A Genome-Wide Scan for Urinary Albumin Excretion in Hypertensive Families


Abstract—Albuminuria increases the risk of cardiovascular events in patients with essential hypertension and diabetic subjects. The heritability (h²) of albuminuria in multiplex hypertensive families is unknown. We calculated the familial aggregation of urine albumin:creatinine ratio (ACR) and performed a genome-wide scan to assess for loci contributing to ACR in participants enrolled in the Hypertension Genetic Epidemiology Network (HyperGEN). To perform the genome scan, we analyzed genotype results from 2589 individuals from 805 families in the Family Blood Pressure Program. ACR and covariates were available in 1727 individuals (mean age, 57.1 years). Estimates of h² were obtained by using variance component methodology as implemented in the SOLAR software package. Linkage was tested between 387 markers spanning the genome at an average interval of 9.32 cM, using SOLAR multipoint analysis. The h² of log urine ACR was 0.49 (P<1×10⁻⁷) after controlling for significant main and interactive effects of age, gender, body mass index, blood pressure, and use of ACE inhibitors or angiotensin-2 receptor blockers. The genome-wide scan revealed a maximum LOD score of 2.73 on chromosome 19 (robust corrected LOD, 2.40; P=0.0099) at marker D19S591 and a LOD score of 2.0 on chromosome 12 (robust corrected LOD, 1.75; P=0.005) at marker PAH. These analyses demonstrate the marked heritability of urine ACR in families enriched for the presence of members with essential hypertension. They suggest that a gene(s) associated with urinary ACR may be present on human chromosomes 19 and 12. (Hypertension. 2003;42:291-296.)

Key Words: albuminuria • nephrosclerosis • blacks • race • hypertension, essential

The heritability and role of inherited factors in the causation of elevated urinary albumin excretion (UAE) among hypertensive subjects remain unknown. Several reports reveal that albuminuria clusters tightly in the diabetic and nondiabetic members of multiplex families with type 2 diabetes mellitus. The presence of microalbuminuria in diabetic individuals portends an increased risk for development of progressive renal failure and subsequent end-stage renal disease (ESRD). In and of itself, ESRD has a strong familial component in diabetes and hypertension. Elevated levels of UAE in diabetic individuals are associated with increased rates of cardiovascular morbidity and mortality. In hypertensive subjects, microalbuminuria is a risk factor for premature cardiovascular morbidity and mortality. The most recent Joint National Commission on Hypertension (JNC VII) report includes microalbuminuria as evidence for the presence of target organ damage. Target organ damage indicates the need for more aggressive control of blood pressure.

It is likely that both genetic and environmental factors contribute to UAE in hypertensive individuals. The fawn-hooded rat, Munich Wistar F1 fromer rat, and Dahl salt-sensitive rat models of hypertensive nephropathy suggest that the genes regulating UAE are independent of those that regulate blood pressure. Previous results from HyperGEN revealed significant evidence for linkage of creatinine clearance to polymorphic markers on chromosomes 1, 3, and 6. These analyses did not assess the effect of albuminuria. Therefore, we performed an analysis of the heritability of urine albumin:creatinine (ACR) and a genome-wide scan for loci contributing to urine ACR to clarify the role of inherited factors on UAE in patients with essential hypertension.

Methods

Population
Participants in the Hypertension Genetic Epidemiology Network (HyperGEN), from the National Heart, Lung and Blood Institute

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* A list of HyperGEN participating institutions and principal staff is given in the Appendix.

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mean arterial pressure (MAP), age, gender, race, body mass index (BMI), medications (ACE inhibitor or angiotensin-2 receptor blocker), and MAP, gender, age, race, medications, age, race, medications, and gender. In addition, the analysis was repeated with and without correction for possible model misspecification, and empirical probability values are reported for the robust LOD scores.20

Results

Genotype data were available from 2589 individuals in 805 families who participated in the FBPP. Of these, 1727 individuals were recruited into HyperGEN and had measurement of urine ACR. The mean age (±SD) of these individuals was 57.1±10.9 years, they had been hypertensive for a mean of 17.12±11.59 years, and they had mean ACR of 64.25±360.90 mg/g (median ACR was 4.9 mg/g). Among these 1727 individuals, there were 1164 sibling pairs, 22 parent–offspring pairs, 61 avuncular pairs, 2 half-sibling pairs, 4 first cousins, 5 identical sibling pairs, and 37 unrelated pairs. The mean family size with ACR data were 2.03 members. The Table contains additional demographic information.

