Effect of Spironolactone on Urinary Excretion of Immunoreactive Prostaglandin E in Essential Hypertension and Primary Aldosteronism

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To determine the effect of aldosterone antagonist on renal prostaglandin E synthesis, the rate of urinary excretion of immunoreactive prostaglandin E was measured radioimmunologically before and during the oral administration of an aldosterone antagonist, spironolactone, in 5 patients with essential hypertension, 3 with primary aldosteronism and 2 with postoperative primary aldosteronism. Spironolactone was administered at an oral dose of 25 mg 4 times daily for about 1 week. In the control state, the rates of urinary prostaglandin E excretion ranged from 151 ng/day to 4,527 ng/day in essential hypertension. The rates were not augmented in primary aldosteronism but decreased after the removal of an aldosterone producing adenoma. No obvious relationship was observed between plasma aldosterone concentration and the rate of urinary prostaglandin E excretion. On the first day of spironolactone administration, the excretion rates of urinary prostaglandin E were markedly increased independently of basal plasma aldosterone level in all cases except one case of essential hypertension. Urinary prostaglandin E excretion was increased with the concomitant increase of urinary Na/K ratio in essential hypertension and primary aldosteronism. After the second day, the augmented urinary prostaglandin E excretion was decreased and the changes of urinary prostaglandin E excretion varied from case to case. These results suggest that synthesis of renal prostaglandin E is not mainly regulated by aldosterone in essential hypertension and primary aldosteronism. — urinary prostaglandin E; plasma aldosterone level; spironolactone; essential hypertension; primary aldosteronism

There are several reports that indicate the important roles of renal prostaglandins in the regulation of renal blood flow and sodium excretion (McGiff and Vane 1975; Lee et al. 1976; Anderson et al. 1976). Recently, McGiff et al. (1972, 1976) reported that another renal natriuretic substance, kinin, increased the synthesis of renal prostaglandins and that the effects of renal kallikrein-kinin system might be modified by the induced renal prostaglandins. On the other hand, Margolius et al. (1974a, b, 1976) reported that the renal kallikrein-kinin system was regulated by

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aldosterone. Moreover, there are some reports that in primary aldosteronism the enhanced urinary kallikrein excretion could be reduced by the administration of aldosterone antagonist (Margolius et al. 1974b; Seino et al. 1977). These results suggest the interaction between mineralocorticoid hormones, prostaglandins and the kallikrein-kinin system.

In this paper, we intended to demonstrate the effect of aldosterone on renal prostaglandin E synthesis by means of administration of aldosterone antagonist, spironolactone.

**Patients and Methods**

Five patients with essential hypertension (EH, 2 men and 3 women, 27–45 years old), 3 with primary aldosteronism (PA, a man and 2 women, 34–47 years old) and 2 with postoperative PA (a man and a woman, 34 and 47 years old) were included in this study. The diagnosis of EH was determined after a series of examinations including routine laboratory tests, intravenous pyelography, radioisotope renography, renoscintigraphy, renal arteriography and determination of plasma aldosterone, 11-OHCS and urinary catecholamines. There was no evidence of severe hypertensive complications in cardiovascular renal system. The patients with PA had persistent hypokalemia, suppressed plasma renin activity (PRA) and increased plasma aldosterone concentration (PAC). Afterward, they were cured by removal of an aldosterone producing adenoma and 2 of the 3 patients were studied again 55 and 39 days after the operation, respectively (Cases No. 6 and No. 7). All patients were studied in Tohoku University Hospital and were allowed to take unrestricted diets during the study period. Antihypertensive medication was discontinued at least 2 weeks before study.

After the control period, spironolactone was administered at an oral dose of 25 mg 4 times daily for 7–8 days in EH and PA and for 5 days in postoperative PA. Twenty-four-hour-urine samples were collected in a refrigerator at 4°C for the determinations of Na, K and prostaglandin E (PGE). Urine samples were stored at -15°C until PGE radioimmunoassay. Blood sampling for PRA and PAC was done with the fasting subjects after at least 2 hr's recumbent position in the morning in the control state.

