HFE C282Y Homozygosity Is Associated With Lower Total and Low-Density Lipoprotein Cholesterol

The Hemochromatosis and Iron Overload Screening Study

Paul C. Adams, MD; James S. Pankow, PhD, MPH; James C. Barton, MD; Ron T. Acton, PhD; Cathie Leiendecker-Foster, MS; Gordon D. McLaren, MD; Mark Speechley, PhD; John H. Eckfeldt, MD, PhD

Background—Previous studies have suggested a positive association of coronary heart disease risk and both serum ferritin concentrations and C282Y heterozygosity. Relationships between serum lipids, C282Y homozygosity, and serum ferritin have not been well established.

Methods and Results—The Hemochromatosis and Iron Overload Screening study screened 101 168 participants in primary care from 5 field centers in the United States and Canada with serum ferritin, transferrin saturation, and HFE genotyping for C282Y and H63D mutations. Serum lipids were measured in a subset of 176 C282Y homozygotes (63 male, 113 female whites) without a prior diagnosis of, family history, or treatment for hemochromatosis and a matched sample of participants with normal transferrin saturation and serum ferritin without C282Y or H63D mutations (wild-type, 123 male, 189 female whites). The proportion of subjects who reported using prescription cholesterol-lowering medications was ∼3 times higher in HFE wild-type subjects than C282Y homozygotes among men (22% versus 7%; P = 0.02) and, in women, 2 times higher (16% versus 8%; P = 0.07). After excluding subjects taking cholesterol medications, C282Y homozygotes had significantly lower mean total and low-density lipoprotein cholesterol concentrations than wild-type subjects, with larger genotypic differences for low-density lipoprotein in men (−0.62 mmol/L; 95% CI, −0.93 to −0.33) than in women (−0.28 mmol/L; 95%, CI, −0.52 to −0.08).

Conclusions—Total mean serum cholesterol and low-density lipoprotein levels were lower in C282Y homozygotes than in HFE wild-type participants. Further studies are required to determine whether this is related to iron overload, HFE alleles, or other factors on C282Y-positive chromosome 6p haplotypes. (Circ Cardiovasc Genet. 2009;2:34-37.)

Key Words: hemochromatosis ■ iron overload ■ iron

Clinical Perspective see p 37

Methods

The authors had full access to and take full responsibility for the integrity of the data. We have read and agree to the manuscript as written. The multicenter, multiethnic, primary care–based HEIRS study performed initial screening after informed consent on 101 168 participants for hemochromatosis and iron overload using TS and SF measurements and HFE genotyping (C282Y and H63D alleles).
A total of 51 subjects fasting for at least 12 hours were enrolled in the study. LDL cholesterol was calculated in serum specimens using the modified Friedewald formula (total cholesterol - triglycerides/5) when triglycerides were less than 4.52 mmol/L. 

### Results

Age and field center adjusted means or percentages of iron, liver, and lipid measures by *HFE* genotype are shown in Table 1. The age range in the HEIRS study was 25 to 100 years, with a median age of 51 years. The use of prescription cholesterol-lowering medications was 22% in *HFE* wild-type men and 16% in *HFE* wild-type women. In *C282Y* homozygotes, these medications were used significantly less often in men (7%, *P*=0.003) and women (8%, *P*=0.05) than in control subjects. After excluding subjects taking cholesterol medications, *C282Y* homozygotes had significantly lower mean total and LDL cholesterol concentrations than wild-type subjects, with larger genotypic differences for LDL in men (−0.62 mmol/L; 95% CI, −0.93 to −0.33) than in women (−0.28 mmol/L; 95% CI, −0.52 to −0.08). There was a significant inverse relationship between LDL and SF (*r*=−0.12, *P*=0.02) and TS (*r*=−0.25, *P*<0.001). Among *C282Y* homozygotes, the correlation between TS and LDL was −0.16 (*P*=0.06; *n*=137). Among wild-type subjects, the correlation between TS and LDL was 0.01 (*P*=0.90; *n*=219). Indirect measures of iron status (SF and TS) were markedly higher in both male and female *C282Y* homozygotes, and there was a statistically significant interaction between ferritin and *HFE* mutation by gender (*P*<0.001; Table 1). Because of this strong association, it was difficult to separate the potential contributions of *HFE* genotype and iron measures to LDL cholesterol levels. LDL cholesterol levels of *C282Y* homozygotes and wild-type subjects were compared according to TS (Figure). In men, age and center-adjusted prevalence of

