Rapid Communications

Association of Salt Sensitivity in Rats With Genes of the Major Histocompatibility Complex

Jaroslav Kuneš, Josef Zicha

Abstract  Dietary sodium intake has long been considered an important factor in the genesis and maintenance of hypertension in both humans and experimental animals. To identify the possible association between salt sensitivity and genes of the major histocompatibility complex (RT1 complex), we studied the blood pressure response to an 8% NaCl diet in normotensive Lewis rats, spontaneously hypertensive rats (SHR), and Lewis.1K congenic rats (congenic to Lewis with the SHR main histocompatibility complex RT1). During the first 4 weeks of a high salt diet, the blood pressure increase was the same in SHR and Lewis.1K congenic rats. Thus, the presence of a small segment of SHR chromosome 20 with genes of the RT1 complex (and closely related genes) in the Lewis genome sensitized the blood pressure of these animals to the hypertensive effects of a high salt diet. Genes of the RT1 complex influenced the salt-induced increase of relative kidney weight more than that of relative heart weight. Our results support the hypothesis that some alleles within or close to the RT1 complex might be responsible for the higher sensitivity of hypertensive individuals to certain environmental stressors, including high salt intake. (Hypertension. 1994;24:645-647.)

Key Words • genetics • hypertension, spontaneous • major histocompatibility complex • salt • blood pressure

Considerable information has accumulated about the importance of salt in the etiology of hypertension, but the precise pathogenetic mechanisms have not yet been resolved. Nevertheless, there is a group of patients with essential hypertension who are salt sensitive; ie, their blood pressure increases when they are offered extra NaCl and falls when a low salt diet is given.1,2 Weinberger et al3 have shown that 51% of human hypertensive individuals but only 26% of normotensive individuals are salt sensitive.

The concept of resistance and sensitivity to salt was largely developed in rats by L.K. Dahl,4 who produced substrains susceptible and resistant to the hypertensive effects of long-term salt loading. Later, specific substrains of spontaneously hypertensive rats (SHR) were identified that are salt sensitive and salt resistant.5,6 We have found that SHR from our Prague colony are highly salt sensitive when fed a diet supplemented with 8% NaCl. Moreover, studies in humans8 and in the Prague set of recombinant inbred strains9 have suggested that genes within or close to the major histocompatibility complex may be linked to the regulation of blood pressure.

In the present study we tested the possibility of whether the genes of the major histocompatibility complex in the rat (RT1 complex) are associated with salt sensitivity.

Methods

Experiments were performed in male Lewis rats, SHR, and Lewis.1K congenic rats bred in our institute. Lewis.1K congenic rats10 represent animals in which the RT1 k haplotype from SHR was transferred to the genetic background of the Lewis strain. Because Lewis.1K rats of our colony were produced by 16 backcrosses (Dr M. Pravenecek, personal communication), the degree of genetic similarity between the Lewis and Lewis.1K strains must be greater than 99%.11 All animals were kept in a temperature-controlled room (23±1°C), were fed a standard diet (1% NaCl), and drank tap water ad libitum. One half of the animals in each group were kept on a high salt diet (8% NaCl) from 6 weeks of age. All procedures followed were in accordance with the guidelines of the Ethics Committee of our institute.

Systolic blood pressure was measured by the same person and at the same time of day in conscious preheated rats once a week by the tail-cuff method. Each rat was acclimated to the holder and tail cuff by three training sessions before the start of definitive blood pressure measurements. The sizes of the holder and tail cuff were increased during the experiment to match the increase in body size. At each session, the mean of at least five readings was taken as the value of systolic blood pressure for each animal. At the end of the experiment, blood pressure values were verified in conscious 12-week-old animals by direct measurement of systolic, mean arterial, and diastolic blood pressures. Briefly, the left carotid artery was cannulated with rats under pentobarbital anesthesia approximately 20 hours before blood pressure determination. Blood pressure was measured on the following day in conscious unrestrained rats for approximately 30 minutes with Statham P23 dB transducers connected to a four-channel Hewlett-Packard recorder.

After blood pressure measurement, animals were killed and the heart (both ventricles without atria) and kidneys were removed and weighed. The relative organ weight was calculated per 100 g body weight.

Data are expressed as mean±SEM. Comparisons among groups were performed with one-way ANOVA. Values of P<.01 were considered statistically significant.

Results

The Figure shows the effects of an 8% NaCl diet on tail-cuff blood pressure. The high salt diet elicited only a moderate increase of systolic blood pressure in the
Lewis strain. On the other hand, during the first 4 weeks of a high salt diet, blood pressure rose similarly in Lewis.1K congenic rats and SHR. This suggested that of a high salt diet, blood pressure rose similarly in the Lewis strain. On the other hand, during the first 4 weeks of feeding the high salt diet induced additional blood pressure elevation in SHR only (Figure). Direct blood pressure measurements at the end of the experiment (Table) confirmed systolic blood pressure values obtained by the tail-cuff method at the age of 12 weeks.

Moreover, the response of mean arterial and diastolic blood pressures to the high salt intake in the individual strains was similar to that of systolic blood pressure (Table).

