Abstract—Renin and aldosterone secretion is often lower in blacks than in whites, characteristics that resemble a milder form of Liddle syndrome in which a mutation in the amiloride-sensitive epithelial sodium channel (ENaC) of the kidney results in enhanced resorption of sodium. In the present study, we looked for evidence that the intrinsic level of ENaC activity is indeed higher in blacks than in whites. In overnight urine samples collected from young people (249 white and 181 black subjects, mean age 13.4 years), the urinary aldosterone/potassium ratio, which is typically very low in Liddle syndrome, was lower in blacks than in whites: 0.421±0.024 (mean±SE) versus 0.582±0.016 nmol/mmol (P<0.0001). In addition, all but 1 of 5 molecular variants in ENaC were much more common in blacks than in whites. G442V in the β-subunit, present in 16% of the blacks and in only 1 white, was associated with parameters reflective of a greater Na retention and potentially a higher ENaC activity: a lower plasma aldosterone concentration (P=0.070), a lower urinary aldosterone excretion rate (P=0.052), a higher potassium excretion rate (P=0.048), and a lower urinary aldosterone/potassium ratio (P=0.027). In a second cohort consisting of 126 black and 161 white normotensive subjects and 232 black and 188 white hypertensive subjects, βG442V did not show a significant association with hypertension (P=0.089). On the other hand, a variant that was twice as common in whites, αT663A, was associated with being normotensive both in blacks (P=0.018) and in whites (P=0.034). Expression of either βG442V or αT663A in Xenopus oocytes did not result in a change in basal Na current, consistent with the variants being in linkage disequilibrium with alleles at active loci. In conclusion, several lines of evidence are presented to suggest that ENaC activity is higher in blacks than in whites, which could contribute to racial differences in Na retention and the risk for hypertension. (Hypertension. 1999;34:631-637.)

Key Words: sodium channels  ■  aldosterone  ■  potassium  ■  hypertension, sodium-dependent  ■  race

In today’s world, dietary intake of sodium is high, and although the kidney excretes most of this Na, some is retained through reabsorption. Blacks appear to absorb more Na than whites, as evidenced by a lower level of plasma renin activity (PRA)5–7 and greater salt sensitivity of blood pressure.5–7 We showed in a cohort of children that blacks also produce less aldosterone,8,9 consistent with a primary renal mechanism for increased Na retention that secondarily suppresses the renin-aldosterone axis. In addition, blacks have a greater predisposition to develop hypertension than whites,10,11 which could be related to the increased Na reabsorption. Why blacks more than whites appear to retain additional Na is unknown.

The amiloride-sensitive epithelial Na channel (ENaC) in the collecting duct of the kidney is the last site for Na reabsorption and one of the most important.12,13 Multiple factors affect its regulation, including aldosterone, which increases its activity.14,15 The channel consists of 3 partially homologous subunits: α, β, and γ.16 Mutations in either β- or γ-ENaC can result in Liddle syndrome, in which constitutive reabsorption of Na leads to hypertension that is often severe, hypokalemia, and suppression of renin and aldosterone secretion.17–22 Blacks show milder but similar features to those in Liddle syndrome, and thus there is interest in ENaC as a contributor to the increased Na reabsorption in blacks. Several molecular variants in ENaC have been described that occur more frequently in blacks than in whites23–25; I showed an association with hypertension.24 Recently, genetic linkage of β- and γ-ENaC to systolic blood pressure was described in white subjects.26

In the present study, we sought to identify associations between variations in ENaC subunits with parameters reflective of Na reabsorption using a study cohort of school-aged young people.27 We measured levels of renin activity and aldosterone, as well as potassium excretion and blood pressure, all of which could be affected by a more active ENaC.

Received April 4, 1999; first decision May 28, 1999; revision accepted June 10, 1999.

