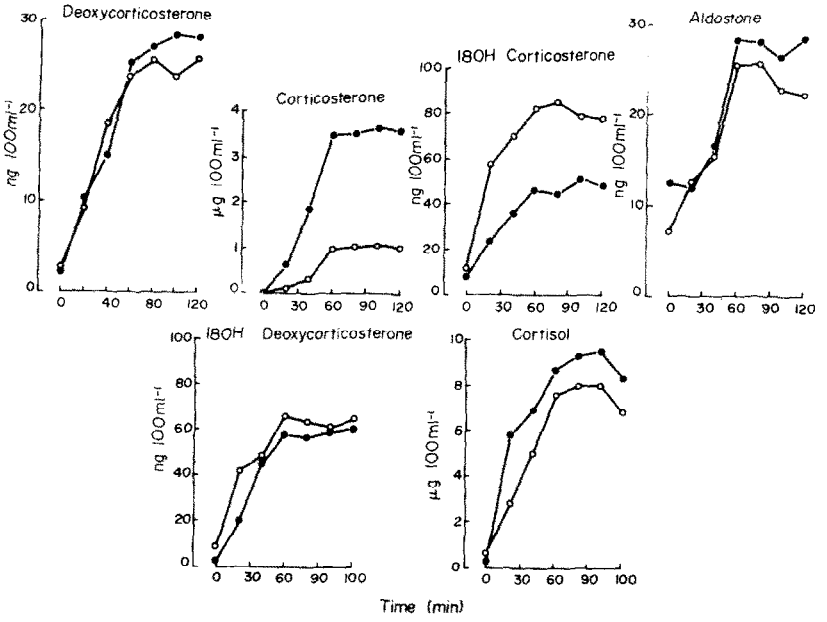

Fig. 1. Effect of a constant rate infusion of Synacthen (0.6 ng Kg^{-1} min^{-1}) on plasma corticosteroid levels of two male subjects. At time 0, their respective corticosteroid concentrations were: deoxycorticosterone 2.1 and 2.6 ng 100 ml^{-1}; corticosterone 0.03 and 0.02 μg 100 ml^{-1}; 18 OH corticosterone 7.4 and 11.7 ng 100 ml^{-1}; aldosterone 12.3 and 7.1 ng 100 ml^{-1}; 18 OH deoxycorticosterone 2.1 and 8.9 ng 100 ml; cortisol 0.2 and 0.6 μg 100 ml^{-1}.

sone-treated normal subjects produced a response pattern (Fig. 2) similar to that previously reported in subjects not receiving dexamethasone [5]. Aldosterone and 18-hydroxycorticosterone concentrations showed a positive and significant correlation ($r = 0.74$, $P < 0.001$ and $r = 0.70$, $P < 0.01$, respectively) (Table 1) with concurrent angiotensin II plasma levels.

3. Effect of simultaneous ACTH and angiotensin II infusions

Graded infusion of angiotensin II into subjects already receiving a constant rate infusion of ACTH raised the plasma levels of all corticosteroids studied (Fig. 3). Positive and significant correlations were obtained between the plasma angiotensin II concentration and not only aldosterone and 18-hydroxycor-

