Effects of Valsartan on Mechanical Properties of the Carotid Artery in Spontaneously Hypertensive Rats Under High-Salt Diet

Carlos Labat, Patrick Lacolley, Malika Lajemi, Marc de Gasparo, Michel E. Safar, Athanase Benetos

Abstract—The aim of this investigation was to examine the preventive effects of valsartan, an AT₁ receptor antagonist, on blood pressure, carotid artery (CA) structure, and functional elastic properties in SHR maintained on different sodium diets.

Methods

All procedures were carried out in accordance with institutional guidelines for animal experimentation. SHR (n = 58) male rats that were 4 weeks of age were obtained from Iffa Credo (France).

In SHR, a HSD (7% NaCl in the food) was administered from the 10th to 20th week of age. Control SHR received a normal-salt diet (NSD; 0.4% NaCl) during the same period. Within each of the 2 groups, the animals received treatment with either placebo or valsartan (30 mg · kg⁻¹ · d⁻¹) administered on the 4th to 20th week of age. Arterial pressure, wall stress, incremental elastic modulus (Einc), medial cross-sectional area, and EIIIA fibronectin isoform were significantly increased in placebo-HSD rats compared with placebo-NSD rats with no change in the ratio of collagen to elastin. Valsartan reduced mean arterial pressure in both NSD and HSD rats but reduced pulse pressure only in NSD rats. In NSD rats, valsartan reduced Einc and medial cross-sectional area. In HSD, valsartan increased Einc and did not modify medial cross-sectional area and fibronectin. In valsartan-treated rats, the ratio of collagen to elastin was greater in HSD than in NSD rats. In conclusion, the effects of AT₁ blockade are greatly influenced by salt intake in SHR. Despite a reduction in mean arterial pressure in HSD rats, AT₁ blockade was not able to prevent the effects of a HSD on pulse pressure, carotid artery stiffness, and hypertrophy. (Hypertension. 2001;38:439-443.)

Key Words: salt □ angiotensin II □ AT₁ blockade □ large artery stiffness □ carotid artery

Previous reports have shown that plasma renin-angiotensin activity is reduced in rats receiving a high-salt diet (HSD). However, a long-term HSD in salt-sensitive animals—Dahl salt-sensitive rats, stroke-prone hypertensive rats, or ANP knockout mice—is known to stimulate plasma renin activity. These observations suggest that the effects observed during perturbation of the renin-angiotensin aldosterone system by a high sodium intake may depend on the animal model used. Recently, Wang and Du observed an increased expression of angiotensin II type 1 (AT₁) receptor mRNA in arterial preparations derived from Wistar and Sprague-Dawley rats maintained on a HSD. In contrast, when Dahl rats were treated with a HSD, decreases in aortic mRNA and AT₁ receptor density were observed. Together, these results suggested that 1 modification associated with high sodium intake may be at the level of the AT₁ receptor. Interestingly, in spontaneously hypertensive rats (SHR) treated with a HSD, an enhanced functional response to angiotensin II and AT₁ receptor antagonists was demonstrated, suggesting that sodium modified AT₁ activity in SHR. Although AT₁ receptor antagonists have been shown to reduce mortality in salt-sensitive animal models (Dahl rats or stroke-prone rats), there is little evidence available that AT₁ receptor antagonists continually block the elevated blood pressure and arterial abnormalities observed in SHR during high sodium intake.

The aim of this investigation was to examine the preventive effects of valsartan, an AT₁ receptor antagonist, on blood pressure, carotid artery (CA) structure, and functional elastic properties in SHR maintained on different sodium diets.
TABLE 1. Effect of Valsartan on Body Weight, Blood Pressure, and CA Parameters in SHR Receiving a NSD or a HSD