From the Department of Medicine, Indiana University School of Medicine (W.T.A., L.J.B., L.Z., J.F.R., M.A.W., C.G., J.H.P.), and the VA Medical Center (M.A.W., C.G., J.H.P.), Indianapolis, Ind; and the Department of Internal Medicine, University of Iowa College of Medicine (P.M.S.), Iowa City, Iowa.
Correspondence to J. Howard Pratt, MD, 541 Clinical Dr, Indianapolis, IN 46202-5111. E-mail johpratt@iupui.edu
© 1999 American Heart Association, Inc.
Hypertension is available at http://www.hypertensionaha.org
In a second cohort consisting of adult subjects, the association of molecular variants with the presence or absence of hypertension was studied. Subjects consisted of blacks and whites.

### Methods

#### Identification of ENaC Variants

DNA was isolated from white blood cells from groups of unrelated adults: normotensive whites, hypertensive whites, normotensive blacks, and hypertensive blacks. There were 96 subjects in each group. Sequences of \( \approx \) 200 to 300 nucleotides were amplified by polymerase chain reaction (PCR). Primers for the amplification reaction in most instances hybridized to a region of intron; their sequences are presented in Table 1. Variants were identified by single-strand conformational polymorphism (SSCP) analysis\(^2\) followed by dideoxy DNA sequence analysis with the Oncor Fidelity DNA Sequencing System. Once identified, the polymorphism was confirmed by allele-specific oligonucleotide hybridization.

#### Subjects

##### Normotensive Young People

The young subjects consisted of children and adolescents who participated in a longitudinal study of blood pressure regulation.\(^2\) None had hypertension, renal or cardiac disease, or diabetes mellitus. The study was approved by the Institutional Review Board of Indiana University–Purdue University of Indianapolis. In the case of minors, informed consent was also obtained from a parent or a legal guardian. Blood samples were obtained for measurement of PRA and the level of aldosterone. Urine samples were collected from bedtime to the next morning for measurement of electrolyte and aldosterone excretion rates, with results expressed per milligram of urinary creatinine. For most of the subjects, urine samples were collected multiple times at 6-month intervals.

##### Normotensives and Hypertensives

An adult cohort was studied that consisted of individuals with and without hypertension. Hypertensives were recruited from clinics at the VA Hospital and Indiana University Hospital in Indianapolis. Additional hypertensives, as well as normotensives, were recruited from local churches and through advertisements in local newspapers. The hypertensives were diagnosed before the age of 50 years. None had hypertension, renal or cardiac disease, or diabetes mellitus. The study was approved by the Institutional Review Board of Indiana University–Purdue University of Indianapolis. In the case of the young cohort, blood pressure was measured in the right arm with a random-zero sphygmomanometer (Hawksley and Sons) while subjects were seated, whereas a standard mercury sphygmomanometer was used in the adult cohort. The first and fifth Korotkoff sounds were designated systolic and diastolic blood pressures, respectively. Three blood pressure readings were obtained, and the average was used for the analysis. In the case of the young cohort, blood pressure was measured multiple times at intervals of 6 months.

#### Expression in Xenopus Oocytes

Two variants showed a significant association with either parameters of Na retention or blood pressure, and therefore, in vitro studies were performed to test for an influence of the molecular change itself on channel function. cDNAs encoding \( \alpha \), \( \beta \), and \( \gamma \)-ENaC in expression vector pMT3 were generated as previously described.\(^3\) Polymorphisms were introduced by site-directed mutagenesis (Mutagen [Bio-Rad] or Quick-change [Stratagene]), and the accuracy of the changes was confirmed by DNA sequencing. We coexpressed \( \alpha \), \( \beta \), and \( \gamma \)-ENaC in Xenopus oocytes by nuclear injection of cDNA (0.2 ng each); either wild-type ENaC or channels containing a polymorphism in the \( \alpha \) (T663A) or \( \beta \) (G442V) subunit were injected along with the 2 wild-type subunits. After incubation of the oocytes overnight in modified Barth’s solution, whole-cell amiloride-sensitive Na current was determined by 2-electrode voltage clamp at \(-60 \text{ mV}. \) Amiloride-sensitive current was the current blocked by a maximal concentration of amiloride (100 \( \mu \text{mol/L} \)); added to the bathing solution. During 2-electrode voltage-clamp recording, oocytes were bathed in a solution containing 116 mmol/L NaCl, 2 mmol/L KCl, 0.4 mmol/L CaCl\(_2\), 1 mmol/L MgCl\(_2\), and 5 mmol/L HEPES (pH 7.4 with NaOH). Because of day-to-day variability in the experiments, we normalized results of the experiments by dividing all measurements within each day by that day’s average current for the wild-type channel. This resulted in the wild-type being normalized to a mean of 1.

