I
n children with familial hypercholesterolemia (FH), functional and morphological changes of the arterial wall become apparent at an early age, indicating that the atherosclerotic process begins shortly after birth. These findings have led to the hypothesis that effective low-density lipoprotein cholesterol (LDL-C)–lowering therapy, usually statin treatment, should be initiated in childhood, generally from the age of 10 to 12 years, to reduce the incidence of cardiovascular disease in later years. Carotid intima-media thickness (cIMT), as assessed by B-mode ultrasound, is considered a valid surrogate marker of cardiovascular disease. Previously, it was shown that age, sex, and LDL-C were strong and independent predictors of cIMT. Multivariable analyses showed that age, male sex, and presence of FH were independent predictors of cIMT.

Methods and Results: B-mode ultrasound measurements of the carotid artery were made in 196 children with FH and 64 of their siblings. Mean (±SE) cIMT in children with FH was significantly greater than that of unaffected siblings (0.398±0.052 versus 0.377±0.045 mm; P<0.001). A significantly greater cIMT value was observed before the age of 8 years. Multivariable analyses showed that age, male sex, and presence of FH were independent predictors of cIMT.

Conclusions: The difference in mean cIMT between children with FH and their unaffected siblings may be significant as early as age 8 years. This study confirms the need for early cholesterol lowering in this high-risk population. These patients participating in a carefully monitored study will help assess the long-term efficacy on cIMT and safety of statin therapy in young children. (Circ Res. 2014;114:307-310.)

Key Words: carotid intima-media thickness ■ hyperlipoproteinemia type II ■ pediatrics

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Rationale: Familial hypercholesterolemia (FH) predisposes patients to premature cardiovascular disease, with the process of atherosclerosis initiated in early childhood.

Objective: As part of an ongoing trial to assess the efficacy and safety of rosuvastatin in children with FH aged 6 to 17 years, we report the differences in carotid intima-media thickness (cIMT) at baseline between children with FH and their unaffected siblings.

Methods

Between January 2010 and January 2011, children aged 6 to <18 years were recruited from 14 lipid clinics in the Netherlands, Belgium, Norway, Canada, and the United States. Children were eligible if they had heterozygous FH and fasting LDL-C >4.92 mmol/L or LDL-C >4.10 mmol/L if there was a family history of premature cardiovascular disease in a first- or second-degree relative. FH was defined by a documented genetic defect or documented evidence of FH in a first-degree relative (LDL-C >4.9 mmol/L in an adult; LDL-C >4.1 mmol/L in a child <18 years of age). Siblings were eligible if they had a documented absence of genetic defect or a documented LDL-C of <3.00 mmol/L, without lipid-lowering medication. The protocol was reviewed and approved by each participating site’s institutional review board, and written informed consent was obtained from participants of age 12 years and all parents. The trial was registered with ClinicalTrials.gov as NCT01078675.

In all FH subjects, a full physical examination was done and venous blood samples were taken. Plasma lipid concentrations were
measured with CDC standardized assays. Ultrasound measurements on all participants were made using the Acuson Sequoia 512 instrument (Siemens AG, Malvern, PA; Erlangen, Germany) equipped with an 85-Mhz linear array transducer. All sonographers were trained and certified before their participation in the study. B-mode scans of the right and left common carotid artery, carotid bulbs, and internal carotid artery were obtained according to strict protocol specifications. Image readings were randomly assigned to image analysts for qualitative and quantitative evaluation. Image analysts were blinded to subjects. Mean cIMT was defined as the mean IMT of the right and left common carotid arteries, the carotid bulb, and the internal carotid far wall segments. For subjects in whom the scan of 1 of the segments had failed, mean IMT was calculated as the mean of the other 2 segments.

We assessed differences in demographic and cIMT between FH subjects and siblings by logistic or linear regression analysis with generalized estimating equations in the SAS procedure GENMOD to account for correlations within families. The same procedure was used to explore the association univariately between mean cIMT (response variable) and demographic and clinical characteristics (explanatory variables). Multivariable analysis was used to identify independent predictors after stepwise backward selection. An equation for difference in cIMT (ΔIMT) was derived by subtracting the equation for children with FH (if GROUP=1), that is, IMTFH=β1AGE+β2GROUP+β3AGE×GROUP+β4+β5AGE, from the equation for their siblings (if GROUP=0), that is, IMTsin=β1AGE, as described previously.2 This calculation resulted in ΔIMT=β2+β3AGE. β’s and SE were derived from the output of a linear regression analysis.

### Table 1. Demographic and Laboratory Data of Children With Familial Hypercholesterolemia, and Age, Sex, and Race of Unaffected Siblings

<table>
<thead>
<tr>
<th></th>
<th>FH (n=196)</th>
<th>Non-FH (n=64)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>12.1±3.3</td>
<td>11.9±3.5</td>
</tr>
<tr>
<td>Male sex, n (%)</td>
<td>88 (44)</td>
<td>33 (52)</td>
</tr>
<tr>
<td>White, n (%)</td>
<td>176 (90)</td>
<td>53 (85)</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>19.3±4.4</td>
<td></td>
</tr>
<tr>
<td>Blood pressure, mm Hg</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Systolic</td>
<td>107.5±10.9</td>
<td></td>
</tr>
<tr>
<td>Diastolic</td>
<td>64.0±8.1</td>
<td></td>
</tr>
<tr>
<td>Lipids, mmol/L</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC</td>
<td>7.87±1.34</td>
<td></td>
</tr>
<tr>
<td>LDL-C</td>
<td>6.10±1.26</td>
<td></td>
</tr>
<tr>
<td>HDL-C</td>
<td>0.90 (0.67–1.25)</td>
<td></td>
</tr>
<tr>
<td>TG*</td>
<td>1.30±0.33</td>
<td></td>
</tr>
</tbody>
</table>

Values are given as mean (SD) or indicated otherwise. BMI indicates body mass index; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; TC, total cholesterol; and TG, triglycerides.