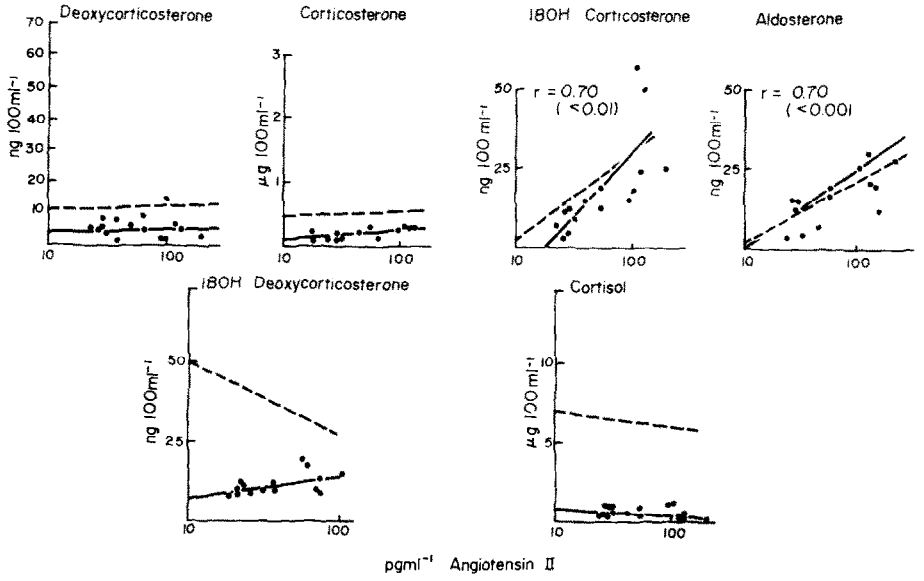

Fig. 2. Relationship between plasma concentrations of angiotensin II and individual corticosteroids, with (solid line) and without (broken line) treatment with dexamethazone. The lines are linear regressions of the pooled data from three subjects. The control regression (broken line) is taken from Mason et al. [5].

Table 1. Effect of dexamethazone and synacthen on the dose-response relationships of angiotensin II and individual corticosteroids

Steroid	Dexamethasone				Dexamethasone + Synacthen			
	b*	a	r†	P	b	a	r	P
DOC	1.4	8.2	−0.08	NS	13.2	19.0	0.23	NS
Corticosterone	0.26	−0.29	0.39	NS	2.8	0.9	0.72	<0.001
18 OH Corticosterone	36.1	−44.2	0.70	<0.01	44.7	−29.5	0.91	<0.001
Aldosterone	19.6	−18.0	0.74	<0.001	25.5	−18.0	0.84	<0.001
18 Hydroxy DOC	3.81	4.9	0.32	NS	71.2	−27.5	0.71	<0.01
Cortisol	1.1	−0.2	−0.15	NS	6.4	5.5	0.58	<0.02

* $y = bx + a$ where y = steroid concentration.
x = log angiotensin II concentration, † correlation coefficient.

ticosterone, but also 18-hydroxy DOC, corticosterone and cortisol ($r = 0.71$, $P < 0.01$, $r = 0.72$, $P < 0.001$ and $r = 0.58$, $P < 0.02$, respectively) (Table 1). Plasma DOC levels showed a positive correlation ($r = 0.32$) with angiotensin II concentration but statistical significance was not achieved. However, a rise of plasma DOC was noted in all cases in response to the angiotensin II infusion.

DISCUSSION

In man, the zona fasciculata of the adrenal cortex synthesises 17α-hydroxycorticosteroids, a large proportion of the DOC, 18-hydroxy DOC and corticosterone, and at least some of the 18-hydroxycorticosterone [15]. The zona glomerulosa produces aldosterone and also contributes small quantities of other 17-deoxycorticosteroids such as 18-hydroxycorticosterone [15]. Thus, apart from the 17α-hydroxycorticosteroids and aldosterone, other products may have a dual origin. Interpretation of changes in plasma concentration in terms of altered activity of a particular biosynthetic pathway may be difficult but still of value [16].

The concentrations of cortisol, corticosterone [17, 18], DOC [19, 20, 21], 18-hydroxy DOC [13, 22] and 18-hydroxycorticosterone [14] respond to ACTH whether infused or endogenously secreted, whereas aldosterone is relatively insensitive to this hormone. Their ACTH:steroid dose-response curves have been compared by Fraser *et al.* [9]. Aldosterone secretion and ACTH levels are more closely related in sodium depleted subjects [23] and in subjects with Conn's syndrome [24]. The apparent association between the diurnal and episodic variations of ACTH and aldosterone levels has been fully discussed elsewhere [25, 26].

In the current experiments, elimination of endogenous ACTH failed to suppress aldosterone levels but subsequent ACTH infusion gave a brisk response (Fig. 1). In the case of all the compounds measured, a plateau of response had been reached by the time angiotensin II was infused.