Demographic Characteristics of HyperGEN Study Population

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>1042 (60.34)</td>
</tr>
<tr>
<td>Male</td>
<td>685 (39.66)</td>
</tr>
<tr>
<td>Race</td>
<td></td>
</tr>
<tr>
<td>Black</td>
<td>834 (48.29)</td>
</tr>
<tr>
<td>White</td>
<td>893 (51.71)</td>
</tr>
<tr>
<td>Age, y</td>
<td>57.14±10.89</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>31.77±6.96</td>
</tr>
<tr>
<td>Mean arterial pressure, mm Hg</td>
<td>91.65±13.20</td>
</tr>
<tr>
<td>Use of medications affecting ACR</td>
<td>699 (40.42)</td>
</tr>
<tr>
<td>ACE inhibitor</td>
<td>672 (36.77)</td>
</tr>
<tr>
<td>Angiotensin receptor blocker</td>
<td>27 (3.76)</td>
</tr>
<tr>
<td>Duration of hypertension, y</td>
<td>17.12±11.59</td>
</tr>
<tr>
<td>Urine ACR mg/g</td>
<td>64.25±360.90</td>
</tr>
</tbody>
</table>

Data listed as mean±SD for continuous measures and n (%) for dichotomous measures. ACR indicates albumin: creatinine ratio.
The heritability ($h^2$) of log urine ACR was 0.49 ($P<1 \times 10^{-7}$) after controlling for significant main and interactive effects of age, gender, race, BMI, blood pressure, and use of ACE inhibitor (ACEi) and angiotensin receptor blocker (ARB) medications.

The genome-wide scan results are depicted in Figure 1. A maximum LOD score of 2.73 was observed on chromosome 19 at 9.0 cM (marker D19S591, $P=0.0004$). A lesser peak with a LOD score of 2.0 was observed on chromosome 12 at 112.0 cM (marker PAH, $P=0.002$). Ten thousand simulations were performed to determine the robust corrected LOD scores and corresponding empirical probability values for the peaks observed on chromosomes 19 and 12. For chromosome 19 at position 9 cM, the robust corrected LOD score was 2.40 (empirical $P=0.0009$) (Figure 2). For chromosome 12 at position 112 cM, the robust corrected LOD score was 1.75 (empirical $P=0.005$) (Figure 3).

**Discussion**

This report reveals that inherited factors appear to play a major role in the regulation of UAE in individuals with essential hypertension. The heritability of urine ACR remained highly significant in theses analyses after controlling for the effects of age, race, blood pressure, BMI, and the use of medications known to reduce proteinuria (ACEi and ARBs). Additionally, the genome scan provided suggestive evidence that genes regulating urine ACR are present on chromosomes 19 and 12.
The marked heritability of urine ACR appears consistent with the findings from two reports in multiplex type 2 diabetic families. A segregation analysis of urine ACR in 1269 white subjects from the Joslin Diabetes Clinic (630 type 2 diabetics and 639 nondiabetic relatives) revealed a significant correlation between median ACR in diabetic and nondiabetic members of the same family. A Mendelian model with evidence for a major gene was most strongly supported in all study subjects. Evidence for Mendelian inheritance was improved when only the diabetic subjects were evaluated, although a single major locus with multifactorial effects was more strongly supported. A segregation analysis of overt proteinuria in 2107 Pima Indians from 715 families revealed that the existence of a major gene effect with Mendelian inheritance as most likely. A dominant model provided the best fit. Taken together, these two reports suggest that urine ACR is regulated by a major gene in type 2 diabetic families.

There are potential limitations in the present analyses. It is now clear that microalbuminuria may be transient in individuals with type 1 diabetes mellitus. Less is known about the natural history of albuminuria in treated and untreated hypertensive patients. Additionally, controversy exists regarding the selection of appropriately sensitive assays for measuring albuminuria. The assay used in this study was extremely sensitive, having a lower limit of detection of 1.3 mg albumin per liter of urine. Although an assay might underestimate the...
true amount of albuminuria, this would tend to bias the results toward the null and probably would reduce the heritability estimates.

