High Calcium Diet Prevents Baroreflex Impairment in Salt-Loaded Spontaneously Hypertensive Rats

Ayumu Ono, Tomoyuki Kuwaki, Wei-Hua Cao, Mamoru Kumada, Toshiro Fujita

Abstract  To investigate the role of the sympathetic control mechanism in the antihypertensive effect of dietary calcium supplementation, we examined whether a high calcium diet affected mean arterial pressure, renal sympathetic nerve activity, heart rate, and overall and central properties of the arterial baroreceptor reflex in salt-loaded young spontaneously hypertensive rats (SHR). Six-week-old SHR were fed either a normal (0.66%) or high (8.00%) salt diet with either a normal (1.17%) or high (4.07%) calcium content for 4 weeks. The arterial baroreceptor reflex was elicited with rats under halothane anesthesia by altering mean arterial pressure with nitroprusside or phenylephrine. The overall property of the arterial baroreceptor reflex was assessed by the median mean arterial pressure (MAP$_{50}$) and maximal gain (G$_{max}$) of the two relations and an attenuation of reflex inhibition of renal sympathetic nerve activity by aortic depressor nerve stimulation. There were no significant differences in mean arterial pressure, renal sympathetic nerve activity, or the overall and central properties of the arterial baroreceptor reflex among the control, high salt/high calcium, and normal salt/high calcium groups. In conclusion, dietary calcium supplementation prevented accelerated hypertension with sympathetic overactivity as well as impairment of the arterial baroreceptor reflex in salt-loaded young SHR. It is suggested that normalization of both tonic and reflex control of sympathetic discharge is involved in the antihypertensive effect of a high calcium diet on salt-induced hypertension. (Hypertension. 1994;24:83-90.)

Key Words • calcium, dietary • sodium • central nervous system • pressoreceptors • sympathetic nervous system • rats, inbred SHR

Although epidemiologic studies have shown an inverse relation between daily oral calcium intake and the level of arterial pressure,1-2 clinical3-4 and experimental5-7 studies do not ubiquitously support the hypothesis that dietary calcium has an antihypertensive effect. With respect to hypertensive rat models, however, dietary calcium supplementation has been shown to consistently prevent salt-induced hypertension in Dahl salt-sensitive rats,8 young spontaneously hypertensive rats (SHR),5-6,9 and angiotensin II-treated rats.7 Moreover, evidence suggests that alteration of sympathetic activity may be involved in the antihypertensive effect of dietary calcium in these models.4-8

The arterial baroreceptor reflex (ABR) is a major mechanism in the central control of arterial pressure, heart rate (HR), and sympathetic vasomotor activity.10 The ABR has been demonstrated to be impaired in certain salt-sensitive animal models11-14 and patients with hypertension.15 Moreover, a high calcium diet in salt-induced hypertensive Dahl salt-sensitive rats augmented the baroreflex response of renal sympathetic nerve activity (RSNA) to alterations of arterial pressure.8 These results suggest that impairment of the ABR contributes to the pathogenesis of salt-induced hypertension and that normalization of the impaired ABR underlies the antihypertensive effect of a high calcium diet.

The present study, conducted in young salt-sensitive SHR,5,6,16 attempted to examine the effects of salt loading and simultaneous calcium supplementation on the sympathetic control mechanism with special attention paid to the ABR. We compared, in addition to basal levels of mean arterial pressure (MAP), HR, and RSNA, the overall and central properties of the ABR among four groups of SHR who were fed either a normal or high salt diet with either a normal or high calcium content. In examining the overall and central properties of the ABR, we made use of the fact that the aortic depressor nerve in the rat consists exclusively of baroreceptor afferents17 and that RSNA reflects sympathetic vasomotor fiber activity.18 Thus, the overall property of the ABR was assessed by the relation between MAP and RSNA and between MAP and HR, and the central property of the ABR was assessed by reflex inhibition of RSNA and HR, which was elicited by electrical stimulation of the aortic depressor nerve.
Methods

Preparation of Animals

Male SHR, confirmed to be salt sensitive by us16-19 and others,20 were purchased from Charles River Japan at 5 weeks of age. The following procedures were all in accordance with the "Guiding Principles for the Care and Use of Animals in the Field of Physiological Sciences Recommended by the Physiological Society of Japan." The animals were maintained at constant humidity (60±5%), constant temperature (23±1°C), and regular light/dark cycle (light period from 6 AM to 6 PM). Beginning on the fifth day after arrival, rats were fed chow containing one of four combinations of NaCl and calcium carbonate for 4 weeks: 0.66% NaCl and 1.17% calcium (control group, n=13), 8.00% NaCl and 1.17% calcium (high salt/normal calcium group, n=14), 8.00% NaCl and 4.07% calcium (high salt/high calcium group, n=12), and 0.66% NaCl and 4.07% calcium (normal salt/high calcium group, n=8). Food and tap water were supplied ad libitum during the entire period of the study.

