Common Hepatic Lipase Gene Promoter Variant Determines Clinical Response to Intensive Lipid-Lowering Treatment

Alberto Zambon, MD, PhD; Samir S. Deeb, PhD; B. Greg Brown, MD, PhD; John E. Hokanson, PhD; John D. Brunzell, MD

Background—The common −514 C→T polymorphism in the promoter region of the hepatic lipase (HL) gene affects HL activity. The C allele is associated with higher HL activity, more dense and atherogenic LDL, and lower HDL2 cholesterol. Intensive lipid-lowering therapy lowers HL activity, increases LDL and HDL buoyancy, and promotes coronary artery disease (CAD) regression. We tested the hypothesis that subjects with the CC genotype and a more atherogenic lipoprotein profile experience the greatest CAD regression from these favorable effects.

Methods and Results—Forty-nine middle-aged men with dyslipidemia and established CAD who were undergoing intensive lipid-lowering therapy were studied. Change in coronary stenosis was assessed by quantitative angiography, HL polymorphism by polymerase chain reaction amplification, HL activity by 14C-labeled substrate, and LDL buoyancy by density-gradient ultracentrifugation. The response to lipid-lowering therapy was significantly different among subjects with different HL promoter genotypes. Subjects with the CC genotype had the greatest decrease in HL activity (P<0.005 versus TC and TT by ANOVA) and the greatest improvement in LDL density (P<0.005) and HDL2-C (P<0.05) with therapy. These subjects had the greatest angiographic improvement, with 96% of them experiencing CAD regression, compared with 60% of TC and none of the TT patients (P<0.001).

Conclusions—In middle-aged men with established CAD and dyslipidemia, the HL gene −514 C→T polymorphism significantly predicts changes in coronary stenosis with lipid-lowering treatment that appear to involve an HL-associated effect on LDL metabolism. This study identifies a gene polymorphism that strongly influences the lipid and clinical response to lipid-lowering drugs. (Circulation. 2001;103:792-798.)

Key Words: coronary artery disease ■ liver ■ genes ■ lipoproteins ■ pharmacogenetics
HDL-C). In addition to lowering LDL-C levels, pharmacologically induced changes in HL activity increase LDL particle buoyancy, leading to CAD regression. Therefore, we hypothesize that CAD patients will respond differently to therapeutic intervention, depending on the HL gene promoter polymorphism.

The primary goals of this study were to test (1) whether in men with established CAD, intensive lipid-lowering therapy can normalize the adverse lipid profile in subjects with the CC genotype at position −514 of the HL gene promoter and (2) whether these changes lead to greater improvement in angiographically documented CAD, thus defining a common genetically determined responsiveness to drug-associated CAD regression. Data addressing these questions will have remarkable potential clinical relevance, given the high frequency of the HL gene polymorphism among white (20% to 25%) and particularly among black (∼45%) and Japanese (∼47%) populations.

Methods

Patients
The study included 49 dyslipidemic men with elevated apoB levels (≥125 mg/dL). The subjects had completed a clinical intervention trial, were ≤62 years old at entry, had CAD diagnosed by coronary angiography, and had DNA available to evaluate HL gene polymorphism. Patients were studied at baseline and after 2.5 years of therapeutic intervention, depending on the HL gene promoter polymorphism. Patients were studied at baseline and after 2.5 years of angiography, and had DNA available to evaluate HL gene polymorphism. Data addressing these questions will have remarkable potential clinical relevance, given the high frequency of the HL gene polymorphism among white (20% to 25%) and particularly among black (∼45%) and Japanese (∼47%) populations.

Blood Collection
Blood specimens were collected in 0.1% EDTA after a 12- to 14-hour fast for plasma lipid measurements and density-gradient ultracentrifugation. Blood was collected in iced lithium-heparin tubes for measurement of HL and lipoprotein lipase activity. Blood samples were immediately processed and stored at −70°C for HL, lipoprotein lipase, and LDL buoyancy evaluation.

Lipid and Lipoprotein Determinations
Plasma, LDL, HDL, and HDL2 cholesterol, triglycerides, and apolipoproteins apoA, AI, and AII were measured at the Northwest Lipid Research Laboratories as previously described. Density-gradient ultracentrifugation for apoB-containing lipoproteins separates lipoprotein particles by the rate of flotation (RF) of lipoproteins in a salt density gradient and is designed to optimize the resolution of apoB-containing lipoproteins into 38 fractions as previously described. LDL RF, a measure of LDL buoyancy, is calculated as the fraction number of the major peak of LDL divided by the total number of fractions collected.

