Exaggerated Renal Vasodilator Response to Calcium Entry Blockade in First-Degree Relatives of Essential Hypertensive Subjects

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SUMMARY Because an inherited renal factor may contribute to essential hypertension in humans, the study of family members is attractive. To assess the determinants of renal vascular tone, graded doses of either diltiazem (10–1000 μg/min) or acetylcholine (1–100 μg/min) were infused into the renal artery in 52 normotensive subjects, 16 with and 36 without a family history of hypertension when they were in balance on either a 10-mEq or 200-mEq sodium intake. Renal blood flow was measured with 133Xe. Restricted sodium intake potentiated renal vascular responses to diltiazem (p<0.01), suggesting a role for angiotensin as a determinant. In four subjects with no family history of hypertension on a 200-mEq sodium intake, angiotensin II in subpressor doses (1 ng/kg/min i.v.) induced renal vasoconstriction and enhanced the renal vasodilator action of diltiazem (p<0.001). In subjects with a family history of hypertension, the renal vascular response to diltiazem was enhanced (p<0.01) despite similar values of plasma renin activity, angiotensin II concentration, and sodium excretion. Because responses to acetylcholine were modified neither by sodium intake nor by family history, specificity for diltiazem was suggested. The intriguing possibility is raised that the enhanced renal vascular response to diltiazem reflects an abnormality in the control of renal vascular tone in the offspring of essential hypertensive subjects. (Hypertension 9: 384–389, 1987)

KEY WORDS • sodium chloride • angiotensin II • acetylcholine • blood flow

Evidence indicating that a functional abnormality, active renal vasoconstriction, contributes to the abnormalities in renal perfusion in patients with mild to moderate essential hypertension stems from the observation of a potentiated renal vasodilator response to nonspecific agents such as acetylcholine. Recent observations that calcium entry blocking agents produce an exaggerated vasodilator response in the limb circulation of some patients with essential hypertension and an enhanced depressor response led us to examine the possibility that such a response would also occur in the renal circulation, a vascular bed with special pathogenetic implications.

Early in the course of this study an unanticipated observation was made: The offspring of patients with hypertension showed an exaggerated renal vasodilator response when compared with subjects from families in which hypertension was absent — a finding that became the central thrust of this study. An unanticipated influence of sodium intake on the renal vascular response to diltiazem led us, in addition, to examine the influence of diltiazem on the renal vascular response to angiotensin II.

Subjects and Methods

The 52 normal subjects ranged in age from 18 to 65 years. They were admitted to the clinical research center of the Brigham and Women's Hospital for evaluation as potential kidney donors. All subjects were placed on a constant, isocaloric diet throughout their hospitalization, including either 10 mEq of sodium per day (25 subjects) or 200 mEq per day (27 subjects). Daily dietary potassium intake (100 mEq) and a fluid intake of 2500 ml/day were also constant. Twenty-four hour urine collections were made each day and analyzed for sodium, potassium, and creatinine content.
A thorough history, physical examination, and laboratory evaluation were employed to rule out major medical illness, as described in detail. The assessment of family history of hypertension was made in several steps. First, health history questionnaires were sent to the patient's home before admission, and the subjects were asked to consult family members about details of family history. On admission a careful review of the history of family members was made. Confirmation was made by communication with the physician responsible for the care of family members when direct contact was impossible. A positive family history of hypertension in the study indicates the existence of one parent whose physician had diagnosed essential hypertension that required medical therapy before the age of 60 years. In one instance both parents were hypertensive. By these criteria, 16 of the 52 subjects (29%) were considered to have a positive family history of hypertension.

Patients were studied in the cardiovascular laboratories of the Department of Radiology at the time of selective arteriography, done as part of the evaluation for living related kidney donation. All subjects gave written informed consent.

For the blood flow studies a 3F Teflon catheter was introduced coaxially through a 7F untapered renal artery catheter to serve as the conduit for drug infusion, while leaving the outer catheter for continuous measurement of arterial blood pressure and for administration of radioxenon to permit assessment of renal blood flow. Protocols were designed to be completed within 20 to 25 minutes so as not to extend the duration of arterial catheterization.