### Statistical Analysis

For normotensive young people, urinary excretion rates and blood pressure were measured in most instances on multiple occasions, and the number of measurements for each of the subjects was not the same (range 1 to 24 per subject). Demographic statistics for the
continuous variables were calculated by repeated-measures ANOVA, which accounted for the varying number of measurements between people and the within-person correlation. The compound symmetrical covariance model was used for the final analyses. Logarithmic transformations of the response were used for all analyses. Included as independent variables in the repeated-measures analysis were sex, age, and BMI, as well as genotype. Genotype was modeled as a linear term, which assumes that the differences in response between 0 and 1 copy of the allele is the same as between 1 and 2 copies. In the adult cohort, the 2-sample t test was used to compare BMI, age, and blood pressures. The effect of genotype on compare BMI, age, and blood pressures. The effect of genotype on the prevalence of hypertension was tested by Fisher’s exact test. For all comparisons of gender frequencies also used Fisher’s exact test. For both cohorts, an exact test for Hardy-Weinberg equilibrium was used to verify that the distributions of the variants were in equilibrium.32 The 2-sample t test was used to compare the in vitro Na current in wild-type and variant groups.

**Results**

**Normotensive Young People**

**Characteristics**

Table 2 depicts characteristics of the young subjects. Although similar in age to the whites, the blacks had a significantly higher mean BMI (P <0.0001). Mean systolic (P = 0.038) and mean diastolic (P = 0.0006) blood pressures were in each case 2 mm Hg higher in the blacks. The mean level of PRA was 19% lower in the blacks than in the whites (P = 0.0041), and the means of the plasma aldosterone concentration and the aldosterone excretion rate were, respectively, 37% and 34% lower in the blacks than the whites (P < 0.0001 for both values). Urinary excretion of K was 19% lower in the blacks than in the whites (P < 0.0001), whereas the serum K concentration was similar in blacks and whites. The findings were consistent with observations made in previous studies of this cohort.9,33 The mean aldosterone/K ratio was significantly lower in the blacks than the whites (0.421 ± 0.024 [mean ± SE] versus 0.582 ± 0.016 mmol/mmol; P < 0.0001) (Figure 1); this was true even though the excretion rate of K in the blacks was lower than in the whites.

**Allele Frequencies**

Nine nucleotide sequence variants were identified by SSCP. Allele frequencies ranged from 3% for βT594M to 45% for αA334T (Table 3). Three of the nucleotide substitutions in α-ENaC and 2 in β-ENaC resulted in an amino-acid substitution, whereas 1 in β-ENaC and all 3 in γ-ENaC were silent substitutions. All variants were in Hardy-Weinberg equilibrium with the exception of the β-ENaC C279T allele in the blacks, for which P = 0.018 (for C279T in whites, P = 0.37). Of the molecular variants, 4 occurred almost exclusively in the blacks (T334A and C168F in α-ENaC and G442V and T594M in β-ENaC), whereas 1 in α-ENaC (T663A) was about twice as common in the whites. To the best of our knowledge, only βG442V25 and βT594M23,24 were described previously. No variant was found to be in linkage disequilibrium with any of the other variants.