Urinary PGE was measured radioimmunologically with commercial assay kits (CA 501, Clinical Assays, Cambridge, Mass. 02142). Two ml of urine sample were used for PGE determination. For the chemical conversion of PGE to PGB, 2 ml of 1 N KOH were added to each urine sample with shaking for 1 hr (Zusman 1972). Then the mixture was acidified with 1 N HCl and extracted with 15 ml of ethylacetate. After the organic solution was dried, the residue was applied to a mini-silicic acid column and the PGB fraction was eluted with 4 ml of the solvent (benzene: ethylacetate, 60:40, by vol, Coladwell et al. 1971; Auletta et al. 1974). The separated PGB was radioimmunologically assayed using PGB, antiserum. PRA was measured by radioimmunooassay (Abe et al. 1972). PAC was measured by radioimmunoassay kits (Midorijuji, Japan). Urinary electrolytes were determined by an autoanalyzer.

**Results**

Table 1 shows the basal clinical data and the values of urinary PGE excretion during the administration of spironolactone in each case.

**Urinary PGE excretion in the control state**

In the control state, the rates of urinary PGE excretion were widely distributed from 151 ng/day to 4,527 ng/day in EH. Two cases of EH (Cases 4 and 5) showed
TABLE 1. Basal clinical data and the values of urinary PGE excretion during the administration of spironolactone. Phase I, phase II, phase III and phase IV represent the states on the first day, 2nd-3rd days, 4th-5th days and 6th-8th days after spironolactone administration, respectively. Urinary PGE excretion rate is presented as an actual value or the mean value of the urinary PGE output in each phase including control state.

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Diagnosis</th>
<th>Mean BP (mmHg)</th>
<th>PRA (ng/ml)</th>
<th>PAC (ng/100 ml)</th>
<th>Urinary Na excretion (mEq/day)</th>
<th>Urinary PGE excretion (ng/day)</th>
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<tr>
<td>1</td>
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<td>135</td>
<td>7.4</td>
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<td>117</td>
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<td>126</td>
<td>4.7</td>
<td>5.8</td>
<td>159</td>
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</tbody>
</table>

Abbreviations; EH, essential hypertension; PA, primary aldosteronism; postop. PA, postoperative primary aldosteronism; PRA, plasma renin activity; PAC, plasma aldosterone concentration.

Fig. 1. Changes in urinary PGE excretion, urine volume, urinary sodium excretion and urinary Na/K ratio after spironolactone administration in 3 patients with essential hypertension. Phase I, phase II, phase III and phase IV are as designated in Table 1.
Fig. 2. Changes in urinary PGE excretion, urine volume, urinary sodium excretion and urinary Na/K ratio after spironolactone administration in 3 patients with primary aldosteronism. Phase I, phase II, phase III and phase IV are as designated in Table 1.

exceedingly high values of urinary PGE output. In PA, the rates of urinary PGE excretion were not augmented. In Case 6 the values were 361 ng/day and 540 ng/day (mean 450 ng/day) in the control state and decreased to 173 ng/day and 203 ng/day (mean 188 ng/day) after the removal of an aldosterone producing adenoma. In Case 7 the rates were 179 ng/day, 250 ng/day, 309 ng/day and 493 ng/day (mean 307 ng/day) in the control state and 159 ng/day after the operation.

Sodium excretion rate ranged from 122 mEq/day to 301 mEq/day and no obvious relationship was observed between sodium excretion and urinary PGE excretion. There was either no obvious relationship between PAC and urinary PGE excretion.

Effect of spironolactone on urinary PGE excretion

On the first day of spironolactone administration the excretion rates of urinary PGE markedly increased independently of basal PAC in all cases except one case of essential hypertension (Case 5). After the second day, the augmented urinary PGE excretion decreased except in Cases 5 and 6. At the end of the study, urinary PGE excretion was increased in 2 cases of EH (Cases 2 and 4) and 2 cases of PA
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Fig. 3. Changes in urinary PGE excretion, urine volume, urinary sodium excretion and urinary Na/K ratio after spironolactone administration in 2 patients with postoperative primary aldosteronism.

(Cases 6 and 8) and was decreased in 2 cases of EH (Cases 1 and 5) and 1 case of PA (Case 7), compared with the values in the control state. In 2 cases of postoperative PA, urinary PGE excretion was obviously increased during the spironolactone administration.

