In vivo Evidence of Regulation by Pituitary-Adrenal Axis of Urinary Epinephrine Excretion in Men

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Abstract

The effect of the pituitary-adrenal axis on epinephrine synthesis in the human adrenal medulla was examined by the estimation of the 24-h urinary epinephrine level after treatment with glucocorticoids in four patients with systemic lupus erythematoses (SLE), one patient with rheumatoid arthritis (RA) and one patient with adrenal pheochromocytoma. 24-h urinary catecholamines (CAs) were measured by HPLC before and after glucocorticoid treatment, dexamethasone or predonisolone was orally given for more than seven days to patients with SLE, RA or isloated ACTH deficiency and five days to a patient with adrenal pheochromocytoma.

In patients with isolated ACTH deficiency, the 24-h urinary epinephrine level was significantly lower than the normal range. In patients with SLE or RA, the 24-h urinary epinephrine level was normal and it was significantly suppressed by therapeutic doses of prednisolone 30-40 mg/day. In a patient with adrenal pheochromocytoma, 24-h urinary epinephrine was extremely high and it was significantly increased after dexamethasone 0.5 mg/day.

These results suggest that epinephrine synthesis in the human adrenal medulla may be dependent on the pituitary-adrenal axis. But the increase in epinephrine synthesis due to dexamethasone in a patient with pheochromocytoma may reflect the direct effect via the feeding artery to the tumor, as previously shown in an in vitro culture system.

Glucocorticoid secreted from the adrenal cortex into the medulla via the adrenal portal veins (Harrison et al., 1960) plays an important role in the regulations of CA biosynthesis. From a detailed study of dopamine-β-hydroxylase (DBH) (Ciaranello et al., 1976) and phenylmethanolamine-N-methyltransferase (PNMT) (Ciaranello et al., 1978) in CA biosynthesis respectively, it is believed that glucocorticoid maintains the levels of DBH and PNMT, synthesized transsynaptically, by inhibiting enzyme degradation. Induction of tyrosine hydroxylase (TH) by glucocorticoid in the rat’s adrenal medulla was reported negative (Axelrod, 1977) or positive by Guidotti et al. (1975). The direct effects of glucocorticoid on CA enzyme induction and CA products have been demonstrated in cultured bovine adrenal chromaffin cells (Hersey & Distefano, 1977; Yanase et al., 1984), rat pheochromocytoma cells (Lucas & thoenen, 1977; Schbert et al, 1980; Tishler et al, 1983) and human pheochromocytoma cells.

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(Yanase et al., 1984). ACTH also regulates CA synthesis not only by the mediation of adrenocortical glucocorticoid but also by the direct effect of maintaining TH activity in the rat adrenal medulla (Mueller et al., 1970; Wurtman et al., 1972). These studies of hormonal regulation of CA biosynthesis are almost all limited to rats. There have been only a few reports on human beings in this field. Urinary excretion of epinephrine arises almost entirely from the adrenal medulla, while that of norepinephrine originates both in sympathetic nerve terminals and the adrenal medulla (Euler et al., 1954). Patients with isolated ACTH deficiency are analogous to hypophysectomized rats. Clinically, the commonest cause of ACTH deficiency is the administration of glucocorticoid. To evaluate the effect of the pituitary adrenal axis on CA synthesis in the human adrenal medulla, we focused on the changes in 24-h urinary excretion of epinephrin in patients with isolated ACTH deficiency, SLE or RA and a patient with adrenal pheochromocytoma before and after glucocorticoid administration.

