Environmental and Physiological Characteristics in Adolescents
Genetically Predisposed to Hypertension

SATORU FUJISHIMA, M.D., OSAMU TOCHIKUBO, M.D.
AND YOSHIHIRO KANEKO, M.D.

To clarify the possible risk factors for the development of hypertension, we examined the influences of heredity and environment on blood pressure regulation and whether or not the physiological condition differed in high school students with different levels of blood pressure. A borderline hypertensive (BH) group, consisting of 75 male students with systolic blood pressure (SBP) consistently above 140 mmHg on two separate occasions, was compared to a normotensive (N) group of 84 male students with SBP below 130 mmHg. In the BH group, 43% of students had a family history of hypertension within two generations of relatives, while 18% had one in the N group (p < 0.05). The BH group was characterized by a gain in weight, a slight increase in 24-hour urinary sodium excretion, a higher heart rate, elevated values of plasma renin and urinary aldosterone, and an elevated sodium concentration in erythrocytes. Nevertheless, urinary excretion of potassium and kallikrein did not differ between the two groups.

In each group, students with familial hypertension had a significantly (p < 0.05) lower 24-hour urinary kallikrein excretion than those without it. Although kallikrein excretion correlated fairly well with aldosterone excretion (r = 0.47, p < 0.01) or creatinine clearance (r = 0.59, p < 0.01) in the BH students without familial hypertension, no such correlations were found in those with familial hypertension.

These results indicate that the abnormal relationships of aldosterone to kallikrein metabolism and of kallikrein to renal function control may be involved as hereditary factors in the development of hypertension.

THE importance of genetic and environmental effects on arterial blood pressure has been well established in experimental animal models and in humans. In the Dahl salt-sensitive and salt-resistant rats, susceptibility to hypertension is definitely a function of differences in genetic substrate with reference to sodium intake.

In human adults, blood pressure levels are likely to associate with race, sex, dietary sodium intake, body weight, psychosocial stress and socioeconomic background. Recently it has been reported in epidemiological surveys, including longitudinal and cross-sectional observations on a large population of children, that a significant tracking of blood pressure levels occurs in early life and that a familial aggregation of blood pressure has been shown in childhood. However, factors

Key Words:
- Urinary kallikrein excretion
- Borderline hypertension
- Adolescents
- Heredity
- Renin-angiotensin-aldosterone system

The Second Department of Internal Medicine, Yokohama City University, School of Medicine, Yokohama, Japan
This work was supported in part by a Scientific Grant from the Japanese Ministry of Education, Sciences and Cultures in 1981.
Mailing address: Satoru Fujishima, M.D., The Second Department of Internal Medicine, Yokohama City University, School of Medicine, 3-46 Urafune-cho, Yokohama 232, Japan

Japanese Circulation Journal Vol. 47, February 1983
associated with blood pressure levels have not been studied extensively in children and adolescents.

A number of genetic abnormalities related to blood pressure regulation have been documented in individuals who had a family history of hypertension. Zinner et al.\textsuperscript{15} have suggested that there is a concomitant genetic influence on both blood pressure and renal kallikrein which is involved in the production of potent vasodilator peptides. Hollenberg et al.\textsuperscript{16} have reported a decrease in renal plasma flow with a concordant rise in plasma renin activity in normotensive offspring of the hypertensives. In addition, the recent in vitro studies have demonstrated the differences in sodium movement across cell membranes in first degree relatives of the hypertensive patients.\textsuperscript{17–19} Hence, such abnormalities may serve to explain, at least in part, the heritable nature of hypertension and the different susceptibilities to the development of essential hypertension.

The purpose of our study was to determine the environmental and physiological factors in 2 groups of high school students with different blood pressure levels as a possible means of identifying the individual who may have a greater risk for essential hypertension. Furthermore, this study was performed to examine the association between urinary kallikrein and factors related to blood pressure regulation, such as urinary electrolyte excretions, aldosterone values, and the glomerular filtration rate, in 2 subgroups of borderline hypertensive students divided by the presence or absence of a family history of hypertension. This latter study was designed to determine a possible heritable role of the renal kallikrein-kinin system in blood pressure regulation.