### Table 1. Adjusted* Means or Percentages of Iron, Liver, and Lipid Measures by *HFE* Genotype

<table>
<thead>
<tr>
<th>Measure</th>
<th>C282Y/C282Y</th>
<th>+/+</th>
<th>Difference or Ratio (95% CI)</th>
<th>C282Y/C282Y</th>
<th>+/+</th>
<th>Difference or Ratio (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transferrin saturation, %</td>
<td>73</td>
<td>32</td>
<td>41 (36 to 46)</td>
<td>64</td>
<td>26</td>
<td>38 (34 to 42)</td>
</tr>
<tr>
<td>Ferritin, µg/L</td>
<td>1036</td>
<td>135</td>
<td>901 (730 to 1072)</td>
<td>380</td>
<td>67</td>
<td>313 (235 to 392)</td>
</tr>
<tr>
<td>ALT, U/L</td>
<td>36</td>
<td>26</td>
<td>10 (3 to 16)</td>
<td>20</td>
<td>23</td>
<td>−3 (7 to 2)</td>
</tr>
<tr>
<td>AST, U/L</td>
<td>39</td>
<td>34</td>
<td>4 (0 to 9)</td>
<td>22</td>
<td>24</td>
<td>−2 (5 to 2)</td>
</tr>
<tr>
<td>GGT, U/L</td>
<td>37</td>
<td>35</td>
<td>2 (9 to 14)</td>
<td>26</td>
<td>24</td>
<td>2 (6 to 9)</td>
</tr>
<tr>
<td>History of liver disease, %</td>
<td>15</td>
<td>5</td>
<td>2.8 (1.1 to 7.3)</td>
<td>8</td>
<td>7</td>
<td>1.1 (0.49 to 2.7)</td>
</tr>
<tr>
<td>Lipid-lowering medication, %</td>
<td>7</td>
<td>22</td>
<td>0.30 (0.11 to 0.83)</td>
<td>8</td>
<td>16</td>
<td>0.51 (0.25 to 1.1)</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L‡</td>
<td>4.91</td>
<td>5.56</td>
<td>−0.65 (−1.01 to −0.28)</td>
<td>5.28</td>
<td>5.51</td>
<td>−0.23 (−0.49 to 0)</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L‡</td>
<td>2.79</td>
<td>3.41</td>
<td>−0.62 (−0.93 to −0.33)</td>
<td>3.00</td>
<td>3.28</td>
<td>−0.28 (−0.52 to −0.08)</td>
</tr>
<tr>
<td>HDL cholesterol, mmol/L‡</td>
<td>1.22</td>
<td>1.22</td>
<td>0 (0 to 0)</td>
<td>1.47</td>
<td>1.50</td>
<td>−0.03 (−0.13 to 0.05)</td>
</tr>
<tr>
<td>Triglycerides, mmol/L‡</td>
<td>2.15</td>
<td>2.23</td>
<td>−0.08 (−0.88 to 0.72)</td>
<td>1.75</td>
<td>1.69</td>
<td>0.06 (−0.21 to 0.31)</td>
</tr>
</tbody>
</table>

* Excluding subjects taking lipid-lowering medications.

†Test of homogeneity across genders in difference in mean or prevalence ratio.

‡Excluding subjects taking lipid-lowering medications.

*Adjusted for age and field center.
hypercholesterolemia was significantly lower in C282Y homozygotes than in wild-type participants (Table 2). This association remained after additional adjustment for history of liver disease, or aspartate aminotransferase, alanine aminotransferase, and y-glutamyl transferase, but was substantially attenuated when adjusted for TS and SF (Table 2). In women, the association between hypercholesterolemia and HFE genotype was not statistically significant in any of the regression models (Table 2). We performed age stratification (<55 and ≥55) and found the LDL effect was larger in younger subjects (0.44 mmol/L) than in older subjects (0.21 mmol/L). A formal test of genotype × age interaction was not significant.

Discussion

CHD has not been reported commonly in long-term studies of C282Y homozygotes, but despite its being the most common cause of death in most Western countries. In this study, we have determined that C282Y homozygotes have a significantly lower total and LDL cholesterol. The direction and magnitude of the effect are very similar to those in a previous study regarding the relationship of serum iron measures and CHD morbidity or aspartate aminotransferase, ALT, AST, and GGT

Discussion

CHD has not been reported commonly in long-term studies of C282Y homozygotes,[12,14] despite its being the most common cause of death in most Western countries. In this study, we have determined that C282Y homozygotes have a significantly lower total and LDL cholesterol. The direction and magnitude of the effect are very similar to those in a previous study in a subset of C282Y homozygotes in the Atherosclerosis and Risk in Communities study.[14]

A key question is whether the magnitude of the difference in total or LDL cholesterol reported in this study is clinically significant. A meta-analysis of clinical trials predicted that every 10% of cholesterol lowering reduces CHD mortality by 15% and total mortality by 11%,[18] In the HEIRS study, male and female C282Y homozygotes had total cholesterol levels that were 12% and 4% lower, respectively, than their wild-type counterparts. Several large population-based studies have not demonstrated any differences in CHD morbidity or mortality events across all HFE genotypes,[12,14] but it is unlikely that these studies were large enough to detect modest reductions in risk for C282Y homozygotes.

