A Gene-Environment Interaction Between Inferred Kallikrein Genotype and Potassium

Steven C. Hunt, Sandra J. Hasstedt, Lily L. Wu, Roger R. Williams

Urinary kallikrein excretion has been shown statistically to be partially determined by a major gene in large Utah pedigrees with the use of segregation analysis. A previous twin analysis of environmental factors influencing urinary kallikrein level showed that urinary potassium twin differences were strongly related to differences in urinary kallikrein. The present study uses 769 individuals in 58 Utah pedigrees to analyze the association of urinary potassium with urinary kallikrein within statistically inferred kallikrein genotypes. Fitting genotype-specific curves relating urinary kallikrein level to 12-hour urinary potassium amount within a major gene, polygene, and common environment model, we showed a significant statistical urinary potassium interaction with the inferred major gene for kallikrein (P=.0002). The heterozygotes (with a frequency of 50%) had a significant association between urinary kallikrein and potassium (slope, 0.51±0.04 SD), whereas there was no association with potassium in the low homozygotes, suggesting a genetic defect involving the kallikrein response to potassium. The model predicted that an increase in urinary potassium excretion of 0.8 SD above the mean in these pedigrees would be associated with high kallikrein levels in the heterozygotes similar to the high homozygotes. A decrease of 1.3 SD in urinary potassium excretion in heterozygous individuals was associated with kallikrein levels similar to the homozygous individuals with low kallikrein. Because in the steady state urinary potassium represents dietary potassium intake, this study suggests that an increase in dietary potassium intake in 50% of these pedigree members, estimated to be heterozygous at the kallikrein locus, would be associated with an increase in an underlying genetically determined low kallikrein level. If kallikrein excretion is found to be a physiological marker related to the risk of developing hypertension, that risk may be able to be modified by achievable increases in dietary potassium intake in heterozygous individuals deficient in potassium intake. (Hypertension 1993;22:161-168)

KEY WORDS • genetics • kallikrein • potassium • hypertension, genetic

LOW levels of urinary kallikrein excretion have been associated with hypertension and increased blood pressure in multiple studies. Segregation analysis on large Utah pedigrees provided evidence that a large proportion of urinary kallikrein level is determined by a major gene and that this major gene is associated with a positive family history of hypertension in adults and youths. Homozygosity for the more common allele at the responsible locus would theoretically make individuals susceptible to the onset of hypertension through low kallikrein levels. Although the major gene explained 51% of the total kallikrein variance, polygenes and random environment explained 27% and 22%, respectively. The 22% variance indicates that environmental factors may play a significant role in determining the level of kallikrein excretion. This is also supported by a large spouse-spouse kallikrein correlation of 0.40 found in another study.

In our previous analysis, statistical methodology was not in place for the unified model to incorporate common environmental influences that may mimic polygenes, confound the major locus means, or reduce the individual residual variation. There was also no implementation for modeling genotype-specific relations of associated variables within the framework of the unified model. With the subsequent availability of that statistical methodology, the specific aims of the present study were to model shared family and individual environmental factors and to investigate their relation to the major gene and polygenic effects previously found.

Dietary electrolytes, primarily sodium and potassium, have been studied extensively in relation to blood pressure levels and kallikrein excretion. In most studies, some people were responsive to either increased or reduced electrolyte amounts, whereas other people were resistant to electrolyte changes. We report here a significant statistical interaction between measurements of urinary potassium excretion and a major gene determining kallikrein levels, presumably representing a renal kallikrein genotype effect on the response of renal kallikrein levels to changes in dietary potassium intake. This interaction may partially explain why the blood pressure levels of some people respond and others do not respond to increased potassium intake in some of the intervention studies.

Methods

We studied the Utah pedigrees used in our earlier study. There were 73 pedigrees containing 1106 indi-
viduals. Individuals on antihypertensive medication, birth control pills, or hormone therapy or who reported any history of kidney disease were excluded. After individuals with missing data were excluded, the final sample was reduced to 769 individuals in 58 pedigrees. Pedigree sizes were 30 pedigrees with 1 to 4 members, 13 pedigrees with 5 to 9 members, 5 pedigrees with 10 to 29 members, 8 pedigrees with 30 to 55 members, and 2 pedigrees with 83 and 109 members. There were 328 descendants in 8 pedigrees ascertained for two or more deaths from stroke before age 75; 234 descendants in 8 pedigrees ascertained for two or more deaths from coronary heart disease before age 55; 162 relatives of 33 probands selected from the Salt Lake City component of the Hypertension, Detection and Follow-up Program; and 45 relatives of 9 probands referred to us through miscellaneous sources.

