Does Renin Determine the Blood Pressure Response to Calcium Entry Blockers?

BERNARD WAEBER, JÜRGEN NUSSBERGER, AND HANS R. BRUNNER

SUMMARY Male Wistar rats with one-kidney, one clip renal hypertension were maintained on either a regular or a low salt diet for 3 weeks after clipping. At that time mean blood pressure in the unanesthetized rats was equally elevated in sodium-depleted ($n = 17$) and in sodium-replete rats ($n = 19$), but plasma renin activity was significantly higher in the former ($p < 0.05$). Infusion of the calcium entry blocker verapamil at a rate of 0.05 mg/kg/minute decreased blood pressure within 60 minutes to a similar extent in rats kept on a salt-deficient diet and in rats fed a regular salt diet. In all rats taken as a group, there was a close, direct correlation ($r = 0.87$, $p < 0.001$) between the magnitude of the blood pressure response to verapamil and the pretreatment blood pressure levels. Verapamil markedly accelerated heart rate and stimulated renin release in all rats. In additional groups of sodium-depleted ($n = 8$) and sodium-replete renal hypertensive rats ($n = 7$), nifedipine administration (4 /µg/kg/min i.v.) within a 45-minute observation period caused a blood pressure fall ($p < 0.001$) and heart rate acceleration ($p < 0.001$) that were comparable in both groups. These findings suggest that in the rat with renal hypertension the short-term blood pressure response to the calcium antagonists verapamil and nifedipine is not influenced by the state of sodium balance and plasma renin activity. In this experimental model of hypertension, the magnitude of the blood pressure lowering effect of calcium entry blockers appears to be proportional to pretreatment blood pressure levels. (Hypertension 7: 223-227, 1985)

KEY WORDS · renal hypertension · verapamil · nifedipine · conscious rats · high and low renin levels

DURING recent years numerous studies have focused on the crucial role of intracellular free calcium in determining vascular smooth muscle tone. 1, 2 Calcium channel blocking agents have been used to reduce intracellular calcium in an attempt to lower the blood pressure of hypertensive patients. Such drugs have proved to be helpful in managing hypertensive patients and appear particularly effective in reducing blood pressure when the rate of renin secretion is low. 3, 4 The present investigation was undertaken in conscious hypertensive rats to assess the effect of a change in sodium balance and renin release on the short-term blood pressure response to the calcium entry blockers verapamil and nifedipine. 5 The experiments were carried out in rats with hypertension induced by partially occluding one renal artery with the contralateral kidney removed (one-kidney, one clip renal hypertension) maintained postoperatively for 3 weeks on either a salt-deficient or a regular salt intake. As was known from previous work, 6, 7 blood pressure during the phase of established hypertension achieves similar high levels in sodium-depleted and sodium-replete rats in spite of the fact that plasma renin activity is elevated only in the former.

Materials and Methods
Male Wistar rats weighing 140 to 180 g (Madörin AG, Füllinsdorf, Switzerland) were used for this study. They were housed in a room with a constant temperature of 23°C and a humidity of about 50%. A solid silver clip (0.2 mm inside diameter) was placed on the left renal artery, and a right nephrectomy was performed with the rats under ether anesthesia. The animals were then returned to their cages and given a regular rat chow diet (Indulab, Buchs, Switzerland) containing 0.113 mmol/g of sodium. Two days later, this diet was replaced in half of the rats by a salt-deficient diet (Indulab, Buchs, Switzerland) contain-
TABLE 1. Basal Characteristics and Plasma Renin Activity of the Study Groups

<table>
<thead>
<tr>
<th></th>
<th>Sodium-replete rats</th>
<th>Sodium-depleted rats</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Vehicle (n = 9)</td>
<td>Verapamil (n = 10)</td>
</tr>
<tr>
<td>Body weight (g)</td>
<td>233 ± 12</td>
<td>241 ± 5.2</td>
</tr>
<tr>
<td>Mean blood pressure (mm Hg)</td>
<td>195 ± 5</td>
<td>196 ± 5</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>407 ± 15</td>
<td>425 ± 14</td>
</tr>
<tr>
<td>Plasma renin activity (ng/ml/hr)</td>
<td>4 ± 1</td>
<td>56 ± 13†</td>
</tr>
</tbody>
</table>

*p < 0.05, †p < 0.001, sodium-depleted versus sodium-replete rats.
§p < 0.01, †p < 0.001, verapamil versus vehicle.
All values are means ± SEM.