\textbf{MATERIALS AND METHODS}

In the autumn of 1980, we asked 5 public schools in Kanagawa for permission to examine the blood pressure of students. The first blood pressure measurement of 2,973 high school students, 1,489 males and 1,484 females ranging in age from 15 to 18 years, was carried out in their schools from February to March, 1981. Casual blood pressure reading was taken twice after 5 min rest in a sitting position using a random-zero mercury sphygmomanometer and phases I and V were recorded. Heart rate was counted from the radial pulse for 30 sec. Their height and weight were also recorded.

Based on this systolic blood pressure (SBP), 216 students with an SBP consistently exceeding 135 mmHg were selected for further examination. They were 169 male students and 47 female students. To facilitate comparisons, a group of normotensive healthy students with an SBP consistently below 130 mmHg was also studied. The second examination, including the second blood pressure measurement, was performed 2 weeks later. After explaining our research procedures in detail, the consents were obtained from both the students and their parents. On the basis of a questionnaire given to the students and their parents, the family history of hypertension and hypertensive complications was studied in regard to two generations of relatives.

According to the SBP on the 2 separate occasions mentioned above, students were divided into the following 3 groups: 1) those with borderline hypertension whose SBP were consistently above 140 mmHg in both measurements, 2) those with normotension with SBP consistently below 130 mmHg and 3) others. In the present study the results obtained from the 75 males who met the criteria for borderline hypertensive (BH) group and the 84 males who met the criteria for normotensive (N) group. Among the 47 females with SBP above 135 mmHg at first examination, only about one-half participated in the second examination and 8 of them were in the BH group. Therefore, we excluded the female data from the analysis.

On the day prior to the second examination, each subject was requested to collect his urine for a 24-hour period for the determination of urinary sodium, potassium, creatinine, aldosterone and kallikrein contents. For collecting 24-hour urine, we used a newly devised portable cup as described elsewhere.\textsuperscript{20} In brief, it is a double-bottomed cup with a pipe-shaped scale and a cock leading to the lower compartment. The cup is so devised that 1% of the urine volume excreted into the upper compartment each time remains in the pipe-scale, and then comes down into the lower compartment by manipulation of the cock.

On the day of the second examination, blood pressure was measured in a sitting position. Thereafter, a 12-lead electrocardiogram was taken at rest in a supine position. In addition, venous blood samples were obtained for measurements of sodium, potassium and creatinine levels, plasma renin activity and erythrocyte sodium
TABLE I CHARACTERISTICS OF BORDERLINE HYPERTENSIVE (BH) AND NORMOTENSIVE (N) GROUPS