Materials and Methods

Materials and protocols

Brief outlines of materials are shown in Table 1. Four patients with SLE (cases 5 to 8, five female, aged 23–56 yr) and one patient with RA (case 9, female, 33 yr) were tested as ones with normal adrenal function. The above patients were virgin cases and they were clinically and serologically diagnosed and all had indication for glucocorticoid therapy. They were serologically normal in liver and renal function. Four patients with isolated ACTH deficiency (case 1–4, one male and three females, aged 36–62 yr) were tested as ones with adrenocortical insufficiency. They were serologically normal in liver and renal function. Four patients with isolated ACTH deficiency (case 1–4, one male and three females, aged 36–62 yr) were tested as ones with adrenocortical insufficiency. All were virgin cases and diagnosed on the basis of findings of low plasma ACTH and low serum cortisol which responded to exogenous ACTH, and other pituitary hormones normal as to both basal and stimulated levels. The basal level of serum cortisol determined by RIA (SRL, Special Reference Laboratory, Tokyo) in case 1 was 0.32 µg/dl and those in cases 2, 3 and 4 were not detected. Basal levels of plasma ACTH determined by RIA (SRL) in cases 1, 2, 3 and 4 were less than the minimal sensitive concentration of 20 pg/ml. Because of their symptoms indicating adrenal insufficiency, they had indications for glucocorticoid replacement. A patient with adrenal pheochromocytoma (case 10, female, 24 yr) was also tested following informed consent to the giving of glucocorticoid. She was diagnosed as MEN IIb and the operation revealed a left adrenal pheochromocytoma which weighed 25 g. Tissue epinephrine and norepinephrine content figures were 11.8 µg/mg and 8.21 µg/mg, respectively. All medications with glucocorticoids were orally given. Patients with SLE or RA were given prednisolone (Shionogi) 30–40 mg/day every day for 7–14 days. Patients with isolated ACTH deficiency were given 0.5 mg/day dexamethasone (MERCK & CO., INS) every day for 7–10 days.

A patient with pheochromocytoma was given 0.5 mg/day dexamethasone for five days. 24-h urinary excretion of CAs were measured for serial 3–4 days before and after the administration of glucocorticoids. All data reported here were mean ± SD for serial three day estimation.

CA determination

Urinary CAs were determined by HPLC (Hitachi 635A) and Trihydroxyindol (THI) reaction in collaboration with SRL (Special Reference Laboratory, Tokyo). HPLC was carried out on a Nucleosil C18 column (0.4 × 15 cm). A sample of 24-h urine was collected in a glass bottle containing 20 ml of 6N HCl. Twenty ml

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Age/Sex</th>
<th>Disease</th>
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<tbody>
<tr>
<td>1</td>
<td>62/M</td>
<td>Isolated ACTH deficiency</td>
</tr>
<tr>
<td>2</td>
<td>36/F</td>
<td>Isolated ACTH deficiency</td>
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<tr>
<td>3</td>
<td>60/F</td>
<td>Isolated ACTH deficiency</td>
</tr>
<tr>
<td>4</td>
<td>46/M</td>
<td>Isolated ACTH deficiency</td>
</tr>
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<td>5</td>
<td>52/F</td>
<td>SLE</td>
</tr>
<tr>
<td>6</td>
<td>56/F</td>
<td>SLE</td>
</tr>
<tr>
<td>7</td>
<td>54/F</td>
<td>SLE</td>
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<tr>
<td>8</td>
<td>23/F</td>
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</tr>
<tr>
<td>9</td>
<td>33/F</td>
<td>RA</td>
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<tr>
<td>10</td>
<td>24/F</td>
<td>Adrenal Pheochromocytoma</td>
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aliquot of the urine was passed through an activated alumina column (Woelm Neutral W-200) according to the method by Ueda et al. (1977) and eluted with 0.1 M phosphate buffer (pH 2.9, flow rate 0.6 ml/min). Activation of alumina was done by the method of Anton and Sayre (1962). The elutes were faced with the necessary reagents for formation of the fluorescent THI derivatives, namely 500 mM K₃HPO₄, followed by 0.1% K₃Fe(CN)₆, 0.1% ascorbic acid and 5N NaOH, and measured with a fluorescence detector (Hitachi, 650-10 LC). The sensitivity of the assay was 10 pg/ml. Normal ranges of 24-h urinary epinephrine in 4228 subjects were 3.0-15.0 µg/day (9.0±6.0, Mean±2SD).

Statistical methods
The statistical significance of the data was assessed with Student's t-test.