Postheparin Plasma Lipase Activity
Total lipolytic activity was measured in plasma as previously described by use of glycerol tri[1-14C]oleate emulsified with lecithin. HL activity, in nanomoles of fatty acids released per minute per milliliter of plasma, is defined as the activity remaining in the postheparin sample after incubation with a specific monoclonal antibody (5D2) that selectively inhibits lipoprotein lipase.

DNA Analysis
Screening for the LIPC −514 C→T polymorphism was carried out by polymerase chain reaction amplification using the primer pair as previously described.

Coronary Angiography
Quantitative coronary angiography was performed, and angiograms were analyzed as previously described. In each subject, an estimate of percentage proximal disease severity was obtained by averaging the severity of the worst lesion found in each of the 9 standard proximal coronary segments. Disease changes were calculated as the difference between percentage proximal disease severity at baseline and after treatment.

Statistical Analyses
Data on and off treatment within the same group were analyzed by paired Student’s t test or the Wilcoxon signed rank test if not normally distributed. Analyses among groups with different HL gene polymorphism were performed by ANOVA and the pairwise multiple comparison procedures (Tukey test). Whenever the data were not normally distributed, the Kruskal-Wallis 1-way ANOVA on ranks was used, as well as the pairwise multiple comparison procedures (Dunn’s method). Relationships between quantitative variables were tested by multiple linear regression analysis. The assumption of Hardy-Weinberg equilibrium was tested in the study groups by means of gene counting and χ2 analysis. The significance level was set at P<0.05.

Results
Of the 49 patients studied, 25 had the CC, 20 the CT, and 4 the TT genotype at position −514 of the HL gene promoter. The observed genotypic frequency in this CAD population was consistent with Hardy-Weinberg equilibrium (χ2 = 0.018, P=0.99). Twenty-five subjects were randomly assigned to receive LC and NC. The numbers of patients treated with LC compared with NC in each HL genotype group were similar (13/12, 10/10, and 2/2 in the CC, TC, and TT groups, respectively). No significant differences were observed between the 2 treatment groups when the effect of different HL genotypes on changes in coronary stenosis, HL activity, LDL concentration and LDL buoyancy (RF), apoB and A-I levels, total HDL and HDL2 cholesterol, body weight, and systolic and diastolic blood pressure was evaluated. In addition, in a multivariate analysis, drug treatment, considered as an independent variable, did not significantly affect the association between HL gene polymorphism and changes in coronary stenosis. Data from the LC and NC groups were therefore pooled and analyzed together.

HL Gene Promoter Polymorphism, Lipids, Lipolytic Activity, and CAD at Baseline
Body weight and systolic and diastolic blood pressure were not different at baseline and did not change with treatment in all groups (data not shown). All patients in this population selected for high apoB and CAD had the atherogenic lipoprotein abnormalities to be expected in such patients. The nature of the abnormalities appeared to differ, however, depending on the HL promoter genotype. At baseline, those with the CC genotype had greater HL activity compared with the TT (283 versus 169 nmol · min−1 · mL−1, P<0.05, Table 1), lower HDL-C (0.05 versus 0.21 mmol/L, P<0.005) despite virtually identical HDL2-C levels across genotypes, and lower LDL buoyancy (0.245 versus 0.283, P<0.01) (Table 2). By contrast, those with the TT promoter variant had higher LDL-C (5.67 versus 4.64 mmol/L, P=0.05) and borderline higher apoB levels (178 versus 152 mg/dL, P=0.08). These high levels and the differences in LDL-C and apoB levels
between groups with different genotypes are not traits attributable to the genetic variation in HL activity.3-12,14-16 Baseline parameters were not significantly associated with changes in CAD with therapy. Only LDL buoyancy showed a trend toward an association with changes in CAD (r=0.27, P=0.06), as we previously observed.20

**Association Between the HL Gene Polymorphism and Changes in HL Activity, LDL Buoyancy, and HDL2 Cholesterol With Lipid-Lowering Therapy**