The mean number of households was 5.5 per pedigree (range, 1 to 39; median, 2). The mean number of visit dates per pedigree was 4.5 (range, 1 to 34; median, 1). Other details of these pedigrees are described elsewhere.10,12 This study followed procedures approved by an institutional review committee, and all subjects gave informed consent.

Sitting blood pressures were measured with an automated blood pressure device (Infrasonde SR-2, Sphygmometrics, Inc, Woodland Hills, Calif) after subjects had been seated for 5 minutes, and four such measurements taken at least 2 minutes apart during the same clinic visit were averaged. Blood pressure response to an isometric handgrip dynamometer after 2 minutes of gripping at 50% of the person's maximum grip was compared with the sitting pressures. Fifth phase blood pressures were used for the diastolic blood pressure. Mean blood pressures were used for the diastolic blood pressure. Urine samples were analyzed for urinary kallikrein excretion with the method of Green and Shaw13 as described by Ash et al.14 Urinary potassium and sodium were collected volume.

Urinary kallikrein, potassium, and sodium were converted to amounts by multiplying by the collected volume. Blood pressures, body mass index (weight/height²), and urinary kallikrein, potassium, and sodium amounts were adjusted using 5-year age- and gender-specific means and standard deviations for ages over 20 and 3-year age groupings for ages 20 and younger for each gender. All variables were standardized to a mean of zero and a standard deviation of 1. Power transformations were applied to urinary kallikrein and potassium before segregation analysis to remove skewness.15 The power transformation estimate for urinary kallikrein was −2.21, changing the skewness from 1.39 to 0.04. For urinary potassium, the transformation estimate was −1.68, changing the skewness from 1.19 to 0.04.

Genetic Analyses

Major locus inheritance of kallikrein levels was modeled using maximum likelihood segregation analysis.16,17 Maximum likelihoods were obtained using PAP and GEMINI.18,19 to which a variance components/major locus model has been added to estimate common environment effects independently from major locus and polygenic effects.20 Additional extensions to the model were made to allow kallikrein levels to be linearly or quadratically dependent within each genotype on levels of other correlated variables, such as urinary potassium level.21,22

The model used for this study assumed that the kallikrein phenotype resulted from a major genetic locus with a large effect, an additive polygenic effect, variance components estimates of a common household (chronic) environmental effect and an effect arising from attending clinic on the same day (which represents acute common environmental effects possibly arising from recent diet, laboratory, screener, seasonal, or other similar shared causes of variation), and random environmental effects, each acting independently. Furthermore, the separation between major gene distributions, usually modeled as the difference between estimated means of each distribution, was modeled using linear and quadratic equations containing slopes and intercepts to estimate the means. No ascertainment correction was used in this study.

Table 1 shows the parameter estimates from the general model and the submodels tested. Parameters include the gene frequency (q) for the allele for high (H) kallikrein levels, means or intercepts for each of the three genotypes (μLH, μUH, μHM), linear and quadratic terms relating potassium excretion levels to kallikrein levels for each genotype (βLH, βUH, βHM and γLH, γUH, γHM), a common standard deviation for each genotype (σ), polygenic heritability (h²), a shared household environmental effect (c₀), and a shared clinic visit effect within pedigrees (c₁). When linear or quadratic terms are included in the model, the μ values represent intercepts, and the combination of the intercept and linear and quadratic terms estimates the distribution means. Otherwise, the μ values represent the means of the distributions. The general model includes transmission parameters (τ₁, τ₂, τ₃) that are tested for departure from the expected Mendelian values of 1, 1/2, and 0, respectively. The transmission parameters are also tested for departure from the three τ values being equal to the gene frequency (environmental model).

The quadratic and linear coefficients were estimates of the relation of urinary potassium to kallikrein and affect phenotypic levels of kallikrein within genotype. After the nonsignificant quadratic effects were set to zero, the linear slopes were used to test for uniform or differing effects of urinary potassium on the kallikrein genotype means, representing environmental effects that are not genotype dependent (uniform) or gene-environment interactions (differing effects), respectively. A general mixed model allowing the τ values to float and including the two variance component effects (same current household and same clinic visit date) and genotype-specific regressions was used for the comparison of submodels.

