Effect of pharmacological inhibition of renin, prostaglandin and kallikrein systems on surgical correction of longstanding two-kidney, one-clip hypertension in the rat

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Summary

1. Longstanding two-kidney, one-clip hypertension in the rat was rapidly reversed by removal of the constricting clip and the reversal was complete by 12 h.

2. Blood pressure was lowered by renin–angiotensin inhibition, caused by saralasin or captopril, but not to normotensive levels. Infusion with indomethacin or aprotinin over a 15 h period did not affect the blood pressure.

3. The pattern of fall in blood pressure after removal of the clip was similar when rats were infused with saralasin, captopril, indomethacin or aprotinin. Blood pressure was normal at 24 h and did not differ significantly from that of unclipped rats given an infusion of glucose.

4. The fall in blood pressure produced by unclipping was not altered by inhibition of the renin–angiotensin system and did not appear to be mediated via the prostaglandin or kallikrein systems.

Key words: Goldblatt hypertension, kallikrein, prostaglandin, renin.

Introduction

The rapid reversal of established two-kidney, one-clip hypertension in the rat [1] remains unexplained. It has been shown that the fall in blood pressure occurs as a result of a decrease in total peripheral resistance [2] and at a time when structural vascular changes have been demonstrated to be still present [3]. This fall in peripheral resistance to normal may be the result of removal of a pressor agent or the activation of a vasodepressor mechanism.

Plasma renin concentrations fall towards normal in longstanding hypertension in this model [4–7] and inhibition of the renin–angiotensin system results in only partial correction of hypertension at this stage [7–9]. Other intrarenal pressor agents have been advocated but remain unproven [10–12].

Alternatively there may be activation of a vasodepressor mechanism by relief of ischaemia. Two such candidates within the kidney are the prostaglandin and kallikrein systems.

To test the hypothesis that one of the above systems may be involved in the lowering of blood pressure seen after unclipping, we have removed the constricting clip from rats with two-kidney, one-clip hypertension of greater than 16 weeks’ duration whilst they were infused with saralasin (a competitive antagonist of angiotensin II), captopril (angiotensin I-converting enzyme inhibitor), indomethacin (a prostaglandin synthase inhibitor) or aprotinin (a potent kallikrein inhibitor).

Methods

Female Wistar rats (160–200 g) were used throughout and all surgical procedures were performed under ether anaesthesia. Two-kidney, one-clip hypertension was produced by placing a silver clip (0.2 mm internal diameter) on the left renal artery through a loin incision: the right kidney was not disturbed. Indirect blood pressures were measured by a light–plethysmographic method [13] and rats with blood pressures in excess of 150 mmHg more than 16 weeks after clipping were used for the study.

Polythene catheters were placed in the carotid artery (P50) and jugular vein (P30) and then
exteriorized between the scapulae and protected by a light, flexible, counterbalanced metal tube. On recovery from the anaesthetic the rats were placed in a plastic container (30 cm x 30 cm) with free access to food and water. Blood pressure was monitored continuously with a Statham P233 transducer connected to a Grass polygraph recorder.

Rats were randomly allocated to one of the following groups, each containing eight rats: sham operation and glucose infusion, or unclipping during glucose, saralasin, captopril, indomethacin or aprotinin infusion. When blood pressure had stabilized after cannulation, infusions of glucose (50 g/l), saralasin (10 μg min⁻¹ kg⁻¹), captopril (2.5 mg/kg initially followed by 8.3 μg min⁻¹ kg⁻¹), indomethacin (1 mg/kg initially followed by 1.3 μg min⁻¹ kg⁻¹) or aprotinin (50 000 units/kg initially followed by 100 units min⁻¹ kg⁻¹) were given through the venous line (P30) at an infusion rate of 0.25 ml/h.

The rats were infused for 15 h (overnight) and then subjected to a further operation, which consisted of either unclipping or a sham procedure, through the original incision. Operation was invariably completed within 10 min and infusions and direct blood pressure recordings were continued throughout this procedure and for the subsequent 24 h.

Plasma renin concentration (PRC) was measured by radioimmunoassay on samples of tail vein blood obtained before the study began [1]. The mean direct arterial blood pressure was calculated from the diastolic plus one-third of the pulse pressure. All results were expressed as mean values ± SEM and paired Student's t-test and one-way analysis of variance were used for statistical comparison. Values for PRC were transformed into logarithms before such comparisons were made, since PRC is not normally distributed.

Results

The initial plasma renin concentration (PRC) was 81 ± 13 pmol of ANG I h⁻¹ ml⁻¹ for hypertensive rats. This was not significantly different from that in age-matched normal rats (65 ± 16 pmol of ANG I h⁻¹ ml⁻¹, n = 8, P > 0.1). There was no significant difference between the groups studied.