Figs. 1, 2 and 3 illustrate the changes in urinary PGE excretion, urine volume, urinary sodium excretion and urinary Na/K ratio in patients with EH, PA and postoperative PA, respectively. The data from Cases 4 and 5 were excluded from the illustration in Fig. 1, because the control levels of urinary PGE excretion were exceedingly high in these cases. In EH, the changes in urine volume, urinary sodium excretion and urinary Na/K ratio were similar to those of urinary PGE excretion (Fig. 1). The parameters increased on the first day and decreased after the second day of spironolactone. In PA, urinary Na/K ratio was lower than that in the EH in the control state and markedly increased on the first day with the augmentation of urinary PGE excretion but was not decreased after the second day of the study. Urine volume and urinary sodium excretion were not obviously changed (Fig. 2). In postoperative PA, no obvious change was observed in urine volume, urinary sodium excretion and urinary Na/K ratio during the period of the study (Fig. 3).
Fig. 4 illustrates the mean changes in urinary PGE excretion, urine volume, urinary sodium excretion and urinary Na/K ratio in EH, PA and postoperative PA. Urinary PGE excretion significantly increased on the first day of the spironolactone medication ($p<0.001$).

**DISCUSSION**

Frolich et al. (1972, 1975) identified PGE and PGF in human urine and they suggested that urinary PGs reflect the synthesis of PGs in the kidney. In this study, we measured the urinary excretion rate of immunoreactive PGE as an indicator of renal PGE synthesis.

Recently, Nasjletti and Colina-Chourio (1976) reported that deoxycorticosterone and aldosterone increased the urinary excretion of kallikrein and PGE and that the augmented urinary PGE excretion was reduced by the administration of kallikrein inhibitor, aprotinin. They suggest that aldosterone may regulate the renal PGE synthesis through the regulation of renal kallikrein-kinin system. Some investigators reported that spironolactone, an aldosterone antagonist, reduced the
augmented urinary kallikrein excretion in PA and suggested that renal kallikrein-kinin system is regulated by the renal effect of aldosterone (Margolius et al. 1974b; Seino et al. 1977).

On the first day of this study, spironolactone increased the urinary excretion of PGE and urinary Na/K ratio which is an indicator of the effect of the aldosterone antagonist on the renal tubules. The increase of urinary PGE excretion and the effect of spironolactone on urinary electrolytes occurred at the same time in EH and PA. In postoperative PA, urinary PGE excretion increased without an augmentation of urinary Na/K ratio. Basal plasma aldosterone level was not related to the increase of urinary PGE excretion after the administration of spironolactone. These findings suggest that the effect of spironolactone on renal PGE synthesis occurs independently of aldosterone antagonistic effect of spironolactone and that spironolactone may have a direct effect on renal PGE synthesis.

Hofmann et al. (1972) reported that aspirin, a specific inhibitor of prostaglandin synthesis (Vane 1971), inhibited the natriuretic response of spironolactone but not of hydrochlorothiazide in mineralocorticoid treated dogs and rats. Tweeddale and Ogilvie (1973) reported the same observation in man. In their paper, aspirin antagonized the natriuretic effect of spironolactone in man. Previously, Elliot (1962) suggested that aspirin inhibits the mineralocorticoid blocking effect of spironolactone through the competition on the receptor sites in the target tissues. Feldman and Couropmitree (1976) also suggested that the non-steroidal anti-inflammatory drugs have an affinity for receptors mediating mineralocorticoid agonist activity. In the present study, spironolactone increased urinary excretion of PGE with the concomitant increase in urinary Na/K ratio, urine volume and urinary Na excretion. This suggests that spironolactone may have a direct effect on renal PGE synthesis and that the augmented renal PGE may participate in the diuresis observed after spironolactone administration. At this time, we cannot explain why the increment of urinary PGE excretion did not persist throughout the study period and the urinary PGE output fluctuated so much after the second day of spironolactone administration.

In the control state, urinary PGE excretion was not augmented in the patients with PA. There was no obvious correlation between urinary PGE and plasma aldosterone level in the control state in EH, PA and postoperative PA. Tan and Mulrow (1977) reported markedly suppressed urinary PGE excretion in 4 patients with PA. From these findings, we suppose that on unrestricted diet, renal PGE synthesis is not mainly regulated by plasma aldosterone level. However, in PA urinary PGE excretion decreased after the removal of an aldosterone producing adenoma. So, there is a possibility that the increased plasma aldosterone may participate in the basal synthesis of renal PGE in PA.

Two cases of EH showed exceedingly high rates of urinary PGE excretion in this study. Tan and Mulrow (1977) recently reported a group of EH in which urinary PGE excretion was markedly suppressed. We suppose that patients with
EH may be divided into some subgroups with regard to urinary PGE excretion. It may be concluded that spironolactone increases renal PGE synthesis independently of plasma aldosterone level and that renal PGE synthesis is not mainly regulated by aldosterone in EH and PA.

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