Results

Mean age was 24.4±17.5 years, ranging from 3 to 83. There were 297 males and 472 females. The range of adjusted, and power-transformed urinary kallikrein amounts was −4.23 to 1.86 SD, and urinary potassium ranged from −3.40 to 2.28.

Table 1 shows the estimated parameters for the different genetic models fit to the data. Under the general model, the separation between mean urinary kallikrein levels in the high and low homozygote distributions was 2.2 SD (at the sample urinary potassium
Contrary to our previous analysis, the dominant model with a linear potassium effect in the high kallikrein allele group was not significant in the heterozygote group. However, the intercept for the heterozygote group with the large positive slope was forced much higher when compared with the large positive slope in the high homozygote group, indicating that the prevalence of the three genotypes appeared to be dominant for the rarer allele ($\chi^2=0.05 [1 df], P=0.82$ versus the dominant model).

In the “best” model that could be fit to the data, the slopes for the two homozygote groups and the polygenic heritability estimate were set at zero ($\chi^2=4.03 [3 df], P=0.26$). The test for a polygenic effect after the two homozygote slopes were fixed at zero was rejected ($\chi^2=0.50 [1 df], P=0.82$). Therefore, the intercept for the heterozygote group with the large positive slope was forced much higher when the slopes were set to zero. However, the intercept for the heterozygote group with the large positive slope was forced much higher when the slopes were set to zero. Therefore, the intercept for the heterozygote group with the large positive slope was forced much higher when the slopes were set to zero. However, the intercept for the heterozygote group with the large positive slope was forced much higher when the slopes were set to zero. However, the intercept for the heterozygote group with the large positive slope was forced much higher when the slopes were set to zero.

Hunt et al. Urinary Kallikrein-Potassium Interaction

| Table 1. Genetic Model Parameters and Estimates for Urinary Kallikrein Excretion and Its Association With Urinary Potassium Excretion |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Parameter       | General         | Codom-Quad      | Codom-Lin       | Codom-1 slope   | Codom-0 slope   | Best            |
| q                | 0.38±0.06       | 0.42            | 0.41            | 0.25            | 0.25            | 0.45±0.04       |
| $\mu_L$         | −0.77±0.09      | −0.77           | −0.78           | −0.57           | −0.66           | −0.83±0.07      |
| $\mu_H$         | 0.05±0.14       | 0.07            | 0.12            | 0.42            | 0.48            | 0.03±0.07       |
| $\mu_{IL}$      | 0.61±0.15       | 0.64            | 0.63            | 0.72            | 0.52            | 0.55±0.11       |
| $\beta_L$       | 0.09±0.06       | 0.10            | 0.11            | 0.26            | (0)             | (0)             |
| $\beta_H$       | 0.49±0.06       | 0.49            | 0.49            | (0)             | (0)             | (0)             |
| $\gamma_L$      | −0.03±0.04      | −0.03           | (0)             | (0)             | (0)             | −0.07           |
| $\gamma_H$      | 0.02±0.04       | 0.02            | (0)             | (0)             | (0)             | 0.07            |
| $\gamma_{IL}$   | 0.02±0.07       | 0.00            | (0)             | (0)             | (0)             | (0)             |
| $\sigma$        | 0.64±0.03       | 0.63            | 0.63            | 0.65            | 0.66            | 0.64±0.03       |
| $h^2$           | 0.07±0.15       | 0.09            | 0.10            | 0.15            | 0.15            | 0.37            |
| $c$             | 0.15±0.15       | 0.18            | 0.19            | 0.15            | 0.33            | 0.21±0.14       |
| $c_S$           | 0.23±0.16       | 0.20            | 0.19            | 0.20            | (0)             | 0.23±0.14       |
| $\tau_1$        | [1.0]           | (1.0)           | (1.0)           | (1.0)           | (1.0)           | =q              |
| $\tau_2$        | 0.40±0.06       | (0.5)           | (0.5)           | (0.5)           | (0.5)           | =q              |
| $\tau_3$        | [0.0]           | [0.0]           | [0.0]           | [0.0]           | [0.0]           | =q              |
| $-2 \ln(L)$     | 1794.354        | 1797.105        | 1797.736        | 1814.989        | 1891.000        | 1801.763        |
| df              | 15              | 14              | 11              | 9               | 7               | 8               | 14              |