At the end of the dietary period, the rats were anesthetized initially with ether. After insertion of tracheal, arterial, and venous canulas, the rats were immobilized by d-tubocurarine chloride (initially, 0.1 mg per rat IV; thereafter, 0.1 mg/h IV) and artificially ventilated with halothane in oxygen-enriched room air. The halothane concentration was kept at 1.0% to 1.5% during surgery and was then reduced to 0.6% to 0.8% during the recording period. Adequacy of anesthesia was evaluated by the absence of increases in arterial pressure, HR, and RSNA to hindlimb toe pinch. End-tidal Pco2 was monitored continuously (Respina H26, Sanet-NEC) and maintained between 3% and 4.5%. Body temperature was maintained at 37±0.5°C by means of a ventral heat source.

Recording of Cardiovascular Variables and RSNA

Arterial pressure was recorded from the abdominal aorta by a polyethylene catheter (0.5-mm inner diameter) inserted through the femoral artery. MAP was recorded by passing the arterial pressure signal through a low-pass filter (corner frequency, 0.2 Hz). HR was computed from the arterial pressure pulse by a tachometer (AT-601G, Nihon Kohden).

The left renal nerve was approached retroperitoneally through a left flank incision and prepared for electrical stimulation with a constant current stimulator. Nerve activity was recorded from the renal nerve with one of the following two methods to evaluate RSNA. In protocol 1 (see below), renal nerve discharges were full-wave rectified and integrated over a 1-second interval. At the end of the experiment, instrumentation noise was recorded after the renal nerve was cut proximally to the recording site and was subtracted from all of the experimental values.

In protocol 2, renal nerve discharges were converted into a train of standard pulses with a window discriminator whose threshold was set slightly above the noise level. A peristimulus time histogram (PSTH) of the evoked RSNA was constructed, along with that of HR, by counting the standard pulses for 32 successive stimulations applied to the aortic depressor nerve. All recorded variables were stored in a tape recorder for further analyses (XR-7000L, TEAC).

Experimental Protocol and Data Analysis

Protocol 1: Comparison of MAP-RSNA and MAP-HR Relations

In 33 rats, MAP was varied within a range of approximately 50 to 230 mm Hg by intravenous bolus injections of different doses of sodium nitroprusside (1 to 30 µg) or phenylephrine (1 to 30 µg). Drugs were dissolved in normal saline to produce a volume of 100 µL for each injection. There was a control period of 5 to 10 minutes between each injection. Peak values of RSNA and HR after each injection were correlated with that of MAP, and the data obtained from a single rat were fitted to the logistic curve using a graphic-assisted fitting program (KALEIDA GRAPH, Synergy Software) to obtain MAP, RSNA and MAP-HR relations. RSNA was expressed as the percentage of the maximal level experimentally attained at or near a MAP of 50 mm Hg, and HR was expressed as beats per minute. We discarded data that had a correlation coefficient less than 0.9.

From the fitted curve the median MAP (MAP0) and maximal gain (Gm) were calculated. MAP0 represents the MAP at which RSNA or HR was the median of the maximal and minimal values and has been used to assess shifting of the logistic curve. Gm is the slope (expressed as a positive value) of the fitted curve at MAP0 and represents the maximal activity of the ABR. With respect to the MAP-HR relation, the HR range, ie, difference between maximal and minimal HR values, was also calculated. These calculated parameters were compared among the four groups to determine the effect of salt loading and calcium supplementation on the ABR.

We also determined the MAP-RSNA relation for each of the four SHR dietary groups by using the mean value of both MAP0 and Gm to reconstruct a single logistic curve representing each group. The same method was used to construct the MAP-HR relation except that the maximal and minimal HR values were also used.