During the 3 weeks after renal artery clipping, the weight gain in sodium-replete (n = 19) and sodium-depleted (n = 17) rats subsequently given verapamil or its vehicle averaged 59.5 ± 5.6 and 47.7 ± 3.5 g respectively (p > 0.05, sodium-replete versus sodium-depleted rats). The basal characteristics of the study groups before administration of verapamil or its vehicle are outlined in Table 1. On the day of the experiment, no significant difference in body weight, base-

FIGURE 1. Time course and magnitude of the blood pressure and heart rate changes induced by verapamil administration in sodium-replete and sodium-depleted conscious rats with one-kidney, one clip renal hypertension.
line mean blood pressure, or pulse rate was observed between the four different groups of rats.

Figure 1 illustrates the time course of the blood pressure and heart rate changes induced by verapamil in sodium-replete and sodium-depleted hypertensive rats. Verapamil administration caused a rapid and pronounced blood pressure fall \((p < 0.001)\) and markedly accelerated heart rate \((p < 0.001)\) in both groups of rats. The changes in these parameters were almost identical throughout the experiment in rats kept on either diet. At the end of the observation period, blood pressure was \(133 \pm 4\) mm Hg in sodium-replete rats and \(126 \pm 5\) mm Hg in sodium-depleted rats. The corresponding heart rate levels averaged \(482 \pm 12\) and \(481 \pm 12\) beats/minute respectively. In contrast, administration of vehicle had no effect on blood pressure or heart rate (Figure 2) in both groups of rats on either sodium diet.

Figure 3 depicts the relationship between baseline blood pressure and the blood pressure response to the 60-minute infusion of verapamil in all rats treated with this agent. A close correlation \((r = 0.87, n = 18, p < 0.001)\) appeared between these two parameters.

Plasma renin activity was measured in all rats at the end of the experiment (Table 1). As expected, renin secretion was markedly higher in sodium-depleted than in sodium-replete rats, in both the vehicle- and verapamil-treated rats. Verapamil infusion markedly increased plasma renin activity in sodium-replete as well as in sodium-depleted rats.

Table 2 summarizes the results obtained with nifedipine in sodium-replete \((n = 7)\) and sodium-depleted \((n = 8)\) renal hypertensive rats. On the day of the experiment there was no significant difference in body weight between the former \((233 \pm 9\) g) and the latter \((214 \pm 7\) g). The blood pressure decrease and the heart rate acceleration induced by this compound were not dependent on the salt intake. Within 45 minutes, blood pressure fell by \(43 \pm 9.9\) mm Hg in sodium-replete rats and by \(45 \pm 6\) mm Hg in sodium-depleted rats. In all animals taken as a group there was a significant correlation between blood pressure before nifedipine

### Table 2. Blood Pressure and Heart Rate Effect of Nifedipine Administration

<table>
<thead>
<tr>
<th></th>
<th>Before nifedipine</th>
<th>After nifedipine (min)</th>
<th>15</th>
<th>30</th>
<th>45</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean blood pressure (mm Hg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sodium-replete rats ((n = 7))</td>
<td>(201 \pm 8.6)</td>
<td>(175 \pm 9^{*})</td>
<td>(160 \pm 8.4^{†})</td>
<td>(153 \pm 8.8^{†})</td>
<td></td>
</tr>
<tr>
<td>Sodium-depleted rats ((n = 8))</td>
<td>(213 \pm 6.3)</td>
<td>(189 \pm 4.6^{*})</td>
<td>(178 \pm 46^{†})</td>
<td>(168 \pm 4.3^{†})</td>
<td></td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sodium-replete rats</td>
<td>(381 \pm 18)</td>
<td>(448 \pm 17^{*})</td>
<td>(464 \pm 14^{†})</td>
<td>(465 \pm 14^{†})</td>
<td></td>
</tr>
<tr>
<td>Sodium-depleted rats</td>
<td>(394 \pm 29)</td>
<td>(461 \pm 21^{*})</td>
<td>(494 \pm 15^{†})</td>
<td>(491 \pm 16^{†})</td>
<td></td>
</tr>
</tbody>
</table>

\(^{*}p < 0.05, {^†}p < 0.001, \) after versus before nifedipine administration.