$q$, Gene frequency; $\mu$, mean kallikrein levels of each genotype when no linear or quadratic potassium relations are modeled or intercepts when potassium is in the model; L, low kallikrein allele; H, high kallikrein allele; $\beta$, linear slopes of urinary kallikrein with urinary potassium within each genotype; $\gamma$, quadratic terms for potassium; $\sigma$, common standard deviation within each genotype; $h^2$, polygenic heritability; $c_S$, common shared household effects and same visit date effects within pedigrees; $\tau$, parent-to-offspring transmission probabilities. Codom-Quad model includes estimated quadratic terms for the relation of urinary potassium to urinary kallikrein within each genotype; Codom-Lin model only includes linear terms; Codom-1 slope model forces all three slopes to be identical for all three genotypes; Codom-0 slope model estimates mean kallikrein levels without regard to potassium levels. Standard errors of estimates are presented. Numbers in brackets are maximized at the boundary of the parameter; numbers in parentheses are fixed at that number. Best model represents best-fitting genetic model.
genotypes was 20.3% for the high homozygotes, 49.5% for the heterozygotes, and 30.2% for the low homozygotes (in the portion of the Utah population represented by these selected pedigrees). Because of the environmental interaction with potassium, the proportion of variance in kallikrein level explained by the major gene depends on the urinary potassium level. Table 2 shows the proportion of variance explained by each of the components modeled at the mean urinary potassium level and at one and two standard deviations above and below the sample mean using the estimates from the best-fitting genetic model. Different levels of potassium excretion influence the observed major gene variation by shifting the heterozygote group to high or low kallikrein excretion levels. The model predicts that in pedigrees with potassium excretion two standard deviations above the mean, the percentage of variance in kallikrein levels explained by the major gene is increased, with a reduction in variation due to unmeasured individual variation. The variance in kallikrein levels contributed by the common environmental components is also lowest at the high urinary potassium level.

The Figure shows the relations between urinary potassium excretion and kallikrein excretion using the model that includes quadratic and linear estimates for each of the three genotypes. The quadratic model is used instead of the best model to show how linear the curves are for all three genotypes over the range of ±2 SD of both variables. The model predicts that urinary kallikrein levels increase with increasing urinary potassium levels in the heterozygotes. However, kallikrein levels remain nearly constant in the low homozygote genotype, suggesting a defect involving the kallikrein response to potassium. Possibly because the kallikrein levels are already high in the high homozygotes, further increases in kallikrein with increasing urinary potassium are not seen.

Assuming a quadratic causal effect, reduction of urinary potassium excretion by three standard deviations below the sample mean would make the heterozygote distribution nearly the same as the low homozygote distribution (the curves never intersect). An increase in urinary potassium excretion by 1.2 SD above the sample mean would make the heterozygote distribution similar to the high homozygote distribution. If the curves are modeled using the best model, the intersections become −1.3 and 0.8 SD, respectively. Because 50% of the population represented by this selected sample of pedigrees are heterozygotes at some kallikrein locus, urinary potassium may have important implications for the urinary kallikrein excretion amount.

After urinary potassium was included in the genetic model, the gene frequency found in our earlier study of 0.15 increased to 0.45 for high levels. This is because a large proportion of the heterozygotes were classified as low homozygotes if their urinary potassium levels were low or were classified as high homozygotes if their urinary potassium levels were high.

Table 3 shows the characteristics of the three inferred genotypes when genotype probabilities are calculated using parameter estimates from the best-fitting genetic model. For each individual, the maximum of the three calculated probabilities was used to assign genotype. The mean value and standard deviation of all study

Table 2. Percentage of Urinary Kallikrein Variance Explained at Different Potassium Amounts Using the Best-Fitting Genetic Model

<table>
<thead>
<tr>
<th>Genetic model component</th>
<th>Urinary potassium (SD)*</th>
<th>-2</th>
<th>-1</th>
<th>0</th>
<th>+1</th>
<th>+2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Major gene</td>
<td></td>
<td>47</td>
<td>37</td>
<td>38</td>
<td>49</td>
<td>62</td>
</tr>
<tr>
<td>Polymorphisms</td>
<td></td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Common household</td>
<td></td>
<td>11</td>
<td>14</td>
<td>13</td>
<td>11</td>
<td>8</td>
</tr>
<tr>
<td>Same clinic date</td>
<td></td>
<td>12</td>
<td>14</td>
<td>14</td>
<td>12</td>
<td>9</td>
</tr>
<tr>
<td>Individual variation</td>
<td></td>
<td>30</td>
<td>35</td>
<td>35</td>
<td>28</td>
<td>21</td>
</tr>
<tr>
<td>Homozygote separation†</td>
<td></td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
</tr>
</tbody>
</table>

*Standard deviations above or below sample mean urinary potassium excretion amount.
†Standard deviations between means of the high and low urinary kallikrein homozygote distributions.