The method for measuring renal blood flow with radioxenon has been described in detail. In brief, four measurements were made over a period of 20 minutes by the injection of serially ascending xenon doses so that the peak count exceeded background by at least 1000. After baseline determination of renal blood flow, one of the two agents employed, diltiazem or acetylcholine, was infused into the renal artery in graded dosage. Because preliminary experiments indicated that a new steady state blood flow increase had been achieved with each agent within 3 minutes of initiating an infusion, blood flow measurements were made between 3 and 4 minutes after each infusion rate was initiated for each agent. Diltiazem infusions provide a dosage ranging from 30 to 1000 µg/min. Acetylcholine doses ranged from 1 to 100 µg/min. No subject received more than three doses, in ascending log-dose increments. Attempts to increase the diltiazem dose to 3000 µg/min led to a fall in blood pressure, so that the study was terminated.

Because diltiazem induced a substantially larger renal blood flow increase in subjects studied on a restricted sodium intake, a second protocol was adopted to assess the possibility that angiotensin II–mediated vasoconstriction, induced by restriction of sodium intake, accounted for the potentiated response. In four additional subjects with no family history of hypertension, external sodium balance was achieved on a 200-mEq sodium intake, and after a baseline renal blood flow measurement, angiotensin II was infused intravenously in a subpressor dose (1 ng/kg/min) to induce stable renal vasoconstriction approximately equivalent to that induced by restriction of sodium intake, as described earlier. A second blood flow determination 5 minutes after initiating angiotensin II infusion documented the new baseline, and with continued intravenous angiotensin II infusion diltiazem was infused at two dose levels directly into the renal artery. For the purposes of this analysis the response to diltiazem was assessed from the new baseline achieved during angiotensin II infusion. The two diltiazem doses employed were 100 and 300 µg/min.

Arterial blood pressure, heart rate, and the electrocardiogram were monitored continuously to ensure patient safety and cardiovascular stability during the study. Arterial and renal vein blood samples were drawn before the infusion and after 20 minutes for measurement of plasma renin activity, plasma angiotensin II concentration, and plasma epinephrine, norepinephrine, and dopamine concentrations. All blood samples were immediately placed on ice, and the plasma was separated and frozen until assay. The methods for measurement of plasma renin activity, angiotensin II, and catecholamines were as previously described. The transit of radioxenon as an index to renal blood flow was also assessed by methods described elsewhere.

Group means have been presented with the standard error of the means as the index of dispersion. Statistical probability was assessed by several approaches. For continuous data the t test was employed, unless multiple comparisons mandated analysis of variance. Dose-response relationships were assessed with the Fisher exact test for central tendency around the median at each dose. We rejected the null hypothesis when a 5% probability or less was found.

The protocols and consent form were approved by the Human Subjects Committee of the Brigham and Women's Hospital, and written informed consent was obtained from each subject.

**Results**

**Baseline Values**

The baseline characteristics of the normotensive subjects with and without a family history of hypertension, including baseline values for blood pressure, renal blood flow, daily sodium excretion at the time of the study, and age, did not differ, apart from anticipated differences in sodium excretion based on sodium intake (Table 1). The data for blood pressure presented in the table were obtained during the hemodynamic study. A similar analysis of blood pressure at the time of admission or during hospitalization also failed to show a difference between the two groups, although the conditions were not standardized, making a comparison difficult.

Baseline endocrine indices for the subgroups studied when stimulated by a 10-mEq sodium intake, clas-
TABLE 1. Baseline Characteristics of Normotensive Subjects Classified by Family History of Hypertension and Sodium Intake

<table>
<thead>
<tr>
<th>Variable</th>
<th>Family history of hypertension</th>
<th>No family history of hypertension</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10 mEq Na+</td>
<td>200 mEq Na+</td>
</tr>
<tr>
<td>Blood pressure (mm Hg)</td>
<td>122 ± 3/68 ± 3</td>
<td>112 ± 8/63 ± 4</td>
</tr>
<tr>
<td></td>
<td>320 ± 30</td>
<td>339 ± 29</td>
</tr>
<tr>
<td>Renal blood flow (ml/min/100 g)</td>
<td>11 ± 2</td>
<td>196 ± 38</td>
</tr>
<tr>
<td>Urinary Na+ excretion (mEq/24 hr)</td>
<td>35 ± 4</td>
<td>42 ± 3</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>35 ± 4</td>
<td>35 ± 5</td>
</tr>
</tbody>
</table>

Values are means ± SEM.