<table>
<thead>
<tr>
<th></th>
<th>NSD Placebo (n=14)</th>
<th>NSD Valsartan (n=15)</th>
<th>HSD Placebo (n=13)</th>
<th>HSD Valsartan (n=14)</th>
<th>P Interaction (ANOVA)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight, g</td>
<td>412±8</td>
<td>389±10</td>
<td>382±8†</td>
<td>378±11</td>
<td>NS</td>
</tr>
<tr>
<td>Mean arterial pressure, mm Hg</td>
<td>182±6</td>
<td>144±3*</td>
<td>214±6†</td>
<td>180±5††</td>
<td>NS</td>
</tr>
<tr>
<td>PP, mm Hg</td>
<td>60±3</td>
<td>42±3*</td>
<td>69±2†</td>
<td>69±3††</td>
<td>&lt;0.002</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>349±11</td>
<td>336±10</td>
<td>347±12</td>
<td>363±11</td>
<td>NS</td>
</tr>
<tr>
<td>Distensibility, mm Hg · 10^-3</td>
<td>2.01±0.16</td>
<td>4.43±0.40*</td>
<td>1.37±0.11†</td>
<td>1.61±0.21†</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Einc, kPa</td>
<td>1176±121</td>
<td>539±53*</td>
<td>1659±137†</td>
<td>1679±279††</td>
<td>0.07</td>
</tr>
<tr>
<td>Wall stress, kPa</td>
<td>210±9</td>
<td>168±9*</td>
<td>242±11†</td>
<td>198±12††</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are mean±SEM.
*P<0.05 vs placebo for the same salt diet; †P<0.05, HSD vs NSD for the same treatment.

Results

Effects of Valsartan on Blood Pressure and CA

All SHR maintained on a NSD survived the treatment period. Of 15 SHR receiving a HSD in the placebo group, 2 (13%) died compared with none of the valsartan-treated rats. SHR maintained on a HSD had lower body weight and higher mean arterial pressure (MAP) and pulse pressure (PP) compared with those of rats receiving a NSD (Table 1). Arterial distensibility was lower, with no change in arterial diameter. The Einc–wall stress curve in HSD rats receiving placebo was shifted to the right in the prolongation of the Einc–wall stress curve observed in the placebo-NSD rats (Figure 1). No significant differences in collagen, elastin densities, and ratio of collagen to elastin were observed between the 2 groups. The higher collagen and elastin contents under a HSD were related to the significant increase in CA MCSA (Table 2) because MCSA was correlated with MAP \((r=0.63)\) and PP \((r=0.80)\).

Valsartan reduced MAP similarly in both NSD and HSD rats (21% versus 16%) (Table 1). A reduction in carotid PP was observed only in NSD rats treated with the AT₁ antagonist. Valsartan administered to NSD rats produced a significant decrease in carotid diameter and a 2-fold increase in arterial distensibility. Valsartan did not modify arterial diam-
higher level of wall stress with no changes in the intrinsic
creased stiffness of the CA in the HSD group was due to a

effect on PP, arterial wall hypertrophy, and stiffness.

In this study, long-term treatment of SHR with the AT1
receptor antagonist valsartan was investigated in animals
in SHR, salt loading has been shown to result in a significant thickening of the aortic media between 10 and 20 weeks of age.26 This increase in wall thickness is associated with an enhanced accumulation of Fn. Previous findings in genetic hypertensive rats indicate marked interactions between a HSD, increased wall thickness, and increase in Fn.27–30 The increase in wall thickness and Fn may contribute to the increase in vascular wall elastic modulus in proportional to the level of circumferential wall stress through an increased number of cell matrix attachments sites.22

Interestingly, the decrease in MAP in HSD rats treated with valsartan was not associated with a significant reduction in CA wall thickness. One possible explanation is that despite similar changes in MAP with valsartan in NSD- and HSD-fed rats, blood pressure values at the end of the treatment period were still too high in HSD rats (180 mm Hg). We can suggest that MAP should achieve a lower blood pressure threshold to significantly reduce arterial wall hypertrophy and to improve arterial compliance. The persistence of arterial wall hypertrophy may also be explained by the absence of reduction in PP, which is a main determinant of arterial hypertrophy.31 Augmentation of Einc for a given value of wall stress in valsartan-treated HSD rats compared with all other groups demonstrates a marked increase in intrinsic stiffness of the wall material. The antihypertensive efficacy of AT1 antagonism, despite a NaCl-induced decrease in the renin-angiotensin system, may be explained by an increase in AT1 receptor messenger RNA levels10 and by an increase in AT1 receptor density.32 The present study shows that AT1 blockade in the presence of a HSD was also associated with an increase in the ratio of collagen to elastin compared with the HSD rats receiving the same treatment. These data support the suggestion that this increase is at least partially responsible for the CA wall stiffness observed in HSD rats receiving the AT1 treatment (Figure 1). The decrease in mean wall stress by valsartan was not reduced in HSD rats treated with valsartan.


discussion

In this study, long-term treatment of SHR with the AT1 receptor antagonist valsartan was investigated in animals maintained on a HSD. The HSD induced an increase in MAP and PP associated with carotid artery hypertrophy and wall stiffness. Valsartan significantly reduced MAP but had no effect on PP, arterial wall hypertrophy, and stiffness.