<table>
<thead>
<tr>
<th></th>
<th>Values</th>
<th>No. of cases</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>BH</td>
<td>N</td>
<td>BH</td>
</tr>
<tr>
<td>Blood pressure (mmHg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>systolic</td>
<td>148 ± 1</td>
<td>117 ± 1</td>
<td>75</td>
</tr>
<tr>
<td>Diastolic</td>
<td>73 ± 1</td>
<td>66 ± 1</td>
<td>75</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>82 ± 2</td>
<td>72 ± 1</td>
<td>75</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>170.3 ± 0.7</td>
<td>169.0 ± 0.7</td>
<td>75</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>63.5 ± 1.2</td>
<td>58.2 ± 0.7</td>
<td>75</td>
</tr>
<tr>
<td>Urinary</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>volume (L/day)</td>
<td>1.02 ± 0.04</td>
<td>0.93 ± 0.04</td>
<td>69</td>
</tr>
<tr>
<td>creatinine (g/day)</td>
<td>1.72 ± 0.06</td>
<td>1.52 ± 0.04</td>
<td>69</td>
</tr>
<tr>
<td>Na (mEq/day)</td>
<td>228 ± 12</td>
<td>194 ± 9</td>
<td>69</td>
</tr>
<tr>
<td>K (mEq/day)</td>
<td>45.0 ± 2.0</td>
<td>42.3 ± 1.8</td>
<td>69</td>
</tr>
<tr>
<td>aldosterone (µg/day)</td>
<td>6.82 ± 0.53</td>
<td>4.54 ± 0.31</td>
<td>68</td>
</tr>
<tr>
<td>kallikrein (unit/day)</td>
<td>79.3 ± 10.7</td>
<td>92.8 ± 11.9</td>
<td>68</td>
</tr>
<tr>
<td>Plasma renin activity (ng/ml/hr)</td>
<td>1.8 ± 0.2</td>
<td>1.0 ± 0.1</td>
<td>25</td>
</tr>
<tr>
<td>Erythrocyte Na concentration (mmol/L-RBC)</td>
<td>9.6 ± 0.3</td>
<td>8.5 ± 0.3</td>
<td>44</td>
</tr>
<tr>
<td>Incidence (%) of familial hypertension</td>
<td>43</td>
<td>18</td>
<td>75</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SEM. p = statistical significance of the difference between the BH and N groups.

Sodium, potassium and creatinine concentrations in blood and urine were measured using an IL flame photometer and standard autoanalyzer technique (Nippon Denshi, MS-24). Plasma renin activity was measured by radioimmunoassay and urinary aldosterone concentration was determined by direct radioimmunoassay using a commercial kit (CEA-IRE-SORIN). Kallikrein activity was measured by the method of Morita et al using a fluorogenic peptide substrate, prolyl-phenylalanyl-arginine-4-methylcoumaryl-7-amide. Urine aliquots were assayed immediately after dialyzing against running tap-water for 24 hours. One unit was defined as a 10 µM solution of 7-amino-4-methylcoumarin liberated per minute at pH 8.0 and 37°C. The intraassay coefficient of variation was 3.0% (n = 9) and the interassay one was 6.2% (n = 12). We found a significant correlation in the urinary kallikrein concentration between this method and the direct radioimmunoassay measurement of glandular kallikrein (r = 0.90, n = 31). The sodium concentration in the erythrocytes was determined using a newly devised method as described elsewhere.

All values were given as mean ± SEM. Non-paired Student's t-test were used for statistical analyses between the groups. A significant difference was considered to exist when p value was less than 0.05.

RESULTS

Characteristics of the study groups are given in Table I. Seventy-five male students belonged to the BH group, representing 5% of the original male students. The prevalence of hypertension or hypertensive complications was found in 43% of the families in the BH group, being significantly higher as compared to 18% in the N group.

The students in the BH group had higher heart rates than those in the N group. Although there was no significant difference in the height between the 2 groups, the weight in the BH group was 63.5 ± 1.2 kg, being significantly different (p < 0.001) from that of the N group, 58.2 ± 0.7 kg. Twenty-four-hour urinary excretion of potassium did not differ in both groups. However, the BH group had a higher 24-hour urinary
excretion of sodium, a higher plasma renin activity and a higher 24-hour urinary aldosterone excretion as compared to the N group. There was a tendency for the sodium concentration in erythrocytes to be slightly higher in the BH group than in the N group. In the BH group, the sodium concentration in erythrocytes was similar between the students with a family history of hypertension (9.6 ± 0.4 mmol/L-RBC) and without it (9.5 ± 0.3), while in the N group it was significantly higher (p < 0.05) in the students with a family history of hypertension (9.1 ± 0.4 mmol/L-RBC) than those without it (8.2 ± 0.3).

Urinary kallikrein excretion for 24 hours did not differ between the two groups. In both groups, however, higher 24-hour urinary excretion of kallikrein was observed in the students without a family history of hypertension than in the students with it (105.0 ± 2.4 vs 47.5 ± 9.1 unit/day in the BH group and 116.5 ± 7.4 vs 57.2 ± 20.0 in the N group, p < 0.05, respectively).