Angiotensin II *in vivo* in man appears to be a specific stimulus to aldosterone and 18-hydroxycorticosterone secretion (see also 5, 6), although in preparations of rat adrenocortical cells incubated *in vitro*

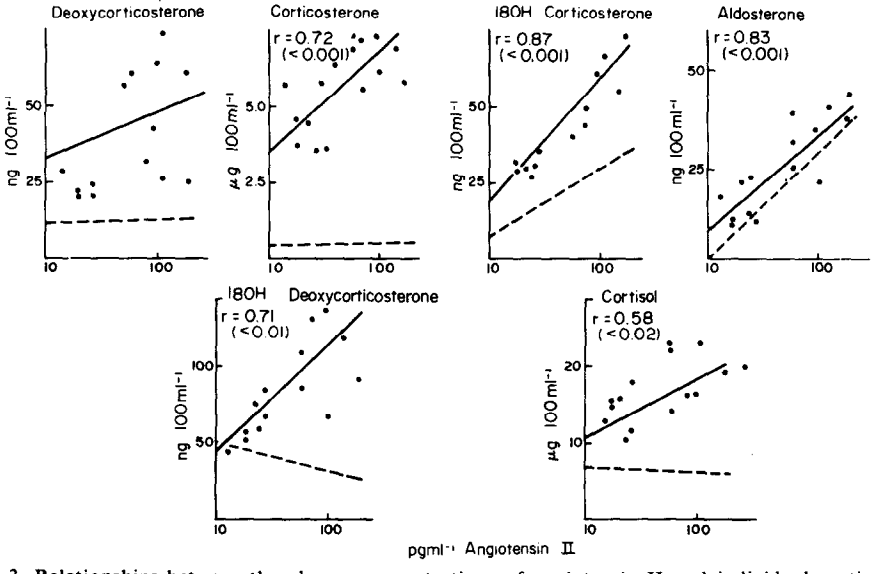

Fig. 3. Relationships between the plasma concentrations of angiotensin II and individual corticosteroids, with (solid line) and without (broken line) simultaneous infusion of Synacthen. The lines are linear regressions. The control regression (broken line) is taken from Mason *et al.* [5].

other products of the zona glomerulosa [27] and the zona fasciculata [28, 29] also respond to the octapeptide. Tan and Mulrow [30] report that DOC levels also respond in dexamethasone-treated human subjects but this has not been confirmed by the current experiments or in other reports. The discrepancy between the response of the adrenal cortex *in vivo* and *in vitro* has not yet been adequately explained. The suggestion that a failure of cortisol to respond to angiotensin II is due to a compensatory fall in ACTH levels brought about by a transient increase in cortisol concentration (i.e. the negative feedback mechanism) is untenable since no cortisol response occurs even when ACTH release is prevented. However, the results of the experiment in which ACTH levels were maintained by infusion (Fig. 3) reveal dramatically that, in these circumstances, angiotensin II elicits a more general response in corticosteroid secretion. From these experiments it can be inferred:

(a) that angiotensin II is capable of exerting an effect at least as early as 21 hydroxylation, since levels of DOC are affected;

(b) that the presence of ACTH is necessary for this response.

Arising from (b) it seems probable that the reason for the lack of response in the absence of exogenous ACTH [6] may be an inhibitory effect of angiotensin II on ACTH and this has been confirmed by a series of experiments in which plasma ACTH levels were found to be inversely correlated with plasma angiotensin II concentration during angiotensin II infusion [10, 16, 31].

The action of angiotensin II on the early biosynthesis of corticosteroids, demonstrated by *in vitro* studies of adrenal tissue [27, 32, 33] is to some extent confirmed by these *in vivo* studies using acute increases of angiotensin II. ACTH is known to act at an early stage of corticosteroid biosynthesis, probably at or before the cleavage of the cholesterol side chain [34, 35]. That ACTH is essential for this more general effect of angiotensin II may possibly mean that the locus of action of the octapeptide is later than this. Preliminary studies have suggested that this locus is the 3β-hydroxysteroid dehydrogenase-isomerase catalysed reactions [36] but further studies are obviously required.

These studies go some way to explaining the discrepancy between the results of *in vitro* and *in vivo* studies of the control of corticosteroid secretions in man. They reveal a previously unreported interaction between ACTH and angiotensin II, the physiological significance of which remains to be discovered.

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