Analysis of Einc–wall stress curves showed that the increased stiffness of the CA in the HSD group was due to a higher level of wall stress with no changes in the intrinsic elastic properties of the vascular wall. Preservation of elastic properties is in accordance with the absence of modification in collagen and elastin densities and the ratio of collagen to elastin with a HSD. In SHR, salt loading has been shown to result in a significant thickening of the aortic media between 10 and 20 weeks of age.26 This increase in wall thickness is associated with an enhanced accumulation of Fn. Previous findings in genetic hypertensive rats indicate marked interactions between a HSD, increased wall thickness, and increase in Fn.27–30 The increase in wall thickness and Fn may contribute to the increase in vascular wall elastic modulus in proportional to the level of circumferential wall stress through an increased number of cell matrix attachments sites.22

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### Table 2. Effect of Valsartan on CA Composition in SHR Receiving a NSD or a HSD

<table>
<thead>
<tr>
<th></th>
<th>NSD (n=14)</th>
<th>Valsartan (n=15)</th>
<th>HSD (n=13)</th>
<th>Valsartan (n=14)</th>
<th>Interaction (ANOVA)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MCSA, mm² · 10⁻³</td>
<td>287±9</td>
<td>223±11*</td>
<td>325±12†</td>
<td>319±18†</td>
<td>&lt;0.03</td>
</tr>
<tr>
<td>Nucleus surface, µm²</td>
<td>6.2±0.2</td>
<td>6.1±0.2</td>
<td>6.1±0.2</td>
<td>7.2±0.2†</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Nucleus number/1-mm section</td>
<td>294±11</td>
<td>284±16</td>
<td>275±12</td>
<td>280±11</td>
<td>NS</td>
</tr>
<tr>
<td>Collagen density, %</td>
<td>16.9±0.6</td>
<td>16.9±0.6</td>
<td>17.7±0.8</td>
<td>17.5±0.6</td>
<td>NS</td>
</tr>
<tr>
<td>Collagen content, mm² · 10⁻³</td>
<td>49±2</td>
<td>37.2±2*</td>
<td>58±4†</td>
<td>51±5†</td>
<td>NS</td>
</tr>
<tr>
<td>Elastin density, %</td>
<td>36.2±1.1</td>
<td>38.8±1.8</td>
<td>36.9±1.6</td>
<td>31.0±1.4†</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Elastin content, mm² · 10⁻³</td>
<td>103±3</td>
<td>86.5±5*</td>
<td>121±8†</td>
<td>94±10*</td>
<td>NS</td>
</tr>
<tr>
<td>Collagen/elastin ratio</td>
<td>0.57±0.03</td>
<td>0.56±0.02</td>
<td>0.57±0.04</td>
<td>0.69±0.03†</td>
<td>&lt;0.05</td>
</tr>
</tbody>
</table>

*P<0.05 vs placebo for the same salt diet; †P<0.05, HSD vs NSD for the same treatment.
expression. In contrast, in the presence of a HSD, valsartan was not able to reduce Fn content. Therefore, persistence of arterial wall rigidity in HSD valsartan-treated rats may also be explained by the maintenance of relatively high levels of Fn despite the decrease in wall stress.

The absence of a reduction in PP by the AT1 antagonist may be responsible for incomplete results on mortality reduction in SHR receiving a HSD. Clinical studies have pointed out the predominant role of PP in the cardiovascular morbidity and mortality in several populations, making PP a major cardiovascular risk factor independent of MAP. Previous studies have shown that salt is a determinant of aortic stiffness and arterial wall hypertrophy. PP may aggravate the effects of a HSD on vascular structure and cardiovascular morbidity and mortality observed in experimental models of hypertension.

In conclusion, the present study showed that the effects of AT1 blockade are greatly influenced by high salt intake in SHR. Valsartan reduced MAP but was not able to diminish large artery stiffness and hypertrophy and PP.

Acknowledgments
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