Protocol 2: Comparison of PSTHs of RSNA and HR on Electrical Stimulation of the Aortic Depressor Nerve

In 47 rats, including those examined in protocol 1, the left aortic depressor nerve was identified at its junction with the superior laryngeal nerve and prepared for electrical stimulation. Stimulation was a 1-second train of negative square-wave pulses of 0.2-millisecond duration, with an intensity of 0.5 to 10 V and frequency of 25 or 100 Hz. The stimulation period was set at 1 second to minimize changes in arterial pressure that could secondarily affect RSNA and HR.

Although the aortic depressor nerve is composed of both myelinated and unmyelinated fibers, it has been demonstrated that the ABR is mediated by myelinated fibers at a stimulus frequency of 100 Hz.22,24,25 At 25 Hz, the ABR is mediated mostly by unmyelinated fibers.22,24,25 Stimulation was repeated 32 times each with an interval of 20 seconds or longer, and the PSTH of RSNA or HR was constructed at each stimulus intensity. The reflex response recorded as the PSTH was measured17-24,26 with the aid of a computer (NEC, PC-9801RX). The area above or below the prestimulus control level of each variable was calculated by integrating the PSTH between the onset and end of the evoked response. A negative sign was used to indicate that the response decreased below that of the control. The area calculated was normalized by dividing it by the prestimulus control level integrated over the 1-second period. Since aortic depressor nerve stimulation elicited reflex inhibition of RSNA and HR, the response was expressed as the percent decrease in these variables.

Statistical Analysis

Data are presented as mean±SEM. Comparison of the data among the four groups of SHR was performed by one-way ANOVA followed by Duncan's method for multiple comparisons among individual means. A value of P<.05 was considered statistically significant.

Results

Basal Hemodynamic Parameters and Body Weight

We compared basal levels of MAP, RSNA, HR, and body weight among the four groups of young SHR fed either a normal or high salt diet with either normal or high calcium intake for 4 weeks (Table 1). MAP and RSNA were significantly greater in the high salt/normal...
TABLE 1. Effect of Salt Loading and/or Calcium Supplementation on Body Weight and Basal Mean Arterial Pressure, Heart Rate, and Renal Sympathetic Nerve Activity in Young Spontaneously Hypertensive Rats

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>High Salt</th>
<th>High Salt+Ca</th>
<th>High Ca</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>13</td>
<td>14</td>
<td>12</td>
<td>8</td>
</tr>
<tr>
<td>BW, g</td>
<td>254±7</td>
<td>242±3</td>
<td>233±3</td>
<td>240±8</td>
</tr>
<tr>
<td>MAP, mm Hg</td>
<td>120±3</td>
<td>142±4*</td>
<td>119±3†</td>
<td>119±4</td>
</tr>
<tr>
<td>HR, bpm</td>
<td>335±11</td>
<td>340±19</td>
<td>322±10</td>
<td>356±9</td>
</tr>
<tr>
<td>RSNA, pulses/s</td>
<td>177±14</td>
<td>336±26*</td>
<td>165±19†</td>
<td>148±28</td>
</tr>
</tbody>
</table>

BW indicates body weight; MAP, mean arterial pressure; HR, heart rate; and RSNA, renal sympathetic nerve activity. Young spontaneously hypertensive rats (SHR) were divided into four groups: control, SHR fed a diet containing 0.66% NaCl and 1.17% calcium; high salt, high salt/normal calcium group fed a diet containing 8.00% NaCl and 1.17% calcium; high salt/high calcium group fed a diet containing 8.00% NaCl and 4.07% calcium; and high Ca, normal salt/high calcium group fed a diet containing 0.66% NaCl and 4.07% calcium. Values are mean±SEM.

*P<.01 compared with control.
†P<.01 compared with high salt.

calcium group compared with the control group. By contrast, in the high salt/high calcium group as well as in the normal salt/high calcium group, MAP and RSNA were not significantly different from those of the control group. Furthermore, the high salt/high calcium group had significantly lower MAP and RSNA compared with the high salt/normal calcium group. These results indicate that simultaneous calcium supplementation prevented salt-induced increases in MAP and RSNA. There was no significant difference in HR or body weight among the four groups.