In the BH group, no statistically significant correlation was found between 24-hour urinary kallikrein excretion and sodium excretion between the students with and without familial hypertension. In this group, however, a positive correlation was found between urinary kallikrein excretion and potassium excretion in 42 students without familial hypertension (r = 0.42, p < 0.01), while this correlation was not evident in 26 students with familial hypertension (r = 0.28, ns). In the BH group without familial hypertension, urinary kallikrein excretion also correlated positively to both urinary aldosterone excretion (r = 0.47, p < 0.01) and creatinine clearance (r = 0.59, p < 0.01) but in the BH group with familial hypertension these correlations were not observed.

Creatinine clearance was 140 ± 5 L/day/1.48 m² in BH students without familial hypertension and 141 ± 6 in those with familial hypertension, showing no significant difference. When the creatinine clearance was plotted against SBP, a significant inverse correlation was obtained in the BH students without familial hypertension (r = −0.44, p < 0.05), but not in the students with familial hypertension.

**DISCUSSION**

In our present study, 75 of 1,489 male students (5%), ranging in age from 15 to 18 years, were found to have borderline hypertension with systolic blood pressure above 140 mmHg in 2 separate measurements. The Task Force on Blood Pressure Control in Children has recently recommended that sustained blood pressure levels above 95 percentile according to age should be considered abnormal. There is now good evidence that the blood pressure at a young age can predict the blood pressure at an older age and that this tracking phenomenon may begin in early life. Therefore, high school students with borderline hypertension may have a greater risk for developing established hypertension as compared to normotensive students.

Nearly one-half of the BH group had a family history of hypertension or hypertensive complications within two generations of relatives, while it was observed in only 18% of the students in the N group. The hereditary nature of essential hypertension has been repeatedly demonstrated, even after taking into account the influence of a common environment. In a study of 1,524 members of 277 families, Ayman has found a susceptibility to hypertension in the offsprings of hypertensive parents. Thus, it follows that an abnormality, which is demonstrable in the adolescents of hypertensive families, but which cannot be found in the adolescents of normotensive families, may well be related to the pathogenesis of hypertension.

In our study, the students in the BH group manifested a number of characteristics which clearly distinguished them from the students in the N group. The former students were heavier in weight in agreement with the findings that individuals who gain more weight show a greater blood pressure in the youths and interventions leading to weight reduction have succeeded in reducing blood pressure levels. These facts suggest that students with borderline hypertension can benefit substantially by weight control.

That salt intake induces hypertension is evidenced by both experimental studies in rats and epidemiological studies in humans. The relationship between salt intake and blood pressure in children and adolescents, however, remains unclear and may depend on whether there is a family history of hypertension. In our present study, 24-hour
urinary sodium excretion was found to be higher in the students of the BH group than in those of the N group. However, in both groups the students with a family history of hypertension did not have a higher 24-hour urinary sodium excretion as compared to the students without a family history. These findings may support the concept that salt acts as a pressor agent in all adolescents rather than in those with a genetic susceptibility predisposed to hypertension. On the other hand, the prevalence of hypertension has been considered to be related to not only a direct function of salt intake but also an inverse function of potassium intake, while our study could not provide strong support for the concept of "blood-pressure-potassium intake".

Our study indicated that plasma renin activity was significantly higher in the BH group than in the N group. Furthermore, 24-hour urinary aldosterone excretion was also considerably higher in the BH group. The explanation for these differences is not clear. In borderline hypertensive adults, some investigators have reported elevated values of plasma renin activity in the resting state, but only a few studies in this field have been performed on children and adolescents. Our result is in contrast to the result of Sinaiko et al. in which plasma renin activity was significantly lower in the upper 0.26% group than lower 5% group of the blood pressure distribution in approximately 10,000 grade school children. The renin-angiotensin-aldosterone system has been shown to be inversely dependent on sodium balance. Despite the fact that sodium excretion was significantly greater in the BH group, we also found elevated plasma renin activity and urinary aldosterone values in this group. Higher heart rates in the BH group could be explained to some extent by assuming that sympathetic stimulation of the heart is stronger in this group. The sympathetic nervous system is intimately involved in the modulation of renin secretion. Thus, it seems likely that the activation of sympathetic nervous system in borderline hypertensive students per se could have an influence on the stimulation of renin-angiotensin-aldosterone system.