**Figure 1.** Urinary aldosterone/K ratio in whites and blacks expressed as mean ± SE (P <0.0001). Asterisks depict median values. Samples were collected overnight. Most individual values are the mean of multiple samples collected at intervals of 6 months. The urinary aldosterone ratio for patients with Liddle syndrome is based on results of Botero-Velez et al.34

---

**Table 2. Characteristics of Subjects in the Young Cohort**

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Whites</th>
<th>Blacks</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>M/F</td>
<td>129/120</td>
<td>82/99</td>
<td>0.20</td>
</tr>
<tr>
<td>Age, y</td>
<td>13.3 ± 1.5</td>
<td>13.4 ± 1.8</td>
<td>0.66</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>21.1 ± 4.5</td>
<td>22.9 ± 4.6</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Systolic BP, mm Hg</td>
<td>104.7 ± 7.8</td>
<td>106.4 ± 8.3</td>
<td>0.038</td>
</tr>
<tr>
<td>Diastolic BP, mm Hg</td>
<td>62.2 ± 6.1</td>
<td>64.4 ± 6.7</td>
<td>0.0006</td>
</tr>
<tr>
<td>PRA, ng·L⁻¹·g⁻¹</td>
<td>0.97 ± 0.60</td>
<td>0.79 ± 0.63</td>
<td>0.0041</td>
</tr>
<tr>
<td>Plasma aldosterone concentration, pmol/L</td>
<td>410.6 ± 214.7</td>
<td>257.1 ± 225.9</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Plasma K concentration, mmol/L</td>
<td>4.21 ± 0.41 (n = 160)</td>
<td>4.28 ± 0.42 (n = 76)</td>
<td>0.15</td>
</tr>
<tr>
<td>Urinary aldosterone excretion, nmol/nmol creatinine</td>
<td>1.85 ± 0.74</td>
<td>1.22 ± 0.86</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Urinary K excretion, mmol/nmol creatinine</td>
<td>3.63 ± 1.3</td>
<td>2.92 ± 1.47</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Urinary aldosterone/K ratio, nmol/mmol</td>
<td>0.58 ± 0.25</td>
<td>0.42 ± 0.28</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Results are reported as counts or as mean ± SD. BP indicates blood pressure.
TABLE 3. Variants Identified in α-, β-, and γ-ENaC

<table>
<thead>
<tr>
<th>Subunit</th>
<th>Exon</th>
<th>Position</th>
<th>Nucleotide</th>
<th>Amino Acid</th>
<th>Whites</th>
<th>Blacks</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>α</td>
<td>6</td>
<td>1082</td>
<td>G→A</td>
<td>A→T (334)</td>
<td>0.031</td>
<td>0.442</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>α</td>
<td>13</td>
<td>2069</td>
<td>A→G</td>
<td>T→A (663)</td>
<td>0.293</td>
<td>0.146</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>α</td>
<td>13</td>
<td>1935</td>
<td>G→T</td>
<td>C→F (618)</td>
<td>0.002</td>
<td>0.080</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>β</td>
<td>1</td>
<td>279</td>
<td>C→T</td>
<td>No change (93)</td>
<td>0.409</td>
<td>0.113</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>β</td>
<td>8</td>
<td>1325</td>
<td>G→T</td>
<td>G→V (442)</td>
<td>0.002</td>
<td>0.083</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>β</td>
<td>12</td>
<td>1781</td>
<td>C→T</td>
<td>T→M (594)</td>
<td>0.006</td>
<td>0.030</td>
<td>0.0059</td>
</tr>
<tr>
<td>γ</td>
<td>2</td>
<td>496</td>
<td>T→C</td>
<td>No change (165)</td>
<td>0.455</td>
<td>0.325</td>
<td>0.0002</td>
</tr>
<tr>
<td>γ</td>
<td>2</td>
<td>571</td>
<td>C→T</td>
<td>No change (190)</td>
<td>0.149</td>
<td>0.095</td>
<td>0.017</td>
</tr>
<tr>
<td>γ</td>
<td>11</td>
<td>1969</td>
<td>C→G</td>
<td>No change (648)</td>
<td>0.192</td>
<td>0.247</td>
<td>0.056</td>
</tr>
</tbody>
</table>