Relation Between MAP and RSNA

The MAP-RSNA relation obtained from individual rats was fitted to a logistic function curve to determine $G_{\text{max}}$ and $MAP_0$ (see "Methods"). The two parameters were subsequently compared among the four groups (Table 2). In the high salt/normal calcium group, $G_{\text{max}}$ was significantly lower and $MAP_0$ was significantly higher than in the control group. Consequently, in the high salt/normal calcium group, the curve was blunted and shifted toward the higher pressure range (Fig 1). By contrast, in the high salt/high calcium group, as in the normal salt/high calcium group, $G_{\text{max}}$ and $MAP_0$ were not significantly different from those of the control group, and the MAP-RSNA relation was not apparently changed (Fig 1). Furthermore, the high salt/high calcium group had significantly greater $G_{\text{max}}$ and lower $MAP_0$ compared with the high salt/normal calcium group.

Relation Between MAP and HR

The MAP-HR relation obtained from the four groups was analyzed in the same manner as the MAP-RSNA relation (Table 2). In the high salt/normal calcium group, $G_{\text{max}}$ was significantly lower and $MAP_0$ was significantly higher than in the control group. Consequently, in the high salt/normal calcium group, the MAP-HR relation was blunted and shifted to the right (Fig 2). By contrast, in the high salt/high calcium group, as in the normal salt/high calcium group, $G_{\text{max}}$ and $MAP_0$ in the MAP-HR relation were not significantly different from those of the control group. Furthermore, the high salt/high calcium group had significantly

TABLE 2. Effect of Salt Loading and/or Calcium Supplementation on $MAP_0$ and $G_{\text{max}}$ of the Relation Between Mean Arterial Pressure and Renal Sympathetic Nerve Activity or Heart Rate in Spontaneously Hypertensive Rats

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>High Salt</th>
<th>High Salt+Ca</th>
<th>High Ca</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>7</td>
<td>10</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td>$MAP_0$, mm Hg</td>
<td>145±7</td>
<td>196±13*</td>
<td>163±7†</td>
<td>158±5</td>
</tr>
<tr>
<td>$G_{\text{max}}$, mm Hg$^{-1}$</td>
<td>1.02±0.06</td>
<td>0.60±0.06*</td>
<td>0.95±0.12‡</td>
<td>1.11±0.13</td>
</tr>
<tr>
<td>$MAP_0$, mm Hg</td>
<td>136±5</td>
<td>169±13*</td>
<td>126±7†</td>
<td>142±4</td>
</tr>
<tr>
<td>$G_{\text{max}}$, mm Hg$^{-1}$</td>
<td>1.07±0.12</td>
<td>0.44±0.12*</td>
<td>0.99±0.09‡</td>
<td>0.89±0.05</td>
</tr>
<tr>
<td>HR range, bpm</td>
<td>51±11</td>
<td>46±9</td>
<td>49±9</td>
<td>40±5</td>
</tr>
<tr>
<td>Maximum HR, bpm</td>
<td>359±12</td>
<td>380±17</td>
<td>362±10</td>
<td>352±11</td>
</tr>
<tr>
<td>Minimum HR, bpm</td>
<td>309±15</td>
<td>334±12</td>
<td>312±13</td>
<td>312±9</td>
</tr>
</tbody>
</table>

$MAP_0$ indicates median mean arterial pressure; $G_{\text{max}}$, maximal gain; MAP, mean arterial pressure; RSNA, renal sympathetic nerve activity; and HR, heart rate. Values are mean±SEM. Rat groups are as in Table 1.

*P<.01 compared with control.
†P<.05, ‡P<.01 compared with high salt.
greater G_{max} and lower MAP_{50} compared with the high salt/normal calcium group. There were no significant differences seen in maximal and minimal values as well as in the range of HR among the four groups.

The results of the MAP-RSNA and MAP-HR relations indicate that salt loading with normal calcium intake in young SHR impaired the overall property of the ABR and that simultaneous dietary calcium supplementation prevented this impairment.

**Reflex Changes in RSNA and HR by Electrical Stimulation of the Aortic Depressor Nerve**

One-second tetanic stimulation of the aortic depressor nerve reflexly elicited transient inhibition of RSNA with bradycardia (Fig 3), which vanished well before the next cycle of aortic depressor nerve stimulation.

At a stimulus frequency of 25 Hz, intended to elicit the ABR originating from unmyelinated aortic depressor nerve fibers and that simultaneous calcium supplementation prevented this impairment.