Lipid-lowering therapy significantly reduced plasma cholesterol, triglyceride, LDL-C, and apoB concentration to a similar extent in all 3 subgroups of patients (Table 2). The HL gene promoter polymorphism was not associated with changes in cholesterol, triglycerides, LDL-C, and apoB levels with treatment. HDL-C increased significantly with therapy in the CC and TC groups (by 35% and 22%, respectively, P<0.01). A 14% increase in HDL-C was seen in the TT group. With lipid-lowering therapy, HL activity decreased significantly in the CC patients (−18%, P<0.01, Table 1), who also had a significant increase in LDL buoyancy (by 12%, P<0.01, Table 2). HL activity and LDL buoyancy did not change significantly in the TT patients.

The HL promoter polymorphism was significantly associated with the degree of changes in HL activity (P<0.005; ANOVA) and LDL buoyancy (P<0.005; ANOVA) with intensive lipid-lowering therapy (Figure 1). Patients with the CC genotype, who at baseline had higher HL activity and smaller, denser LDL particles, had a greater decrease in HL activity (Figure 1A) as well as a greater increase in LDL buoyancy (Figure 1B). The TT patients, who at baseline had lower HL and more buoyant, larger LDL particles, had no change in HL and LDL buoyancy with therapy. Intermediate changes were observed in the TC group. In addition, pairwise comparisons of these data indicated that both the TC and TT groups are significantly different from the CC subjects for changes in HL activity (P<0.01 for both comparisons) and changes in LDL buoyancy (P<0.05 for both comparisons). Finally, changes in HL activity were not significantly associated with changes in LDL-C or apoB with therapy.

With intensive lipid-lowering therapy, absolute changes in HDL2-C were not significantly different among groups with

### TABLE 1. Plasma HL and LPL Activity

<table>
<thead>
<tr>
<th>HL Gene Promoter Polymorphism</th>
<th>CC (n=25)</th>
<th>TC (n=20)</th>
<th>TT (n=4)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>Treatment</td>
<td>Δ, %</td>
</tr>
<tr>
<td>HL, nmol·min⁻¹·mL⁻¹</td>
<td>283±60</td>
<td>231±52</td>
<td>−18†</td>
</tr>
<tr>
<td>LPL, nmol·min⁻¹·mL⁻¹</td>
<td>187±86</td>
<td>189±70</td>
<td>1</td>
</tr>
</tbody>
</table>

Values are expressed as mean±SD. LPL indicates lipoprotein lipase.

*P<0.01 vs baseline.
†P<0.05 vs baseline CC.
‡P<0.005 vs on treatment CC.

