SHORT COMMUNICATION

Oestrogen hypertension in rats

T. SARUTA, R. NAKAMURA, I. SAITO, K. KONDO AND S. MATUKI
Department of Internal Medicine, University of Keio School of Medicine,
Shinanomachi, Tokyo, Japan

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Summary

1. A large dose of oestrogen elevated the blood pressure in male Wistar rats.
2. Plasma renin substrate and renin activity increased significantly but plasma renin concentration was unchanged.
3. The increase in blood pressure induced by oestrogen was significantly reduced by salt loading, plasma renin concentration was suppressed and the increase in plasma renin activity was reduced.
4. The increase in plasma renin activity induced by the increase of plasma renin substrate concentration may play a role in oestrogen-induced elevation of blood pressure.

Key words: blood pressure, oestrogen, renin-angiotensin system.

Introduction

Since the first report of hypertension induced by oral contraceptives (Brownrigg, 1962), numerous reports (Laragh, Sealey, Ledingham & Newton, 1967; Woods, 1967; Tyson, 1968; Weinberger, Collins, Dowdy, Nokes & Leutscher, 1969; Skinner, Lumbers & Symonds, 1969; Saruta, Saade & Kaplan, 1970) have suggested that the administration of oral contraceptives containing oestrogens leads to the development of hypertension in women. The mechanism by which oral contraceptives containing oestrogen cause hypertension is still controversial and alterations in the renin-angiotensin system induced by oestrogen may play a role. Skinner et al. (1969) suggested that failure to suppress renin secretion adequately in the presence of an oestrogen-induced increase in renin substrate may contribute to the development of hypertension. Our previous findings in women taking oral contraceptives (Saruta et al., 1970) supported this hypothesis.

In this study, the effects of oestrogen upon blood pressure in male Wistar rats were studied and components of the renin-angiotensin system measured.

A part of this work was presented at the fifth Asia and Oceania Congress of Endocrinology at Chandigarh, India, 1974.

Methods

Two series of identical experiments (A and B) were performed with seventy-five male Wistar-strain rats weighing 130–150 g. In each experiment the rats were divided into four groups. Animals of group 1 were fed on stock chow diet and given tap water to drink. No treatment was given. Animals of group 2 were fed in the same way and received daily an intramuscular injection of oestrogen (5 μmol/kg; 1.5 mg/kg) and an intraperitoneal injection of stilboestrol (56 μmol/kg; 15 mg/kg). Animals of group 3 were fed on stock chow diet and given NaCl solution (170 mmol/l) as a drinking solution in order to suppress renin production. They also received daily injections of oestradiol and stilboestrol disulphate in the same amounts as group 2 animals. Animals of group 4 were fed on stock chow diet and given
NaCl solution (170 mmol/l) as a drinking solution. The systolic pressure and body weight of all the animals were checked at intervals of 3 or 4 days. All animals were killed by decapitation after 15 days and blood was taken for renin assay.

Systolic blood pressure was measured by the modified tail-water plethysmographic method (Okamoto & Aoki, 1963); three readings from each rat were averaged.

Plasma renin activity, plasma renin concentration and plasma renin substrate were determined by the method of Skinner (1967). For determination of plasma renin concentration it was necessary to dialyse plasma to pH 2-8 instead of pH 3-3 against a citric acid–phosphate buffer in order to destroy the high concentrations of renin substrate completely. In spite of this, renin was not denatured or lost. A uniform renin substrate for the measurement of plasma renin concentration was prepared from a 24 h nephrectomized rat plasma. This was shown to be free from angiotensinase by the survival of not less than 90% of added angiotensin after 24 h at 37°C and pH 7-5, and this gave, in every case, a zero-order enzymatic reaction. The standard rat renin for the measurement of plasma renin substrate was prepared by the method of Lever, Robertson & Tree (1964). It was shown to be free from both substrate and angiotensinase, and the addition of the standard renin to angiotensinase-free plasma in a ratio of 4:1 at pH 7-5 and 37°C produced a maximum yield of angiotensin within 15 min.

The significance of differences between mean values was calculated by using Student’s *t*-test.

**Results**

Changes in body weight and systolic blood pressure are shown in Table 1 together with the results of assay of the renin components. The oestrogen-treated groups, regardless of salt loading, showed a significantly slower rate of growth. The untreated control group showed an average increase in weight of 58% in 15 days, and the oestrogen-treated group showed an average increase in weight of 18%.