At a stimulus frequency of 100 Hz and an intensity of 1 to 10 V, intended to elicit the ABR originating from myelinated baroreceptor afferents, the percent decrease in RSNA was attenuated significantly in the high salt/normal calcium group compared with the control group (marked by an asterisk in Fig 4A). In the high salt/high calcium group, RSNA inhibition was significantly greater at stimulus intensities of 1 to 10 V compared with the high salt/normal calcium group (marked by a dagger in Fig 4A) but was not significantly different from that of the control or normal salt/high calcium groups. These data suggest that salt loading in young SHR centrally impaired the ABR mediated by myelinated aortic depressor nerve fibers and that simultaneous calcium supplementation prevented this impairment.

With respect to the reflex HR response, there were no significant differences among the four groups at a stimulus frequency of either 25 or 100 Hz (Fig 5).

**Discussion**

This study was intended to clarify the role of the sympathetic control mechanism in the development of salt-induced hypertension and the antihypertensive effect of a high calcium diet. For that purpose we examined whether high dietary intake of salt and/or calcium affected MAP, RSNA, HR, and the ABR in young SHR. In keeping with previous reports, high salt intake without calcium supplementation resulted in accelerated hypertension with renal sympathetic overactivity in young SHR. Furthermore, we obtained the following two major results: (1) A high salt diet impaired both overall and central properties of the ABR. Only the reflex arising from myelinated baroreceptor afferents was affected centrally. (2) A high calcium diet...
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Preservation of Baroreflex by Dietary Calcium

100 Hz, 1V  25 Hz, 5V

Control

High salt

High salt + Ca

High Ca

Fig 3. Peristimulus time histogram records of renal sympathetic nerve activity in response to electrical stimulation of the aortic depressor nerve. Records represent each of four groups of spontaneously hypertensive rats as defined in Fig 1. Histograms were constructed by superimposing 32 responses to tetanic stimulation of the aortic depressor nerve by a 1-second train of square-wave pulses having an intensity of 1 V and frequency of 100 Hz (left) or 5 V and 25 Hz (right). Horizontal bar above each tracing indicates duration of electrical stimulation. Abscissa is the number of renal nerve discharges recorded over 10 milliseconds.

In the present study, salt loading attenuated the reflex responses of RSNA and HR to stimulation of arterial baroreceptors in young SHR. This result is consistent with previous studies in hypertensive animal models and humans, with the exception of one report by Calhoun et al. They demonstrated that salt loading in salt-sensitive SHR for 2 to 3 weeks enhanced rather than attenuated the response of lumbar sympathetic nerve activity (LSNA) to stimulation of arterial baroreceptors with rats under conscious, unrestrained conditions. We suspect that the apparent discrepancy between their results and ours is primarily due to the following two differences in experimental conditions.

First, we selected RSNA, not LSNA, to monitor the response of the ABR. Although efferent renal sympathetic postganglionic fibers innervate vascular and non-vascular elements of the kidney, both groups of fibers behave in an identical manner so that RSNA can be considered to reflect the activity of sympathetic vasoconstrictor fibers. The lumbar sympathetic nerve on the other hand is composed of postganglionic muscle and skin fibers of various modalities such as vasomotor, sudomotor, and piloerector fibers. As demonstrated in single-fiber preparations in laboratory animals and microneurograms in humans, these fibers behave differently with respect to their rate and temporal pattern of spontaneous discharges as well as their responses to various inputs such as baroreceptor, chemoreceptor, or thermal stimulation. For this reason, these two sympathetic nerves may respond differently to baroreceptor stimulation in normotensive and/or hypertensive animals. In fact, the reflex change in RSNA to baroreceptor stimulation was found to be attenuated in SHR and renal hypertensive rabbits, whereas in LSNA it was not.

Prevented salt-induced accelerated hypertension and renal sympathetic overactivity. It also prevented the impairment of the overall and central properties of the ABR.

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In another series of experiments, we used young Wistar-Kyoto rats to examine the effects of halothane anesthesia on sympathetic activity in spontaneously hypertensive rats (SHR) and Wistar-Kyoto rats (WKY). We conducted these experiments to understand how halothane anesthesia affects the arterial baroreceptor reflex (ABR) in SHR, a model of hypertension.