### TABLE 2. Clinical and Lipid Parameters

<table>
<thead>
<tr>
<th>Hepatic Lipase Gene Promoter Polymorphism</th>
<th>Baseline</th>
<th>Treatment</th>
<th>Δ, %</th>
<th>Baseline</th>
<th>Treatment</th>
<th>Δ, %</th>
<th>Baseline</th>
<th>Treatment</th>
<th>Δ, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cholesterol, mmol/L</td>
<td>6.63±1.0</td>
<td>4.81±1.1</td>
<td>−27†</td>
<td>6.81±1.3</td>
<td>4.92±1.1</td>
<td>−28†</td>
<td>7.60±1.4</td>
<td>5.31±0.4</td>
<td>−30*</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>2.43±1.3</td>
<td>1.97±1.0</td>
<td>−19‡</td>
<td>2.15±1.5</td>
<td>1.76±0.8</td>
<td>−18*</td>
<td>1.93±1.3</td>
<td>1.43±0.4</td>
<td>−27*</td>
</tr>
<tr>
<td>LDL-C</td>
<td>4.64±0.8</td>
<td>2.87±1.0</td>
<td>−38‡</td>
<td>4.84±1.4</td>
<td>3.03±1.0</td>
<td>−37†</td>
<td>5.67±1.4</td>
<td>3.42±0.6</td>
<td>−40*</td>
</tr>
<tr>
<td>HDL-C</td>
<td>0.88±0.1</td>
<td>1.19±0.3</td>
<td>35‡</td>
<td>0.96±0.1</td>
<td>1.16±0.2</td>
<td>22‡</td>
<td>1.09±0.2</td>
<td>1.24±0.3</td>
<td>14</td>
</tr>
<tr>
<td>HDL2-C</td>
<td>0.05±0.02</td>
<td>0.22±0.15</td>
<td>335‡</td>
<td>0.10±0.08</td>
<td>0.23±0.09</td>
<td>128‡</td>
<td>0.21±0.11†</td>
<td>0.31±0.24</td>
<td>49</td>
</tr>
<tr>
<td>HDL3-C</td>
<td>0.85±0.1</td>
<td>0.94±0.1</td>
<td>11†</td>
<td>0.85±0.10</td>
<td>0.93±0.08</td>
<td>9</td>
<td>0.89±0.15</td>
<td>0.92±0.09</td>
<td>3</td>
</tr>
<tr>
<td>ApoB, mg/dL</td>
<td>152±23</td>
<td>104±25</td>
<td>−31‡</td>
<td>157±33</td>
<td>108±26</td>
<td>−31‡</td>
<td>178±39</td>
<td>115±23</td>
<td>−35*</td>
</tr>
<tr>
<td>ApoA-I, mg/dL</td>
<td>126±12</td>
<td>141±19</td>
<td>12‡</td>
<td>129±15</td>
<td>143±20</td>
<td>11*</td>
<td>133±12</td>
<td>138±24</td>
<td>5</td>
</tr>
<tr>
<td>ApoA-II, mg/dL</td>
<td>29±6</td>
<td>28±3</td>
<td>−3</td>
<td>27±4</td>
<td>27±6</td>
<td>0</td>
<td>32±3</td>
<td>27±4</td>
<td>−15</td>
</tr>
<tr>
<td>LDL Rf</td>
<td>0.245±0.024</td>
<td>0.274±0.032</td>
<td>12‡</td>
<td>0.254±0.047</td>
<td>0.268±0.039</td>
<td>6 (P=0.08)</td>
<td>0.283±0.043</td>
<td>0.287±0.044</td>
<td>1.4</td>
</tr>
<tr>
<td>Coronary stenosis, %Smax</td>
<td>35.6±12.1</td>
<td>33.5±12.2</td>
<td>−2.1†</td>
<td>39.4±12.2</td>
<td>38.3±13.1</td>
<td>−1.1</td>
<td>31.1±3.3</td>
<td>35.1±6.1</td>
<td>4.0 (P=0.08)</td>
</tr>
</tbody>
</table>

Values are expressed as mean±SD. %Smax indicates mean percent diameter reduction of worst lesions in each of the 9 standard proximal coronary segments.

*P<0.05 and †P<0.01 vs baseline, same genotype.
‡P<0.01 vs baseline CC.
different genotypes. When changes in HDL\(_{2}\)-C were expressed as percent of baseline concentration, however, the carriers of the C allele had a significantly greater increase in HDL\(_{2}\)-C than subjects in the TT group (335\%, 128\%, and 49\% in CC, TC, and TT, respectively, Table 3 and Figure 1C, \(P<0.05\) by ANOVA).

ApoA-I levels increased significantly in CC and TC subjects. Changes in apoA-I among groups with different genotypes failed to reach statistical significance (\(P=0.79\) by ANOVA).

**HL Gene Promoter Polymorphism, Coronary Atherosclerosis, and Response to Treatment**

Lipid-lowering therapy resulted in a significant improvement of coronary stenosis in CC patients (\(\Delta\%S_{\text{prox}} = -2.1, P<0.01\)) and to a lesser extent in the TC group (Table 2), whereas progression of stenosis was observed in the TT group (\(\Delta\%S_{\text{prox}} = 4.0, P=0.08\)).

The HL gene promoter polymorphism was associated with significantly different degrees of coronary stenosis regression with lipid-lowering therapy (\(P=0.01\) by ANOVA). In the CC group, 96\% of patients (24 of 25) experienced no worsening or improvement in mean coronary stenosis severity, compared with 60\% (12 of 20) in the TC and 0\% (0 of 4) in the TT group (\(\chi^2 = 16.43; P<0.001\), Figure 2). Changes in coronary stenosis continued to be statistically different in the CC versus CT patients (\(P=0.01\)) after exclusion of the TT subjects. In addition, after adjustment for baseline HL activity levels, the HL gene promoter polymorphism continued to be significantly associated with changes in coronary stenosis (\(r=0.45, P<0.01\)).