The influence of the RT1 complex on changes of relative heart and kidney weights induced by the high salt diet was different in both of these organs (Table). The genes within or close to the SHR RT1 complex had no effect on relative heart weight because the salt-induced increase of this parameter was the same in the Lewis.1K congenic strain as in the Lewis strain (9% versus 11%, respectively). On the other hand, the high salt diet increased relative kidney weight more in Lewis.1K congenic rats than in Lewis rats (27% versus 17%, respectively). The marked increase in relative organ weights observed in SHR fed the 8% NaCl diet might be related to the extreme blood pressure elevation in this group (Table).

Discussion

In our study we have tried to elucidate the role of genes of the major histocompatibility complex (RT1 complex) in the salt sensitivity of SHR. Using the Lewis.1K congenic strain, we demonstrated that at least part of the blood pressure sensitivity to a high salt diet could be associated with genes within or close to the RT1 complex. This is in good agreement with the hypothesis that the changes of several genes within the RT1 complex (ie, 21-hydroxylase, tumor necrosis factor-α, hsp70, etc) might be responsible for the higher sensitivity of hypertensive individuals to certain environmental stimuli. This is also supported by the previous results in the Prague set of recombinant inbred strains in which we found a significant association between the RT1 complex and blood pressure. It was even shown in humans with essential hypertension that at least one of the genes responsible for the genetic susceptibility to this disease is located in or near the human leukocyte antigen complex. On the other hand, the study of Lodwick et al failed to prove the association of blood pressure with the hsp70 gene within the major histocompatibility complex. This suggests that gene-gene or gene-environment inter-

**Table: Characteristics of 12-Week-Old Lewis, Lewis.1K Congenic, and Spontaneously Hypertensive Rats Fed Normal (1% NaCl) or High Salt (8% NaCl) Diet**

<table>
<thead>
<tr>
<th>Strain and Diet</th>
<th>n</th>
<th>BW, g</th>
<th>SBP, mm Hg</th>
<th>MAP, mm Hg</th>
<th>DBP, mm Hg</th>
<th>HW/BW, mg/100 g</th>
<th>KW/BW, mg/100 g</th>
</tr>
</thead>
<tbody>
<tr>
<td>LEW 1% NaCl</td>
<td>12</td>
<td>271±12</td>
<td>130±4</td>
<td>107±3</td>
<td>89±3</td>
<td>242±2</td>
<td>650±6</td>
</tr>
<tr>
<td>8% NaCl</td>
<td>11</td>
<td>285±17</td>
<td>141±4</td>
<td>107±5</td>
<td>82±6</td>
<td>272±3*</td>
<td>760±8*</td>
</tr>
<tr>
<td>LEW.1K 1% NaCl</td>
<td>20</td>
<td>223±3</td>
<td>139±3</td>
<td>109±2</td>
<td>88±2</td>
<td>247±2</td>
<td>645±8</td>
</tr>
<tr>
<td>8% NaCl</td>
<td>20</td>
<td>245±8*</td>
<td>170±3*</td>
<td>137±4*</td>
<td>114±4*</td>
<td>270±5*</td>
<td>818±9*†</td>
</tr>
<tr>
<td>SHR 1% NaCl</td>
<td>15</td>
<td>260±5</td>
<td>191±8†</td>
<td>161±6*</td>
<td>133±4†</td>
<td>334±5†</td>
<td>617±5†</td>
</tr>
<tr>
<td>8% NaCl</td>
<td>14</td>
<td>239±8*</td>
<td>262±18†</td>
<td>211±15†</td>
<td>171±11†</td>
<td>519±20†</td>
<td>951±62†</td>
</tr>
</tbody>
</table>

BW indicates body weight; SBP, systolic blood pressure; MAP, mean arterial pressure; DBP, diastolic blood pressure; HW/BW, heart weight–body weight ratio; KW/BW, kidney weight–body weight ratio; LEW, Lewis rats; LEW.1K, Lewis.1K congenic rats; SHR, spontaneously hypertensive rats. Values are mean±SEM.

*P<.01 vs 1% NaCl.
†P<.01 vs LEW (fed the same diet).
actions could be responsible for the discrepant results obtained in different crosses. Thus, other genes out of the respective congenic segment might cause the additional late blood pressure rise in salt-loaded SHR. The genetic bases and mechanisms of salt-sensitive blood pressure changes are complex, multifactorial, and probably interrelated. There is evidence that neural mechanisms, central and peripheral effects of vasopressin, atrial natriuretic factor, norepinephrine, or angiotensin II, increased membrane permeability for sodium, and many other mechanisms could be implicated in salt-sensitive blood pressure changes. This multifactorial mechanism provides convincing evidence for a polygenic component of the salt sensitivity of blood pressure. Unfortunately, no appreciable progress in identifying specific genes for salt sensitivity has yet been made. Weinberger et al reported that among human genetic markers only phenotypes of haptoglobin were found to be associated with differences in salt sensitivity. Involvement of the immune system in the pathogenesis of spontaneous hypertension is highly probable. Recently, Fannon et al demonstrated that immune system abnormalities were present even in prehypertensive SHR, supporting the hypothesis that this disturbance precedes and does not adapt to the hypertensive status.

Because of the complexity of the pathophysiology of hypertension, it is rather difficult to elucidate the exact relation between the major histocompatibility complex and salt-induced blood pressure rise. Further study will be necessary to define more clearly by which mechanisms the altered gene or genes in the histocompatibility complex could increase blood pressure and induce higher sensitivity of hypertensive individuals to environmental stimuli including salt intake.

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References

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