Halothane has an inhibitory effect on the ABR in laboratory animals and humans, so it might have preferentially attenuated the arterial baroreceptor–renal sympathetic reflex of salt-loaded rats without calcium supplementation. However, this is unlikely for the following reasons: (1) In our experiments, during the recording period after the initial surgical procedure, the halothane concentration was reduced and maintained at 0.6% to 0.8%. At this level of halothane anesthesia, the reflex response of sympathetic fibers should be comparable to that of the conscious state. (2) In another series of experiments, we observed that salt loading significantly enhanced G_{max} of the MAP-RSNA relation in halothane-anesthetized young Wistar-Kyoto rats.

Evidence suggests that salt loading modifies the central neural control of sympathetic activity in SHR. For example, in SHR but not in WKY, a high salt intake resulted in a greater increase in RSNA induced by stressful environmental stimulation (air jet to the face). Furthermore, a recent study we conducted demonstrated that both hypothalamic and renal norepinephrine turnover rates were augmented in salt-loaded young SHR. Therefore, we investigated whether salt loading altered the central property of the arterial baroreceptor–RSNA reflex. This was demonstrated by salt-induced attenuation of the RSNA reflex response to electrical stimulation of myelinated fibers of the aortic depressor nerve, which consists exclusively of baroreceptor afferents. Interestingly, in both SHR and WKY, a resetting of the stimulus–response curve (a functional representation of the baroreceptor–afferent fiber complex) was greater in myelinated fibers than unmyelinated fibers. We should stress that the duration of electrical stimulation of the aortic depressor nerve to obtain the PSTH was intentionally set short (1 second) in the present study. As a result, repeated stimulation of the aortic depressor nerve caused minimal reflex changes in HR. This had little effect on MAP and did not distort the aortic depressor nerve–RSNA reflex. Therefore, lack of a significant alteration of the aortic depressor nerve–HR reflex in salt-loaded SHR does not automatically imply that this reflex is not impaired centrally.

As previously demonstrated, dietary calcium supplementation exerts an antihypertensive effect in hypertensive patients and animals and the sympathetic nervous system participates in this process. The present study examined whether simultaneous dietary calcium supplementation affected MAP, RSNA, HR, and the ABR in salt-loaded young SHR. A high calcium diet prevented salt-induced hypertension with sympathetic overactivity and impairment of the overall and central properties of the ABR. These findings are consistent with the report by Peuler et al in Dahl salt-sensitive rats, although they did not examine the renal component of the ABR. Our results clearly demonstrate that in salt-loaded young SHR, prevention of central impairment by a high calcium diet is associated with normal ABR function.

Evidence suggests a direct action of dietary calcium on the central nervous system. In angiotensin II–induced hypertensive rats to which calcium was given orally, calcium was transported into the central nervous system and caused a decrease in MAP. In addition, changes in calcium concentration in the lateral ventricle and in certain central nervous system regions such as the nucleus tractus solitarius and the site of projection of primary afferents of the aortic depressor nerve were associated with changes in MAP. Also, intraventricular infusion of calcium channel antagonists altered the baroreceptor reflex response of HR in rats. The possibility does remain, however, that a high calcium diet initially prevents salt-induced hypertension and then secondarily prevents deterioration of the ABR. Hypertension per se can also cause a resetting of the ABR, which may be reversed by antihypertensive drugs.

There are some possibilities that dietary calcium may lower arterial pressure in part because of the peripheral sympathetic mechanism. Hatton et al demonstrated that SHR fed a low calcium diet had larger pressor
responses to environmental stress compared with those fed a high calcium diet, although both rat groups showed similar plasma norepinephrine release during the stress. Moreover, a high calcium diet could attenuate development of hypertension and α1-adrenergic receptor-mediated responses relative to a low calcium diet in young SHR. These results suggest that altered postsynaptic responses to adrenergic agonists are involved in the antihypertensive effect of dietary calcium. We are aware that the contents of NaCl and calcium in the diet used in this study are high and do not necessarily reflect clinically relevant values. We had three reasons for using diets containing 8.00% NaCl and/or 4.07% calcium. First, diets containing 8% NaCl have been widely used in SHR to increase arterial pressure without undesirable side effects. Second, diets containing 4% calcium have been consistently shown to reduce arterial pressure and/or sympathetic activity in hypertensive animal models. By contrast, there does not seem to be general agreement as to whether diets containing less than 4% calcium lower overall and central properties of the arterial baroreceptor reflex, all of which were prevented by simultaneous dietary calcium supplementation. These results suggest that an unimpaired baroreflex reflex with intact peripheral and central mechanisms underlies the antihypertensive effect of a high calcium diet.

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