The initial direct mean blood pressure was similar in all groups (Table 1, P > 0.05). There was no significant change in blood pressure during the preoperative infusion period in rats infused with glucose, indomethacin or aprotinin. Infusion of saralasin lowered blood pressure, but not significantly (P > 0.05), whereas the fall seen during the infusion of captopril was significant (P < 0.01). After operation the blood pressure of sham-operated rats rapidly returned to preoperative levels. In all rats that were unclipped the pattern of response was similar. By 6 h the blood pressure of unclipped rats was significantly lower than that of sham-operated rats (Table 1; P < 0.01) but not significantly different between the unclipped groups (P > 0.05). This was also the case at both 12 h and 24 h after operation (Table 1). All rats that were unclipped had significant falls in blood pressure at 6, 12 and 24 h compared with respective preoperative values (P < 0.01).

At 24 h post-unclipping mean blood pressure in all groups of unclipped rats was not significantly different from that in age-matched normal rats (123 ± 3.4 mmHg; n = 8, P > 0.1).

Discussion

The fall in blood pressure in this study after a 15 h infusion of saralasin was small and confirms

<table>
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<tr>
<th>Group (n = 8)</th>
<th>Direct mean blood pressure (mmHg)</th>
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<tr>
<td></td>
<td>Pre-infusion</td>
</tr>
<tr>
<td>Sham operation</td>
<td>171 ± 5.6</td>
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<tr>
<td>Unclip. 5% glucose</td>
<td>173 ± 5.8</td>
</tr>
<tr>
<td>Unclip. saralasin</td>
<td>174 ± 4.8</td>
</tr>
<tr>
<td>Unclip. captopril</td>
<td>186 ± 5.7</td>
</tr>
<tr>
<td>Unclip. indomethacin</td>
<td>178 ± 7.5</td>
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<tr>
<td>Unclip. aprotinin</td>
<td>192 ± 7.0</td>
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</tbody>
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One-way analysis of variance

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<th>P &gt; 0.05</th>
<th>P &lt; 0.01</th>
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The fall in blood pressure after unclipping has been reported to be mediated via a fall in total peripheral resistance [2] and this in the presence of vascular hypertrophy implies that smooth muscle tone is subnormal. The alternative explanation to that of removal of a vasopressor is that a vasodepressor substance is released. Three intrarenal vasodepressor systems have been proposed: prostaglandins, kallikrein and the renomedullary lipids [17].

Indomethacin is a potent prostaglandin synthase inhibitor [18] and previous work in rats with two-kidney, one-clip hypertension has demonstrated that those treated with indomethacin over several days developed higher pressures than did controls, and this was associated with a significant suppression of prostaglandin E synthesis [19]. In the present experiments, with the same dose of indomethacin there was a small but insignificant rise in direct conscious blood pressure in hypertensive rats (P > 0.1). There was no alteration in the blood pressure response to unclipping, with a similar pattern of response seen to that of glucose-infused unclipped rats: blood pressure was normal by 24 h.

The kallikrein system has potent vasodilator properties, which are mediated by the generation of kinins. Aprotinin inhibits conversion of inactive prekallikrein into kallikrein and thereby suppresses kinin generation [20]. The regimen used in the present study was similar to that used by Kramer et al. in conscious rats [21], based on previous work [22, 23]. This regimen was effective in reducing urinary immunoreactive PGE₂ by 60% [21]. It could be expected from previous studies that kinin generation would be significantly suppressed by the dose of aprotinin used in this study, although complete inhibition cannot be assumed. No effect on blood pressure was seen when hypertensive rats were infused for 15 h and the response of blood pressure after unclipping was similar to that seen in glucose-infused rats. A previous study has demonstrated that urinary kallikrein levels fall in the 8 h after unclipping in this model of hypertension [24]. It is possible that kallikrein acts as a local hormone, its effect possibly being mediated via prostaglandins [25] but the present experiments suggest that neither is necessary for the surgical correction of hypertension in this model.

We have previously demonstrated that the presence of an intact renal medulla is essential for the full reversal of hypertension by surgical correction in this model [26]. The present studies suggest that the renomedullary factor is not influenced by indomethacin or aprotinin and therefore operates independently of the prostaglandin and kallikrein systems.

Two-kidney, one-clip hypertension in the rat is fully reversed within 24 h of removal of the constricting clip even when hypertension is of more than 16 weeks’ duration. This reversal is not significantly influenced by continuous infusion of saralasin, captopril, indomethacin or aprotinin.

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References