The analysis for linear trends in the proportion of subjects who experience progression or regression in coronary stenosis demonstrated a highly statistically significant difference in progression/regression based on genotype (\(\chi^2 = 15.215, P<0.0001\)).

The association between HL gene polymorphism and CAD benefit remained significant (\(P<0.005\)) after adjustment for drug-induced changes in cholesterol, triglycerides, HDL-C, HDL\(_{2}\)-C, apoA-I, and apoB levels, with the latter being the only significant predictor of CAD outcome in addition to HL genotype (Table 3, model 2). Finally, when changes in HL activity (Table 3, model 3) and changes in LDL buoyancy (Table 3, model 4) were included in the model, the association between HL genotype and degree of CAD regression with treatment was no longer significant. This suggests that the association between HL gene polymorphism and CAD improvement with therapy is mediated by the effect of this polymorphism on HL activity and subsequently on LDL buoyancy.

**Discussion**

In men with established CAD and dyslipidemia, the HL gene \(-514\ C\rightarrow T\) polymorphism significantly predicts coronary stenosis regression during intensive lipid-lowering treatment. This association appears to be mediated by the modulating effect of this polymorphism on specific drug-induced changes in lipoprotein metabolism. Homozygous CC patients exhibited a greater decrease in HL activity and a greater increase in LDL buoyancy with lipid-lowering therapy than both homozygous and heterozygous carriers of the T allele. Thus, the HL gene promoter polymorphism is responsible for a differential lipoprotein and angiographic response to lipid-lowering drugs.

Data from the 21 patients in the placebo group of the original clinical trial strongly support the presence of a specific, modulating effect of the HL gene polymorphism on CAD response to therapy. Regardless of their HL promoter genotype, these patients showed a similar degree of disease progression (\(\Delta\%S_{\text{prox}}\): CC [\(n=12\), +2.45; CT [\(n=7\], +2.36; TT [\(n=2\], +2.42] and no significant changes in HL activity, LDL density, and lipoprotein variables (data not shown).

Patients in the present study were middle-aged men selected for having dyslipidemia and CAD as diagnosed by angiography. \(^{21}\) When initially evaluated, the severity of coronary artery stenosis was not significantly different among groups with different HL promoter genotypes, in agreement with similar previous observations.\(^{15,18}\) Their lipoprotein profiles showed interesting differences, however, suggesting that the presence of CAD in these patients may have been accounted for by the presence of different lipid risk factors. Specifically, higher HL activity, small, dense LDL particles, and lower HDL\(_{2}\)-C levels characterized the atherogenic po-
tential of the lipid profile in CC patients. Previous observations showed that high HL activity is indeed associated with smaller and denser LDL particles, as well as lower HDL₄-C, and large epidemiological studies have demonstrated that both low HDL-C (and HDL₂-C)²⁵ and the presence of small, dense LDL¹⁰,¹¹ are risk factors for CAD. Conversely, patients with the TT genotype had significantly higher LDL-C and apoB levels and developed CAD despite the presence of more buoyant, less atherogenic LDL and higher HDL₂-C levels. Indeed, both LDL-C and apoB levels are strong independent risk factors for CAD⁵–⁶ and are not associated with HL activity levels.⁷

Our group recently reported that cholesterol-lowering therapy with an HMG-CoA reductase inhibitor or nicotinic acid in association with a resin not only affects lipoprotein levels (particularly LDL-C) but also significantly decreases plasma HL activity and increases LDL buoyancy.²⁰ HL-mediated changes in LDL buoyancy strongly predicted CAD regression with therapy. Two concurrent and independent lipoprotein pathways accounting for drug-associated CAD regression were identified (Figure 3): the well-known one leading to changes in LDL-C and apoB levels and the new pathway of HL-mediated improvements in LDL buoyancy.²⁰ The present study was designed to investigate the genetic contribution to the HL-mediated pathway associated with CAD response to therapy. No significant association was seen between changes in LDL-C and apoB and changes in HL activity in this study. This observation, as well as previous data²⁰ showing that a decrease in both plasma LDL-C and apoB levels with colestipol was associated with increased HL activity (opposite of what was seen in the present study), suggests that these 2 pathways may be at least partly independent of each other. Conversely, the present experimental design does not allow testing of the possibility that lipoprotein changes associated

---

**Figure 1.** Change in HL activity (A), LDL buoyancy (B), and percent HDL₂-cholesterol (C) during intensive lipid-lowering therapy in patients with different HL gene promoter genotypes (CC, TC, and TT). Values are expressed as mean±SD (error bars). See Methods for details.