The increase in renal blood flow induced by diltiazem at any dose was potentiated in the subjects with a family history of hypertension (p < 0.001). The data for studies performed in nine subjects with no family history and eight subjects with a family history of hypertension on a restricted sodium intake are presented in Figure 2, while data for all studies normalized for diet and dose of the agent are shown in Figure 3. Two subjects with a family history of hypertension on a high salt diet received diltiazem, and six received acetylcholine — three, respectively, on each sodium intake.

Response to Acetylcholine

Sodium intake did not influence the renal vascular response to acetylcholine in subjects with no family history of hypertension (see Figure 1), nor did a family history of hypertension influence the response (see Figure 3).

Diltiazem–Angiotensin II Interaction

Intravenous infusion of angiotensin II in a subpressor dose of 1.0 ng/kg/min induced the anticipated fall in renal blood flow (from 364 ± 30 to 256 ± 33 ml/100 g/min; p < 0.001) but had no influence on arterial blood pressure, which was unchanged in each of the four subjects studied. The subsequent superimposition of diltiazem at doses of 100 and 300 μg/min (Figure 4) revealed clear evidence of potentiation (p < 0.001) of the renal vasodilator response to diltiazem.

Discussion

Two observations in this study were initially unanticipated; first, that sodium intake would influence the renal vascular response to diltiazem; second, that family history of hypertension would influence that response. Although neither was part of the original working premise, their possible influence was recognized early; a substantial portion of the study was performed prospectively; and a large number of subjects was studied. There seems little doubt of the influence of either factor.

The renal vascular response to a shift in sodium intake is dominated largely by changes in the local concentration of angiotensin II. Thus, for example, in normal humans the renal blood flow increase induced by the angiotensin II antagonist saralasin14 and by con-
verting enzyme inhibition are substantially larger in subjects ingesting a low sodium intake. The similarity of the influence of sodium intake on the latter agents and on diltiazem, as well as the similarity in the magnitude of the response, raised the intriguing possibility that a similar mechanism was involved. Our study with acetylcholine made it clear that restriction of sodium intake does not potentiate the renal vascular response to all vasodilators nonspecifically. The smooth muscle action of angiotensin II is dependent on extracellular calcium ion flux, which is blocked by calcium channel blockers in other systems. The reversal of angiotensin II-induced renal vasoconstriction in this study is also consistent with such an explanation.

Calcium entry blockers are especially effective as antihypertensive agents in low renin hypertension. This phenomenon may reflect an unrelated mechanism, but too little is known about the control of the renal circulation in low renin hypertension to link the observation in this study to the clinical response to calcium channel blockade.

The renal vascular response to diltiazem, but not to acetylcholine, was also potentiated in about half of the offspring of subjects with hypertension—presumably essential hypertension. Nothing in this study would allow us to ascertain unequivocally whether the two observations—the potentiation of diltiazem’s action on the renal blood supply in the presence of angiotensin II or sodium restriction and in association with a family history of hypertension—are related. The intriguing observation by Uneda et al., that the offspring of patients with essential hypertension showed a potentiated renal vascular response to converting enzyme inhibition with captopril, may be relevant. Both studies may reflect the reversal of a renal vascular action of angiotensin II, but such an explanation must be considered to be speculative. In the study in which a converting enzyme inhibitor was employed, a real possibility existed that the renal vasodilatation reflected the local accumulation of vasodilators, such as bradykinin or prostaglandins, but it is substantially less likely that the diltiazem used in the present study acted through a similar mechanism. The feature common to the two studies is reversal of angiotensin II-mediated vasoconstriction.