Relationships of the ENaC Variants to Parameters Reflective of ENaC Activity

Each variant was analyzed for its association with plasma aldosterone, PRA, and urinary aldosterone and K excretion. Only G442V in β-ENaC showed significant associations, and this was in the blacks, in whom the allele frequency was 0.083; there was only 1 carrier of the allele among the whites. Relationships to βG442V are depicted in Table 4 and Figure 2. The magnitude of the effect is described by the estimated slope of the regression line, which is equivalent to the average change between individuals with 1 or 2 copies of βG442V. In the presence of βG442V, the overnight aldosterone excretion rate was lower (P=0.052), K excretion was higher (P=0.048), and the urinary aldosterone/K ratio was lower (P=0.027) (Figure 2); in each case, the direction of the change was consistent with a higher level of intrinsic ENaC activity in the presence of βG442V. βG442V showed a marginal association with the plasma aldosterone concentration (P=0.070) but did not associate significantly with the level of PRA (P=0.20). Because excretion rates were integrated values and were often measured multiple times, they provided more stable values and were thus possibly more representative of ENaC activity than were values derived from single plasma samples. This could explain the stronger associations with the urinary excretion rates. All the other variants showed no significant association with any of the parameters, and none of the variants were significantly related to blood pressure.

Hypertensives and Normotensives

Characteristics of Subjects

Table 5 depicts the characteristics of normotensive and hypertensive subjects. Black hypertensives were on average ~9 years older than black normotensives, and white hypertensives had a mean BMI that was 1.6 kg/m² greater than white normotensives.

TABLE 4. Relationship of βG442V to Levels of PRA and Plasma Aldosterone, Urinary Aldosterone and K Excretion Rates, and Urinary Aldosterone/K Ratio in Black Subjects

<table>
<thead>
<tr>
<th>Independent Variable</th>
<th>Parameter Estimate</th>
<th>P</th>
<th>Parameter Estimate</th>
<th>P</th>
<th>Parameter Estimate</th>
<th>P</th>
<th>Parameter Estimate</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of copies of βG442V</td>
<td>−0.18</td>
<td>0.21</td>
<td>−0.25</td>
<td>0.07</td>
<td>−0.26</td>
<td>0.052</td>
<td>0.13</td>
<td>0.041</td>
</tr>
<tr>
<td>Female sex</td>
<td>0.08</td>
<td>0.49</td>
<td>0.11</td>
<td>0.34</td>
<td>0.39</td>
<td>0.0006</td>
<td>0.015</td>
<td>0.77</td>
</tr>
<tr>
<td>Age (y)</td>
<td>−0.01</td>
<td>0.66</td>
<td>0.056</td>
<td>0.048</td>
<td>−0.06</td>
<td>0.0004</td>
<td>−0.05</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>0.03</td>
<td>0.12</td>
<td>−0.001</td>
<td>0.94</td>
<td>0.012</td>
<td>0.26</td>
<td>−0.002</td>
<td>0.66</td>
</tr>
<tr>
<td>K excretion (mmol/mmol creatinine)</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>0.33</td>
<td>&lt;0.0001</td>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Na excretion (mmol/mmol creatinine)</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>−0.052</td>
<td>&lt;0.0001</td>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Intercept</td>
<td>0.28</td>
<td>4.48</td>
<td>−0.27</td>
<td>5.38</td>
<td>5.51</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Parameters are either regression slopes for continuous predictors or difference between groups for categorical predictors. For example, a 15-year-old black girl with a BMI of 22 kg/m² and 1 copy of the variant would have a predicted PRA (log value) of −0.06×15 (age)+0.08 (sex)+[−0.03×22] (BMI)+[−0.18×1] (genotype)+0.28 (y-intercept)=−0.63 (ng·L⁻¹·s⁻¹).
Relationships to the ENaC Variants

For none of the variants was the assumption of Hardy-Weinberg equilibrium rejected. \(\beta\)G442V, which showed an association with lower aldosterone and K excretion rates in the other cohort, was not a significant predictor of hypertension. Although the association was of marginal significance (\(P = 0.089\)), this resulted only from the fact that the 9 subjects homozygous for \(\beta\)G442V were hypertensive. There was no significant difference in allele frequencies for \(\beta\)G442V in normotensive (0.117) and hypertensive (0.120) subjects (\(P = 1.00\)). The \(\alpha\)T663A genotype, which was more common in whites than blacks, showed a significant association with being normotensive both in whites (\(P = 0.034\)) and in blacks (\(P = 0.018\)). The allele frequencies for \(\alpha\)T663A were 0.234 and 0.152 in black normotensives and hypertensives, respectively (\(P = 0.006\)), and 0.370 and 0.287 in white normotensives and hypertensives, respectively (\(P = 0.023\)). Thus, \(\alpha\)T663A appeared to be protective against hypertension. None of the other molecular variants showed a significant relationship to either hypertension or being normotensive. (See Table 6.)