---
Figure 2. Change in coronary disease severity with intensive lipid-lowering therapy in patients with different HL gene promoter genotypes (CC, TC, and TT).

Figure 3. Differential effect of intensive lipid-lowering therapy on change in HL activity, LDL buoyancy, and HDL-C cholesterol levels in carriers of the CC and TT genotypes at the HL promoter −514 locus.

with drug-induced decrease in HL activity may also be beneficial if they were not associated with reduction in plasma concentration of atherogenic lipoprotein particles, and specific studies are needed to address this question.

Previous observations demonstrated that HL activity, LDL buoyancy, and HDL-C but not LDL-C or apoB levels are significantly associated with the −514 polymorphism of the HL gene promoter, making it an interesting candidate to study the contribution of genetic factors to individual susceptibility for CAD regression. Our data indeed demonstrated that this polymorphism had no significant impact on the lipoprotein pathway leading to changes in LDL-C and apoB levels (Figure 3). HL genotype strongly influenced the LDL buoyancy–mediated pathway, however, promoting CAD regression (Figure 3). Patients with the CC genotype, in addition to improving LDL-C and apoB concentrations, normalized their HDL-C levels and LDL buoyancy, which characterized the atherogenic potential of their lipid profile at baseline (ie, small, dense, atherogenic LDL and low HDL-C). The greater magnitude of the increase in LDL buoyancy and HDL-C (as percentage of baseline value) was accounted for by a greater decrease, with treatment, in HL activity among CC patients compared with both TC and TT subjects. A shift toward larger and more buoyant LDL particles reduces their atherogenic potential because of diminished susceptibility to oxidative modification in the subendothelial space, which triggers the sequence of inflammatory responses believed to be crucial for lipid accumulation and plaque destabilization in the atherosclerotic process. In addition, normalization of HDL-C levels is consistent with a more efficient reverse cholesterol transport, a key pathway to reduce CAD risk and progression.

This study was not designed to provide clinical cardiovascular event end points; therefore, we do not have a direct measurement of the effect of HL gene promoter polymorphism on clinical CAD outcomes. The implications of our results, however, are most likely to be clinically relevant because a direct association between anatomic changes in coronary stenosis and future clinical events has been demonstrated by angiographic trials.

In summary, the present study provides compelling evidence that in white, middle-aged men with established CAD, the HL gene −514 C→T polymorphism significantly predicts changes in coronary stenosis with lipid-lowering therapy. In addition, this study provides the pathophysiological mechanism to account for the effect of this genetic polymorphism on CAD response to treatment, highlighting how current routine lipid measurements may not enable physicians to distinguish between responders and nonresponders. Screening for these variants in the HL gene promoter region identifies CAD patients who will benefit most from lipid-lowering strategies, as well as subjects who appear to be resistant to HL-mediated CAD regression. In these resistant patients, a more aggressive LDL-C–targeted and overall risk-reducing approach might be warranted. The relevance of these findings is emphasized by the high frequency of this polymorphism, ranging from 20% to 47%, depending on the population studied. Therefore, screening for this genetic variant could become an important parameter influencing the choice of treatment strategies for cardiovascular risk reduction and their cost-effectiveness.

Acknowledgments
This study was supported by NIH grants HL-30086, HL-64322, and NIH RR-37. Dr Zambon is supported by a Paul Beeson Physician Faculty Scholarship from the American Federation for Aging Research.

References
6. Sacks FM, Moyé LA, Davis BR, et al. Relationship between plasma LDL concentrations during treatment with pravastatin and recurrent coronary


Common Hepatic Lipase Gene Promoter Variant Determines Clinical Response to Intensive Lipid-Lowering Treatment
Alberto Zambon, Samir S. Deeb, B. Greg Brown, John E. Hokanson and John D. Brunzell

Circulation. 2001;103:792-798
doi: 10.1161/01.CIR.103.6.792

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/103/6/792