Plot shows relation of urinary potassium amounts to urinary kallikrein amounts for each of the three kallikrein genotypes using estimates from the genetic model with linear and quadratic terms for urinary potassium. Statistical interaction between urinary potassium and kallikrein genotype was significant at a value of \( P = .0002 \). Standard deviations (SD) on the axes are from the whole sample and are not the standard deviations within genotype. Percentages of each genotype refer to the population from which these pedigrees were sampled and do not necessarily represent the general Utah population.
The presence of an environmental interaction with genotype may take various forms, because environment could shift the distribution of one genotype more than the other two, shift the distribution in the opposite direction, or have no effect on one or two genotypes. An example of an environmental effect only in the heterozygotes, as seen in this study, has been recently reported. In a study of iron loading in Africa,\textsuperscript{23} individuals homozygous for the normal alleles for transferrin saturation and with increased dietary iron intake were protected from the effects of dietary iron intake from beer brewed in iron drums. Those who were heterozygous had levels of transferrin saturation similar to the normal homozygotes if they had low dietary iron intake but had significant increases in transferrin saturation up to the level of the abnormal homozygotes if they had increased dietary iron intake through drinking the beer brewed in iron drums. Thus, the genetic transmission at the iron-loading locus appeared to be recessive for high levels of transferrin saturation if dietary iron was not increased and dominant if dietary iron was increased.

Similarly, in populations in which dietary potassium is low, genetic transmission of the determinants of urinary kallikrein excretion will appear dominant for low levels, and in populations in which dietary potassium is high, genetic transmission will appear to be recessive for low kallikrein levels. If a kallikrein gene can be linked to blood pressure, our results imply that homozygotes for high kallikrein levels may be protected from environmental risk factors such as low potassium intake; homozygotes for low levels may develop hypertension, regardless of their potassium intake; and heterozygotes may have blood pressures that depend on the level of potassium intake. However, the association of blood pressure with kallikrein was weak in this study, and genetic linkage of kallikrein levels to hypertension in humans has not yet been done.

Other studies have shown that dietary potassium changes alter both urinary potassium and kallikrein excretion.\textsuperscript{24-27} The correlation between urinary potassium and kallikrein apparently comes completely from genetic transmission at the iron-loading locus appeared to be recessive for low kallikrein levels and in populations in which dietary potassium is high, genetic transmission will appear to be recessive for low kallikrein levels. If a kallikrein gene can be linked to blood pressure, our results imply that homozygotes for high kallikrein levels may be protected from environmental risk factors such as low potassium intake; homozygotes for low levels may develop hypertension, regardless of their potassium intake; and heterozygotes may have blood pressures that depend on the level of potassium intake. However, the association of blood pressure with kallikrein was weak in this study, and genetic linkage of kallikrein levels to hypertension in humans has not yet been done.

Other studies have shown that dietary potassium changes alter both urinary potassium and kallikrein excretion.\textsuperscript{24-27} The correlation between urinary potassium and kallikrein apparently comes completely from genetic transmission at the iron-loading locus appeared to be recessive for low kallikrein levels and in populations in which dietary potassium is high, genetic transmission will appear to be recessive for low kallikrein levels. If a kallikrein gene can be linked to blood pressure, our results imply that homozygotes for high kallikrein levels may be protected from environmental risk factors such as low potassium intake; homozygotes for low levels may develop hypertension, regardless of their potassium intake; and heterozygotes may have blood pressures that depend on the level of potassium intake. However, the association of blood pressure with kallikrein was weak in this study, and genetic linkage of kallikrein levels to hypertension in humans has not yet been done.

Other studies have shown that dietary potassium changes alter both urinary potassium and kallikrein excretion.\textsuperscript{24-27} The correlation between urinary potassium and kallikrein apparently comes completely from genetic transmission at the iron-loading locus appeared to be recessive for low kallikrein levels and in populations in which dietary potassium is high, genetic transmission will appear to be recessive for low kallikrein levels. If a kallikrein gene can be linked to blood pressure, our results imply that homozygotes for high kallikrein levels may be protected from environmental risk factors such as low potassium intake; homozygotes for low levels may develop hypertension, regardless of their potassium intake; and heterozygotes may have blood pressures that depend on the level of potassium intake. However, the association of blood pressure with kallikrein was weak in this study, and genetic linkage of kallikrein levels to hypertension in humans has not yet been done.