Expression of \(\beta\)G442V and \(\alpha\)T663A in Xenopus Oocytes

We compared whole-cell amiloride-specific Na current in oocytes expressing the wild-type ENaC with that of the mutated ENaC. Neither \(\beta\)G442V nor \(\alpha\)T663A had an effect on the Na current. Our results with \(\beta\)G442V were similar to those of Persu et al.\textsuperscript{25} The average normalized current was 1.00±0.58 (mean±SD) for the \(\beta\)G442V wild type and 0.97±0.67 for the variant (\(P = 0.81\)), and for \(\alpha\)T663A, the average normalized current was 1.00±0.46 for the wild type and 1.13±0.92 for the variant (\(P = 0.52\)).

Discussion

In the present study, the urinary aldosterone/K ratio, which is low in patients with Liddle syndrome,\textsuperscript{34} was lower in the blacks, consistent with greater Na retention, as might occur with a more active ENaC. The molecular variant \(\beta\)G442V, which occurs almost exclusively in blacks, was significantly related to a lower aldosterone excretion rate and a lower urinary aldosterone/K ratio in a group of healthy young people. In black adults, however, \(\beta\)G442V did not show a

### Table 5. Characteristic of the Normotensive and Hypertensive Subjects

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Normotensive</th>
<th>Hypertensive</th>
<th>(P)</th>
<th>Normotensive</th>
<th>Hypertensive</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>M/F</td>
<td>38/88</td>
<td>140/92</td>
<td>0.019</td>
<td>62/99</td>
<td>103/85</td>
<td>0.003</td>
</tr>
<tr>
<td>Age, y</td>
<td>44.3±10.3</td>
<td>53.1±9.9</td>
<td>&lt;0.0001</td>
<td>49.9±9.0</td>
<td>51.4±11.4</td>
<td>0.20</td>
</tr>
<tr>
<td>BMI, kg/m(^2)</td>
<td>28.8±6.5</td>
<td>29.4±5.5</td>
<td>0.40</td>
<td>27.0±5.1</td>
<td>29.6±5.2</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Systolic BP, mm Hg</td>
<td>119.2±10.5</td>
<td>144.5±19.4</td>
<td>&lt;0.0001</td>
<td>120.4±13.1</td>
<td>146.5±18.9</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Diastolic BP, mm Hg</td>
<td>75.9±7.8</td>
<td>91.3±12.5</td>
<td>&lt;0.0001</td>
<td>76.9±9.3</td>
<td>93.3±11.2</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Results are reported as counts or as mean±SD. BP indicates blood pressure.
significant association with hypertension \( (P=0.089) \). A second molecular variant, \( \alpha T663A \), which was twice as common in whites, was associated with being normotensive in blacks and in whites, 2 independent population groups. \( \alpha T663A \) appeared to reduce the risk for hypertension. Thus, several lines of evidence suggest that intrinsic (non–aldosterone stimulated) ENaC activity may be higher in blacks than in whites.