A genome scan for renal function (creatinine clearance) has previously been reported in members of the HyperGEN study.\textsuperscript{11,12} In these reports, the heritability of creatinine clearance was 0.17 and 0.18 among black and white subjects, respectively. The best evidence for linkage in black subjects was found on chromosome 3 (LOD = 3.61 at 214.6 cM) and in white subjects at chromosome 3 (LOD = 3.36 at 115.1 cM). In this genome scan for urine ACR, we did not identify any evidence for linkage in these regions on chromosome 3. The linkage peaks for urine ACR (chromosomes 19: LOD = 2.73 at 9.0 cM, robust corrected LOD 2.40, P = 0.0009; and chromosome 12: LOD = 2.00 at 112.0 cM, robust corrected LOD 1.75, P = 0.005) do not overlap with those that regulate renal function in these individuals.

The LDL receptor (LDLR) locus regulating atherosclerosis susceptibility is located on 19p13.3 to 13.2,\textsuperscript{22,23} within our broad region of linkage. Polymorphisms in the LDLR gene could conceivably result in altered urinary ACR. Recent reports reveal that elevated urinary ACR and excess cardiovascular morbidity and mortality rates are strongly associated.\textsuperscript{5,6} Type 2 diabetic individuals with microalbuminuria are at far greater risk for cardiovascular death than of progression to renal replacement therapy.\textsuperscript{4} The Heart Outcomes Prevention Evaluation (HOPE) study demonstrated the impact of microalbuminuria on cardiovascular event rates in nondiabetic individuals.\textsuperscript{6} Elevated urinary ACR can be reduced by intake of lipid-lowering drugs (particularly the LDL-lowering statin class).\textsuperscript{24} Reductions in serum lipids may also slow progression of renal disease.\textsuperscript{24} Therefore, elevated urinary albumin excretion could result from generalized endothelial dysfunction with concomitant large and small vessel atherosclerosis. It is more probable that another gene on chromosome 19 or 12 directly affects urinary protein excretion, since a previous report in HyperGEN families failed to demonstrate linkage between markers on chromosome 19 and serum LDL levels.\textsuperscript{25}

Perspectives
This is the first report analyzing the heritability of urine ACR in members of multiplex hypertensive families. Elevations in urinary ACR are well-recognized risk factors for the development of heart attack and stroke. The heritability of log urine ACR was high (0.49) after controlling for the main and interactive effects of age, gender, race, BMI, blood pressure, and medications known to alter UAE. Additionally, suggestive evidence for linkage to urine ACR was detected on chromosomes 19 and 12. These results suggest that the genes regulating susceptibility to albuminuria may reside in these chromosomal regions. The important role of inherited factors in the development of albuminuria suggests that familial clustering of urine ACR may contribute, in part, to the observed familial aggregation of cardiovascular disease. It is important that additional large, family-based analyses in hypertension and cardiovascular disease attempt to reproduce these results.

Appendix

HyperGEN Participating Institutions and Principal Staff
New York Center/University of Utah Field Center: Steven C. Hunt, Roger R. Williams (deceased), Hilary Coon, Paul N. Hopkins, Janet Hood, Lily Wu, Jan Skippin; University of Alabama at Birmingham Field Center: Albert Oberman, Cora E. Lewis, Michael T. Weaver, Phillip Johnson, Susan Walker, Christie Oden; Boston University/Framingham Field Center: R. Curtis Ellison, Richard H. Myers, Yuqing Zhang, Luc Djoussé, Jemma B. Wilk, Greta Lee Splansky; University of Minnesota Field Center: Donna Arnett, Aaron R. Folsom, Mike Miller, Jim Pankow, Gregory Feitl, Barb Lux; University of North Carolina Field Center: Gerardo Heiss, Barry I. Freedman, Kari North, Kathryn Rose, Amy Haire; Data Coordinating Center, Washington University: D.C. Rao, Michael A. Province, Ingrid B. Borecki, Avril Adelman, Derek Morgan, Karen Schwander, David Lehner, Aldi Kraja, Stephen Mandel; Central Biochemistry Laboratory, University of Minnesota: John H. Eckfeldt, Catherine Lieendecker-Foster, Ronald C. McGlennen, Greg Rynders, Michael Y. Tsai, Jean Buckx; Molecular Genetics Laboratory, University of Utah: Mark Leppert, Steven C. Hunt, Jean-Marc Lalouel, Robert Weiss; National Heart, Lung, and Blood Institute: Susan E. Old, Millicent Higgins (retired), Cashell Jaquish, Martha Lundberg, Marianna Gerschenson.

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References


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