Differences in serum cholesterol concentrations could result from long-term liver damage due to iron overload in C282Y homozygotes. It seems unlikely to be related to liver disease with decreased cholesterol synthesis because most participants had relatively mild iron overload. Liver biopsies were infrequently performed in the HEIRS study and cirrhosis was a rare observation.[19] Among patients with chronic hepatitis C, total cholesterol levels were 0.31 to 0.41 mmol/L (12 to 16 mg/dL) lower among those with significant liver fibrosis than in those with no significant fibrosis.[20,21]

It is important to establish whether the observations in this study regarding the relationship of serum iron measures and cholesterol and LDL cholesterol values apply to patients without HFE mutations. The magnitude of the differences in cholesterol would have public health significance if they could be extrapolated to the general population. The effects of oral iron supplements on serum lipids have not been clearly established, and many multivitamin preparations that claim to lower cholesterol contain iron. Experimental iron overload in rats has been found to lower LDL and raise high-density lipoprotein.[22] Excess iron could affect cholesterol metabolism due to increased intracellular oxidative stress, membrane peroxidation, and altered activity of liver enzymes involved in cholesterol metabolism and lipoprotein formation.[23] Patients with hemochromatosis typically have low levels of hepcidin that increase ferroportin expression on macrophages, which decrease intracellular iron. Foam cells in the arterial wall are deteriorated macrophages and may have decreased iron and less oxidative damage.[23]

It is important to recognize that elevated SF may be associated with CHD because SF may be a marker of obesity, the metabolic syndrome, diabetes, and inflammation. In this study, the selection of controls with a normal SF reduced the chance of including wild-type participants with metabolic syndrome, and therefore the differences observed here may be more marked than in the general population.

An unresolved question arising from these studies is whether the decreased cholesterol in C282Y homozygotes is related to excess iron, or a genetic effect of the HFE gene itself or other genes in close proximity such as those in the HLA region which are in linkage disequilibrium with the HFE locus.[24] The HEIRS study design resulted in a subset of C282Y homozygotes with a broad range of SF levels and included nonexpressing patients with a normal SF. Further studies should include the study of serum lipids in C282Y homozygotes before and after phlebotomy therapy. In conclusion, we have confirmed the previous observation, that C282Y homozygotes have a lower total and LDL cholesterol than matched controls without HFE mutations. This may be a factor in the reportedly normal life expectancy that has been reported in patients in population studies and an elucidation of the underlying basis for these observations could be relevant to population health.

Acknowledgments

The authors acknowledge the assistance of all the HEIRS study investigators and its participants for making this study possible.

Sources of Funding

The HEIRS Study was initiated and funded by the National Heart, Lung, and Blood Institute in conjunction with the National Human

### Table 2. Association of HFE Genotype and Hypercholesterolemia

<table>
<thead>
<tr>
<th>Model</th>
<th>Covariates</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Prevalence Ratio*</td>
<td>95% CI</td>
</tr>
<tr>
<td>1</td>
<td>Age and center</td>
<td>0.39</td>
<td>0.20–0.75</td>
</tr>
<tr>
<td>2</td>
<td>Model 1 variables + history of liver disease</td>
<td>0.38</td>
<td>0.20–0.73</td>
</tr>
<tr>
<td>3</td>
<td>Model 1 variables + ALT, AST, and GGT</td>
<td>0.43</td>
<td>0.22–0.83</td>
</tr>
<tr>
<td>4</td>
<td>Model 1 variables + SF and TS</td>
<td>0.81</td>
<td>0.31–2.13</td>
</tr>
</tbody>
</table>

ALT indicates alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma glutamyl transferase. *Prevalence of hypercholesterolemia in C282Y homozygotes vs wild-type homozygotes.
Clinic, the study was supported by contracts N01-HC05185 (University of Minnesota); N01-HC05186, N01-CM-07003-74, and the Minority Community Clinical Oncology Program (Howard University); N01-HC05188 (University of Alabama at Birmingham); N01-C05189 (Kaiser Permanente Center for Health Research); N01-HC05190 (University of California, Irvine); N01-HC05191 (London Health Sciences Centre); and N01-C05192 (Wake Forest University). Additional support was provided by University of Alabama at Birmingham General Clinical Research Center grant M01-RR00032; Howard University General Clinical Research grant M01-RR10284; University of California, Irvine/University of California at San Diego/University of California at Irvine Satellite General Clinical Research Center grant M01-RR00827, sponsored by the National Center for Research Resources, National Institutes of Health; Howard University Research Scientist Award U1H-HL03679-05 from the National Heart, Lung, and Blood Institute and the Office of Research on Minority Health (to V.R.G.); and the Southern Orient Disorders Center (to J.C.B., R.T.A.).

Disclosures

None.

References


HFE C282Y Homozygosity Is Associated With Lower Total and Low-Density Lipoprotein Cholesterol: The Hemochromatosis and Iron Overload Screening Study
Paul C. Adams, James S. Pankow, James C. Barton, Ron T. Acton, Cathie Leiendecker-Foster, Gordon D. McLaren, Mark Speechley and John H. Eckfeldt

Circ Cardiovasc Genet. 2009;2:34-37; originally published online January 23, 2009; doi: 10.1161/CIRCGENETICS.108.813089
Circulation: Cardiovascular Genetics is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2009 American Heart Association, Inc. All rights reserved.
Print ISSN: 1942-325X. Online ISSN: 1942-3268

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circgenetics.ahajournals.org/content/2/1/34

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation: Cardiovascular Genetics 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 Circulation: Cardiovascular Genetics is online at:
http://circgenetics.ahajournals.org//subscriptions/