A significant increase of mean systolic pressure was observed in the oestrogen-treated group in the two different experiments (Table 1). In the oestrogen plus salt-loading group, systolic pressure also tended to increase, but in both experiments the extent of the increase was significantly less than that in the group

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Number</th>
<th>Weight (g) Before</th>
<th>After</th>
<th>Blood pressure (mmHg) Before</th>
<th>After</th>
<th>Plasma renin (pmol h⁻¹ ml⁻¹) Activity</th>
<th>Concentration</th>
<th>Renin substrate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Expt. A</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>10</td>
<td>141 ±8</td>
<td>207±8</td>
<td>125±7</td>
<td>127±9</td>
<td>3.9 ±0.3</td>
<td>9.5±1.0</td>
<td>353±54</td>
</tr>
<tr>
<td>Oestrogen</td>
<td>10</td>
<td>141±2</td>
<td>160±9</td>
<td>125±8</td>
<td>142±7</td>
<td>6.9±1.0</td>
<td>8.6±1.2</td>
<td>929±154</td>
</tr>
<tr>
<td>Oestrogen + saline</td>
<td>10</td>
<td>140±3</td>
<td>165±6</td>
<td>122±13</td>
<td>130±6</td>
<td>4.5±0.8</td>
<td>6.7±1.2</td>
<td>950±88</td>
</tr>
<tr>
<td>Saline alone</td>
<td>10</td>
<td>140±8</td>
<td>194±15</td>
<td>128±5</td>
<td>132±8</td>
<td>3.8±0.4</td>
<td>7.7±0.7</td>
<td>419±43</td>
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<tr>
<td>Expt. B</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>5</td>
<td>122±2</td>
<td>211±2</td>
<td>117±7</td>
<td>123±6</td>
<td>4.3±0.5</td>
<td>8.7±0.4</td>
<td>400±24</td>
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<tr>
<td>Oestrogen</td>
<td>10</td>
<td>122±4</td>
<td>150±6</td>
<td>118±4</td>
<td>152±12</td>
<td>6.1±1.3</td>
<td>8.6±1.1</td>
<td>866±72</td>
</tr>
<tr>
<td>Oestrogen + saline</td>
<td>10</td>
<td>124±4</td>
<td>158±11</td>
<td>118±10</td>
<td>139±9</td>
<td>4.8±1.1</td>
<td>6.7±1.2</td>
<td>869±121</td>
</tr>
<tr>
<td>Saline alone</td>
<td>10</td>
<td>120±2</td>
<td>194±19</td>
<td>119±8</td>
<td>129±8</td>
<td>3.4±1.0</td>
<td>5.8±0.9</td>
<td>430±38</td>
</tr>
</tbody>
</table>
treated with oestrogen alone. In the group treated with salt loading alone, no significant increase in systolic pressure was observed.

Plasma renin and renin substrate values in all the experiments are shown in Table 1. In both experiments, the group treated with oestrogen alone showed significant increases in plasma renin activity and plasma renin substrate compared with those in the control group. On the other hand, plasma renin concentration was unchanged despite the oestrogen treatment.

In the oestrogen plus salt-loading group, plasma renin substrate increased significantly, as with the group treated with oestrogen alone. The increase in plasma renin activity, however, was not significant ($P > 0.05$), probably because of the significant suppression of plasma renin concentration.

In all the oestrogen-treated rats, with or without salt loading, there was a significant positive correlation between the increase of blood pressure during the treatment with oestrogens and plasma renin activities determined at the end of the experiment ($r = 0.35, P < 0.05$).

Discussion

It is known that oral contraceptives containing oestrogens induce hypertension in women but it is unknown whether hypertension is induced in other species by oestrogens or oestrogen–progesterone combinations. Nasjletti, Matsunaga, Tateishi & Masson (1971) have reported that injection of diethylstilboestrol (0.2 pmol/day) was not associated with an increase in blood pressure in female Sprague–Dawley rats. In this study we have induced hypertension in male Wistar rats by using two kinds of oestrogen, namely oestradiol (5 pmol/kg) intramuscularly and stilboestrol disulphate (56 pmol/kg) intraperitoneally.

With these doses, average increases of systolic pressure of 17 mmHg in one experiment and 34 mmHg in the other experiment were observed. Assay of renin components revealed that plasma renin substrate was two to three times higher than in the control group and plasma renin activity was also significantly increased on the fifteenth day of these treatments. Plasma renin concentration was, however, unchanged. These findings were similar to those seen in women taking oral contraceptives. Menard & Catt (1972) reported that plasma renin activity fell to slightly below the normal value by the sixth day of oestrogen treatment in rats. As we employed several times the amount of oestrogen used by these workers in order to induce hypertension, this difference in the amount of oestrogen may partially explain the discrepancy between their results and ours.

Alterations of the renin–angiotensin system induced by oestrogen may be important in causing the hypertension induced by oral contraceptives. In this experiment oestrogen clearly increased systolic pressure. Furthermore, the elevation of systolic pressure may be related to an increased amount of angiotensin, measured as plasma renin activity, because the elevation of systolic pressure was reduced by suppression of plasma renin activity by salt loading. Also, there was a significant relationship between the changes in systolic pressure and plasma renin activities, determined at the end of the experiment, in all the oestrogen-treated rats. However, we cannot exclude the possibility that factors other than the renin–angiotensin system may also play a role in the elevation of systolic pressure induced by oestrogen.

Acknowledgments

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References


SKINNER, S.L. (1967) Improved assay methods for renin