Basal renal blood flow was not different in subjects

Figure 1. Change in renal blood flow (RBF) in response to graded doses of diltiazem and acetylcholine. Note that restriction of sodium intake potentiated the response to diltiazem (p < 0.001), but not to acetylcholine.

Figure 2. The influence of family history (FH) of hypertension on the renal vascular response to diltiazem. All studies in this figure were performed with the subjects on a restricted sodium intake. The response was potentiated in the normotensive offspring of hypertensive parents (p < 0.01), although the threshold dose was unchanged. RBF = renal blood flow.

Figure 3. The influence of family history of hypertension on the renal vascular response to diltiazem and acetylcholine. Each point has been normalized to the median for the response by dose, by diet, and by whether the subject did (FH+) or did not (FH-) have a family history of hypertension. Note the approximately normal distribution of the points for both agents in the group with no family history of hypertension and for acetylcholine in the subjects with a family history of hypertension. Exactly 50% of the points in the group with a family history of hypertension receiving diltiazem showed a potentiated response. The 95% confidence intervals for each agent, defined in normal subjects with no family history of hypertension, is shown as a stippled line. RBF = renal blood flow.
Angiotensin II (All) was infused at a rate of 1 ng/kg/min, which was sufficient to reduce renal blood flow (RBF) by about 20% without a pressor response. Note the potentiated response to diltiazem when superimposed on angiotensin-induced renal vasoconstriction.

with or without a family history of hypertension, nor was calculated renal vascular resistance, as arterial blood pressure was essentially identical. If an appropriate level of angiotensin or some other vasoconstrictor stimulus was present in the kidney to account for the potentiated increase in renal blood flow in response to diltiazem in the subjects with a family history of hypertension, activation of an opposing vasodilator stimulus is the most plausible explanation for a similar basal renal blood flow. There is ample precedent in the renal blood supply, where vasodilator prostaglandin release often offsets the constrictor response to agents such as angiotensin II.\textsuperscript{19} It would be reasonable to speculate that the renal vasoconstriction that characterizes early hypertension in many patients reflects as much the loss of a compensating dilator response as an inappropriate constrictor state. Bianchi et al.\textsuperscript{3} described an increase in renal blood flow in some offspring of essential hypertensive subjects. Perhaps the offsetting mechanism was more pronounced in the subjects they studied.

We have described a group of subjects with essential hypertension, called nonmodulators.\textsuperscript{21} Several lines of evidence have suggested that they have an inappropriate concentration of angiotensin in their kidneys,\textsuperscript{22, 23} an abnormality that limits their ability to handle a sodium load.\textsuperscript{23, 24} Several observations may link that group to this study. First, a family history of hypertension is exceedingly common in nonmodulators, exceeding 80%.\textsuperscript{6, 24} Second, in the present study, plasma aldosterone concentration in subjects with a family history of hypertension studied on a low salt diet was strikingly less than that in the subjects with no family history of hypertension, despite an identical plasma angiotensin II concentration. This relative unresponsiveness of aldosterone release to angiotensin II in subjects on a low salt diet is one of the characteristics of nonmodulators.\textsuperscript{21}

Evidence for a genetic factor in the pathogenesis of essential hypertension, in at least some patients, appears to be overwhelming.\textsuperscript{7, 8, 25, 26} Precisely what is inherited remains in doubt, but it is the subject of much speculation. We found that about half of the subjects with a family history of hypertension showed a potentiated renal vascular response to diltiazem, and it seems unlikely that half would go on to become hypertensive. Ayman\textsuperscript{27} reported that hypertension developed in about 28% of the offspring of a single hypertensive parent. On the other hand, what is inherited may be a predisposition that will only express itself if the appropriate environmental stimulus is applied. In this case one might speculate that an abnormality in the control of the renal blood supply is inherited — evident in an enhanced vasodilator response to converting enzyme inhibitors\textsuperscript{15, 18, 20} and to calcium channel blockade — and is expressing itself as a limited ability to handle a salt load. Genetic factors in sodium handling have been clearly documented.\textsuperscript{25} From this baseline characteristic a predictable series of events may follow.

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References