All values are means \(\pm\) SEM.
administration and the magnitude of the drug-induced reduction observed at the end of the experiment \( (r = 0.60, p < 0.05) \).

**Discussion**

Free intracellular calcium has been demonstrated to play a key role in regulating the contractile state of the vascular smooth muscle cell.\(^1\)\(^,\)\(^2\) Evidence emerging during the last few years has suggested that the development of both genetic hypertension in the rat\(^6\)\(^-\)\(^11\) and essential hypertension in humans\(^12\)\(^,\)\(^13\) is linked to an abnormality in cellular calcium metabolism. It now appears possible that the cellular defect of calcium handling is caused primarily by a derangement in transmembranous sodium transport.\(^1\) In support of such a mechanism is the recent observation that calcium channel blockers, which inhibit the flux of extracellular calcium ions across the cell membrane, have the greatest antihypertensive effect in patients with low plasma renin levels\(^14\) — that is, in patients who presumably have an excess of total body sodium\(^15\) as well as an intracellular sodium accumulation.\(^16\)

In the present study the short-term blood pressure and heart rate effects of verapamil were investigated in rats with hypertension induced by clamping one renal artery and removing the contralateral kidney. In these rats the factors involved in the maintenance of high blood pressure typically depend on the state of sodium balance. In rats kept on a regular salt intake, blood pressure is renin dependent only during the developmental phase of hypertension.\(^6\) A few weeks following renal artery clipping, possibly as a result of a progressive salt retention,\(^11\) plasma renin levels are no longer elevated and blood pressure becomes unresponsive to the blockade of the renin-angiotensin system.\(^6\) In our rats the magnitude of the short-term blood pressure response to calcium entry blockade with verapamil or nifedipine was not influenced by the state of sodium balance nor on the degree of activation of the renin-angiotensin II system.\(^5\)\(^,\)\(^6\) Unlike nifedipine, verapamil, has been shown to interact in vitro with \(\alpha\)-adrenergic receptors.\(^20\) Such an action unrelated to calcium channel blockade could potentially interfere with the interpretation of our results. The fact that we obtained comparable results with both inhibitors suggests that \(\alpha\)-blockade was not the main mechanism responsible for the blood pressure lowering effect of verapamil.

In the present study two different calcium channel blockers, verapamil and nifedipine, were administered to rats with the same hypertension model because verapamil, unlike nifedipine, has been shown to interact in vitro with \(\alpha\)-adrenergic receptors.\(^20\) Such an action unrelated to calcium channel blockade could potentially interfere with the interpretation of our results. The fact that we obtained comparable results with both inhibitors suggests that \(\alpha\)-blockade was not the main mechanism responsible for the blood pressure lowering effect of verapamil.

On the other hand, in several aspects the present experimental findings confirm those previously observed in hypertensive patients during short-term blockade of calcium channels. First, as in the patients,\(^4\)\(^,\)\(^18\) heart rate was accelerated during verapamil and nifedipine infusion in both sodium-depleted and sodium-replete rats, most probably as a consequence of a baroreceptor reflex-mediated increase in sympathetic nerve activity. Second, as in hypertensive patients renin secretion was markedly stimulated by verapamil administration in rats. The reduction in arterial pressure per se as well as the compensatory stimulation of sympathetic nerve activity may well be responsible for the effect of verapamil on renin release.\(^21\) Finally, in all rats taken as a group, the magnitude of the blood pressure decrease induced by both verapamil and nifedipine administration was directly proportional to the pretreatment blood pressure level. Again, this finding is in total agreement with clinical experience.\(^4\)\(^,\)\(^14\)\(^,\)\(^18\)

**Conclusion**

Our data indicate that the acute vasodilating effect of calcium entry blockade in the conscious rat with renal hypertension does not depend on the state of sodium balance nor on the degree of activation of the renin-angiotensin system, but that it is directly related to pretreatment blood pressure.

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