Other studies have shown that dietary potassium changes alter both urinary potassium and kallikrein excretion.\textsuperscript{24-27} The correlation between urinary potassium and kallikrein apparently comes completely from genetic transmission at the iron-loading locus appeared to be recessive for low kallikrein levels and in populations in which dietary potassium is high, genetic transmission will appear to be recessive for low kallikrein levels. If a kallikrein gene can be linked to blood pressure, our results imply that homozygotes for high kallikrein levels may be protected from environmental risk factors such as low potassium intake; homozygotes for low levels may develop hypertension, regardless of their potassium intake; and heterozygotes may have blood pressures that depend on the level of potassium intake. However, the association of blood pressure with kallikrein was weak in this study, and genetic linkage of kallikrein levels to hypertension in humans has not yet been done.
were ascertained in a manner for which it is difficult to correct the genetic estimates, the exact percentage of the general population that is heterozygous is not known. The correction should be small for the coronary disease-ascertained pedigrees, because the deceased probands were not included in the analysis, and the correction depends on the correlation between familial death from coronary heart disease and kallikrein levels in the probands. The correction may have more impact for the pedigrees ascertained for hypertension and stroke. However, given the weak correlation of urinary kallikrein with blood pressure and family history of these disease end points in these pedigree members, the correction is expected to be small.

In the absence of a marker for the genetic locus associated with kallikrein, one might be able to clinically distinguish the three kallikrein genotypes using kallikrein level and the kallikrein response to potassium supplementation. Those individuals with large changes in kallikrein after potassium supplementation would be heterozygotes, whereas those who did not change kallikrein level could be divided into the two homozygote genotypes by their low or high kallikrein levels. Peak urinary kallikrein levels are reached after about 1 week of fixed changes in dietary potassium and remain altered at least up to 1 month. Although the slopes describing the association between urinary kallikrein and potassium appeared to be linear over the range of ±2 SD, the numbers of observations at either end of the curves are small. As the potassium values approach the extremes, it is probable that kallikrein excretion is no longer linearly related to potassium. A direct test of a causal effect of dietary potassium on kallikrein levels within genotype and an assessment of the ability to classify a person to a particular genotype must await an intervention study.

The polygenic effect on kallikrein levels became nonsignificant in these analyses after common environment components were introduced into the model as opposed to the larger polygenic variation found for many other variables associated with blood pressure. As the environmental factors that affect kallikrein levels are further identified, the unexplained variance will be reduced and improve the power of linkage studies. Because the gene is so common, it should be considered as a susceptibility gene rather than a causal gene. If it is a susceptibility gene for hypertension, approximately 30% of the population from which these pedigrees were sampled may be at risk for hypertension because of their low kallikrein levels. An additional percentage of heterozygotes may be at increased risk if they have low urinary potassium excretion.

**Blood Pressure and Kallikrein**

Most cross-sectional studies have shown a relation between urinary kallikrein excretion and blood pressure or hypertension. We have previously shown that low kallikrein levels are associated with a positive family history of hypertension, stroke, and coronary heart disease. Also, the kallikrein genotype for low kallikrein levels is identified in youths as frequently as in adults. In this study, in which the average age was 24, mean sitting blood pressures were not different by genotype. However, people with the low kallikrein genotype appeared to be less responsive to an isometric stress test. Low renal kallikrein levels may cause increased local vasoconstriction. Because of a steady state of increased vasoconstriction, further constriction may be diminished below normal in these individuals. Some studies have shown increased blood pressure responses to various stress tests, so this finding needs further research to determine its significance.

The small sitting and stressed blood pressure differences among the three genotypes might be expected if the kallikrein gene is a susceptibility gene as proposed, requiring other factors superimposed on this defect for the development of hypertension. Also, many other factors control blood pressure, and their combination may obscure the actual relation of kallikrein with blood pressure. The lack of a strong trend in blood pressures in this young family cohort suggests that the contribution of the kallikrein genotype to the actual rise in blood pressure may take many years to appear. This would make it important to identify individuals who do not have the high, protective kallikrein genotype and would be susceptible to the effects of other risk factors for the development of hypertension.