Secretion of renin and aldosterone is often suppressed in blacks compared with whites,1–3,8,9 consistent with an ENaC that is functionally more active. In the present study, a reduced aldosterone excretion rate in the blacks was coupled to a K excretion rate that was greater than would have been predicted based on the normal relationship of K to aldosterone. K is a major stimulus of aldosterone secretion35; as dietary intake of K increases or decreases, as reflected in the urinary excretion of K, aldosterone secretion responds accordingly, and urinary excretion rates of aldosterone and K would be expected to change more or less in parallel. If, on the other hand, the channel functions at a higher level without additional aldosterone, then the ratio of excreted aldosterone to excreted K would be lower, aldosterone secretion being suppressed by the increase in reabsorbed Na and K excretion continuing relatively unabated in the collecting duct where K secretion is coupled to the reabsorbed Na. Also, aldosterone secretion would be less responsive to stimulation by K when the renin-angiotensin system was suppressed16,37; a lower amount of aldosterone would be secreted for any given level of intake of K. Indeed, a very low urinary aldosterone/K ratio has been used to identify patients with Liddle syndrome.33 A lower ratio would also occur with overproduction of another mineralocorticoid, although in a previous study, we found that, if anything, levels of other mineralocorticoids were lower in blacks.38 A lower urinary aldosterone/K ratio could result from a decrease in 11\( \beta \)-hydroxysteroid dehydrogenase, as occurs in apparent mineralocorticoid excess,9 or from modifications in the regulation of ENaC by accessory factors40 such as the ubiquitin ligase Nedd4, which participates in removal of ENaC from the cell surface.41

\( \beta G442V \) showed a significant association with aldosterone and K excretion rates in the cohort consisting of children and adolescents. Although none were hypertensive (some will presumably become hypertensive eventually), this group provided us the opportunity to study the relationship of genotype to parameters representative of Na balance and potentially to the origins of hypertension. These parameters could not have been measured as accurately in adults, in whom there may be confounding factors related to age, or in hypertensives, in whom there are confounding factors related to the hypertension itself and certainly to its treatment. This might explain why the association of \( \beta G442V \) with hypertension was not detectable. On the other hand, \( \alpha T663A \) was associated with being normotensive and showed no association with parameters of Na balance in the young cohort. In the case of \( \alpha T663A \), quite the opposite might occur, with age being important to its physiological expression. Our studies did not consider potential physical interactions between variants, such as between \( \alpha A334T \) and \( \beta G442V \), which are both quite common in blacks.

In our search of the coding regions, no polymorphism was found in the proline-rich sequence that is altered in Liddle syndrome.42 With the exception of \( \alpha T663A \), the common molecular variants were more prevalent in the blacks than in the whites (Figure 3), which raises the question of whether such modifications in the channel in blacks contribute to the efficiency of Na reabsorption and to the salt-sensitive hypertension that is common in blacks. In a study of blacks from London,24 although not in a study of US blacks,23 the \( \beta T594M \) variant was found to be associated with hypertension and a lower level of PRA. In the present study, no significant association of \( \beta T594M \) with hypertension was observed, nor were there associations with parameters representative of Na balance and potentially to the origins of hypertension. These parameters could not have been measured as accurately in adults, in whom there may be confounding factors related to age, or in hypertensives, in whom there are confounding factors related to the hypertension itself and certainly to its treatment.
In summary, parameters reflective of Na retention, together with significant associations of ENaC variants with either the state of Na balance or with hypertension, indicate that there may be increased ENaC activity in blacks. Different levels of intrinsic ENaC function may contribute to racial differences in Na reabsorption and the risk for hypertension.

Acknowledgments

This study was supported by NIH grants R01-HL-35795 and M01-RR00750, a Merit Review grant from the US Department of Veterans Affairs, and the American Heart Association, Indiana Affiliate (L.J.B.). Studies performed at the University of Iowa College of Medicine were supported by a fellowship from the Roy J. Carver Charitable Trust (P.M.S.) and by NIH grants HL-03575, HL-58812, and DK-52617. The authors are grateful for the excellent laboratory assistance of Priya Kulkarni.

References

Genetic Variants in the Epithelial Sodium Channel in Relation to Aldosterone and Potassium Excretion and Risk for Hypertension

Walter T. Ambrosius, Laura J. Bloem, Lifen Zhou, John F. Rebhun, Peter M. Snyder, Mary Anne Wagner, Chunlu Guo and J. Howard Pratt

Hypertension. 1999;34:631-637
doi: 10.1161/01.HYP.34.4.631

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1999 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/34/4/631

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Hypertension can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Hypertension is online at:
http://hyper.ahajournals.org//subscriptions/