The impairment of renal kallikrein, producing kinins which are potent renal vasodilators and natriuretic substances, has been shown in experimental and human hypertension. Although this abnormality may occur as a consequence of hypertension, the possibility that low kallikrein production may play a role in the etiology of hypertension remains. However, we could not find any significant difference in 24-hour urinary kallikrein excretion between the borderline hypertensive and the normotensive students. This is in agreement with the observation by Lawton et al. that abnormalities in urinary kallikrein were not found in mild hypertensives with normal renin as compared to age-matched normotensive subjects. Finally, it has not yet been proved that this decrease is directly involved in the pathogenesis of hypertension.

In an epidemiological study, it has been shown that there is a familial clustering of urinary kallikrein concentration. Since this concentration is affected not only by the amount of kallikrein excreted but also by the urine volume, it is necessary to investigate whether there is a familial clustering of urinary kallikrein excretion. Accordingly, we studied the relation of 24-hour urinary kallikrein excretion to the family history of hypertension. In each group, students who had a family history of hypertension had a significantly lower urinary kallikrein excretion than those without it. Furthermore, in borderline hypertensive students with a negative family history urinary kallikrein excretion correlated well with urinary excretion of aldosterone which is one of the factors in renal kallikrein release but there was no such correlation in the students with a positive family history. It is well known that the young offspring of normotensive parents are most unlikely to be hypertensive, whereas those of hypertensive parents are quite likely to be so. Consequently, our results may suggest that an abnormality in the relationship of aldosterone to kallikrein metabolism might be involved as one hereditary factor in the development of hypertension.

Guyton et al. have proposed that the kidneys function as the final common pathway for blood pressure regulation by their control of salt and water excretion. The renal kallikrein-kinin system appears to be involved in regulating not only electrolyte and water transport but renal circulatory homeostasis. Decreases in renal kallikrein would reduce renal plasma flow and glomerular filtration rate. In fact, Kramer et al. observed decreases in renal blood flow, glomerular filtration rate, urinary volume, sodium and potassium excretion on the administration of aprotinin, an inhibitor of kallikrein and other
proteases, to volume-expanded rats. In the present study involving borderline hypertensive students without a family history, we supported the concept that the decrease in urinary kallikrein excretion was associated with a decrease in glomerular filtration rate. However, in those students with familial hypertension, we failed to find such a correlation. This failure was apparently not attributable to the effects of a decrease in renal kallikrein, since no difference in glomerular filtration rate was evident between the two subgroups in borderline hypertension. It may be, therefore, suggested that the absence of the association between kallikrein excretion and the glomerular filtration rate may contribute to the development of genetic hypertension.

In summary, our results showed that it is indeed possible to demonstrate environmental and physiological differences in adolescents with different blood pressure levels. Furthermore, abnormalities in the relation of aldosterone to kallikrein metabolism and in that of kallikrein to renal function control appear to account for the increased blood pressure levels in borderline hypertensive adolescents with familial hypertension, and they may contribute to their propensity to develop hypertension.

REFERENCES


25. AYMAN D: Heredity in arteriolar (essential) hypertension: A clinical study of the blood pressure of 1,524 members of 277 families. Arch Intern Med 53: 792, 1934

26. CORONI-HUNTLEY J, HARRAN WR,


43. KANEKO Y, FUJISIMA S, TOCHIKUBO O, UNEDA S, FUJIKI Y, ODA H: Relation of urinary kallikrein excretion to family history of hypertension in high-school students. In Ninth Scientific Meeting of the International Society of Hypertension, Mexico City, Mexico, 1982