Our blood pressure results do not disagree with most other published cross-sectional studies, because nearly all of those studies compared hypertensive patients with normotensive patients. In our study, all of the hypertensive patients were removed because of the confounding effects of medications. Even when highly significant results are found between normotensive and hypertensive patients, correlations with blood pressures within these groups are often weak or nonexistent. However, the association of kallikrein with blood pressure or hypertension is not yet clearly understood. In a recent study, the greatest kallikrein decreases were observed in malignant essential hypertension, with only moderate reductions of kallikrein in nonmalignant hypertension compared with normotensive patients. Holland et al did not find decreased kallikrein levels in hypertensive patients unless they had mild renal insufficiency, suggesting that altered kallikrein levels result from hypertensive renal injury. Thus, the susceptibility imposed by low kallikrein levels may interact in many different ways with other factors, resulting in different forms of or reasons for increased blood pressure.

For the present, one must therefore rely on the weight of evidence from most cross-sectional studies that urinary kallikrein is associated with hypertension. Ten-year follow-up of this family cohort is nearing completion and may provide evidence for a prospective relation of kallikrein to increases in blood pressure. Also, if the results found in rats are consistent with those in humans, the study by Pravenec et al provides further evidence of the involvement of kallikrein with blood pressure determination. They found cosegregation of blood pressure with a marker for the kallikrein gene family in the spontaneously hypertensive rat, with an increase in blood pressure of 10 to 15 mm Hg in the rats that inherited the spontaneously hypertensive rat allele. Markers at the structural kallikrein locus in humans have not yet been tested for linkage to kallikrein levels. If this locus or another locus were found to be linked to kallikrein level, genotypes of people in linked pedigrees would be more accurately determined,
improving the ability to test for blood pressure differences by genotype.

**Urinary Potassium and Kallikrein**

Dietary potassium is known to be a potent modulator of urinary kallikrein excretion. Horwitz et al. measured urinary kallikrein excretion at three different levels of dietary potassium intake. Kallikrein excretion increased with increasing dietary potassium in both normotensive and hypertensive subjects. Changes in urinary aldosterone paralleled the urinary kallikrein changes, suggesting that aldosterone may be closely involved with the kallikrein changes. In a double-blind, randomized, crossover study of dietary potassium supplementation for two periods of 4 weeks each, urinary kallikrein levels significantly increased by 52% as urinary potassium levels increased by 124%. In addition, systolic and diastolic blood pressures significantly fell by 6 and 4 mm Hg, respectively. The change in urinary potassium correlated with the change in urinary kallikrein (r = .50).

Using monozygous twins to identify environmental determinants of kallikrein excretion, we found that the most significant variable (out of a large number of anthropometric, biochemical, and dietary variables) that determined differences in kallikrein excretion between each twin pair was the difference in urinary potassium levels. Urinary sodium differences were not significantly related to kallikrein differences in the twins. This study also showed that kallikrein differences between twins correlated with systolic blood pressure differences between the twins.

In another study that compared mildly and more severely hypertensive patients with normotensive patients, the mildly hypertensive and normotensive patients had an increase in urinary kallikrein after a potassium load, but the more severely hypertensive patients had no increase in kallikrein levels. The more severely hypertensive patients also had the lowest kallikrein levels, implying that a greater proportion were homozygous for low kallikrein levels and did not respond to the potassium supplementation. The mildly hypertensive group might have had more heterozygotes who would have responded to the potassium load.

In summary, we have identified the significant environmental interaction between factors determining levels of urinary potassium and an unknown gene determining urinary kallikrein excretion. This interaction is primarily manifested as a kallikrein response to potassium levels in the heterozygote group. Although the evidence for a blood pressure association with the gene for kallikrein levels was equivocal, further studies with greater statistical power, including unmedicated hypertensive individuals and including measures of and modification of urinary potassium and aldosterone levels, are needed to better define the pathophysiology associated with abnormal kallikrein levels. Molecular identification of the gene associated with altered kallikrein levels would also be an important advance in our understanding of this system that could have important effects on blood pressure and the development of hypertension.

**References**


A gene-environment interaction between inferred kallikrein genotype and potassium.
S C Hunt, S J Hasstedt, L L Wu and R R Williams

_Hypertension_. 1993;22:161-168
doi: 10.1161/01.HYP.22.2.161

_Hypertension_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1993 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/22/2/161