Results

Individual basal levels and responses of 24-h urinary epinephrine after administration of glucocorticoid in patients with isolated ACTH deficiency and in patients with SLE or RA are shown in Fig. 1 and Fig. 2, respectively. The statistical comparisons of 24-h urinary epinephrine levels before and after glucocorticoid administration were based on the mean level of 24-h urinary epinephrine for serial 3-4 days. In patients with isolated ACTH deficiency, the basal level of urinary epinephrine (3.38±1.64 µg/day, Mean±SD) was quite low compared with the normal range (9.0±3.0 µg/day) and it was significantly increased to 7.16±2.74 µg/day by replacement with dexamethasone 0.5 mg/day (P<0.05). In patients with SLE or RA, the basal level of 24-h urinary epinephrine (8.36±2.84 µg/day) was within the normal range and it was significantly suppressed to 3.93±1.38 µg/day by the administration of prednisolone 30-40 mg/day (Fig. 3). In a patient with adrenal pheochromocytoma, the basal level of urinary epinephrine (143.8±4.5 µg/day) was extremally high and it was significantly in-

Fig. 1. Individual 24-h urinary epinephrine levels for serial 3-4 days before and after administration of dexamethasone (Dex) 0.5 mg/day in patients with isolated ACTH deficiency (cases 1-4)
Discussion

In patients with isolated ACTH deficiency, the basal level of 24-h urinary epinephrine was significantly low. This result supports the theory of plasma epinephrine deficiency in hypocorticotropic hypopituitary children (Rudman et al., 1981), while Luft and Euler (1956) reported that the daily urinary excretion of epinephrine in 20 cancer patients was not altered by hypophysectomy. But a blunted increase in urinary epinephrine during insulin hypoglycemia was reported in 20 patients with hypophysectomy (Luft & Euler, 1956) and a 2-year boy with ACTH deficiency (Hung & Migeon, 1968). These results strongly indicate that the pituitary-adrenal axis plays an important role in maintaining epinephrine synthesis in man. In our data, the replacement dose of glucocorticoid significantly increased the diminished level of urinary epinephrine in patients with isolated ACTH deficiency. However, more elaborate examinations may be needed to draw a conclusion because the previous report showed that decreased epinephrine synthesis in hypophysectomized rats was unresponsive to a replacement dose of glucocorticoid, but return to a normal level took place after massive amounts of glucocorticoid were given (Mueller et al., 1970; Wurtman et al., 1972). Long term treatment with prednisolone in patients with SLE or RA significantly suppressed the 24-h urinary epinephrine excretion. Endogenous glucocorticoid produced under the influence of pituitary ACTH is about 100 times more potent than its exogenous effect in maintaining PNMT activity in the adrenal
medulla (Wurtman et al., 1972). In addition, PNMT activity in men with normal adrenocortical function may be already maximum because we failed to prove an increase in urinary epinephrine in two patients with Cushing’s disease (7.84 ± 2.14 µg/day, 6.78 ± 2.23 µg/day, Mean ± SD.). Accordingly, our results in patients with SLE or RA may be explained by the suppression of ACTH and cessation of action of endogenous glucocorticoid despite the increase in serum prednisolone concentration.

On the other hand, five day’s treatment with dexamethasone 0.5 mg/day significantly
increased the urinary epinephrine in a patient with adrenal pheochromocytoma despite the suppression of the pituitary-adrenal axis. Because this tumor is large enough to compress the adrenal cortex, this raises the question whether normal perfusion through veins from the adrenal cortex to the tumor may exist. Our results are probably due to the direct effect of dexamethasone via the feeding artery to the tumor, as previously shown in an \textit{in vitro} culture system (Yanase \textit{et al.}, 1984).

In summary, the regulation, by the pituitary-adrenal axis of epinephrine synthesis was suggested in the human adrenal medulla \textit{in vivo}. In adrenal pheochromocytoma, the regulation of epinephrine synthesis by glucocorticoid seemed to be less dependent on the pituitary-adrenal axis and rather controlled via the feeding artery to the tumor.

**Acknowledgement**

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**References**


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