### Table 2. Determinants of Carotid Intima-Media Thickness in Children With Familial Hypercholesterolemia at Baseline

<table>
<thead>
<tr>
<th></th>
<th>Univariate Analysis</th>
<th>Multivariable Analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Regression Coefficient (SE)</td>
<td>P Value</td>
</tr>
<tr>
<td>Age, y</td>
<td>0.004 (0.001)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Male sex</td>
<td>0.21 (0.006)</td>
<td>0.001</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>0.002 (0.001)</td>
<td>0.021</td>
</tr>
<tr>
<td>MAP, mm Hg*</td>
<td>0.001 (&lt;0.001)</td>
<td>0.064</td>
</tr>
<tr>
<td>LDL-C, mg/dL</td>
<td>&lt;0.001 (&lt;0.001)</td>
<td>0.787</td>
</tr>
<tr>
<td>HDL-C, mg/dL</td>
<td>&lt;0.001 (&lt;0.001)</td>
<td>0.197</td>
</tr>
<tr>
<td>TG, mg/dL†</td>
<td>0.002 (0.008)</td>
<td>0.841</td>
</tr>
<tr>
<td>Previous statin use</td>
<td>0.19 (0.011)</td>
<td>0.100</td>
</tr>
</tbody>
</table>

BMI indicates body mass index; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; MAP, mean arterial pressure; and TG, triglycerides.

*Mean arterial pressure=(Systolic blood pressure+(2×Diastolic blood pressure))/3.
†Log-transformed.

Results

In total, 196 children with FH were enrolled. From 53 families of these children, 64 unaffected siblings were included as controls. Children with FH and their siblings were comparable with respect to age (mean age [SD]: 12.1±3.3 versus 11.9±3.5 years, respectively; P=0.32) and sex (male sex: 44% versus 52%, respectively; P=0.39; Table 1). Mean cIMT (±SE) was 0.398±0.052 mm in children with FH and 0.377±0.045 mm in healthy siblings, which remained significant after adjustment for age, sex, and family relations (P<0.001). Associations between baseline variables of FH patients and cIMT are shown in Table 2. After stepwise backward elimination, multivariable regression analysis identified age and sex as independent predictors for cIMT. When siblings were also included in the multivariable model, age (regression coefficient [SE], 0.004 [0.001]; P<0.001), sex (regression coefficient, 0.022 [0.006] for males; P<0.001), and FH status (regression coefficient, 0.022 [0.007] for FH; P=0.002) revealed independent predictors for cIMT.

In the Figure, the difference in cIMT between children with FH and their siblings was plotted against age. A significant difference in cIMT between FH and controls was observed before the age of 8 years. In fact, FH patients showed an increase of 0.0041 mm/year compared with an increase of 0.0032 mm/year in siblings.

Discussion

In the present study, we show that children with FH have greater mean cIMT values as compared with their unaffected siblings before the age of 8 years, and we also report that age, sex, and presence of FH were independent predictors of...
carotid arterial wall atherosclerosis. These results reaffirm the findings of our previous study,2 but extend them to the age of 8 years as compared with 12 years in our previous study.

In our previous study, we found that the difference in cIMT increased with age between FH patients and siblings. We, therefore, expected a similar increase in the present study. However, our current data were not definitive. Interestingly, although the progression with age is almost similar in FH patients between the previous and the present study (0.005 versus 0.0041 mm/year), the mean progression with age in the siblings was <0.001 mm/year in the previous study versus 0.0032 mm/year in the present study (all adjusted for age and family relations). Because all non-FH controls were siblings of FH patients, genetic and environmental variation between the 2 groups was kept to minimum. Generally speaking, apart from FH, no major differences in risk factors for cardiovascular disease would be expected between the 2 groups. However, because the first study was performed >10 years before the present study, one possible explanation could be the rise in childhood obesity during the past decade. This might manifest more in siblings, because children with FH receive lifestyle advice on a frequent basis. If children in the present cohort were more obese, with all its metabolic sequelae, this might explain the faster progression in these siblings. However, to delineate these data in siblings further, more imaging studies including such healthy siblings as controls are needed.

Furthermore, despite the appearance of significant differences in the mean IMT results between FH patients and unaffected siblings, individual children cannot be distinguished on the basis of their cIMT results because of the extensive overlap in the data.

In our present study, in children aged 6 to 18 years, the difference in cIMT between FH subjects and unaffected siblings may be significant as early as age 8. This finding again underscores the importance of early LDL-C-lowering therapy,6,7 usually with statins, from younger age than recommended at present, because treatment should preferably be started before atherosclerosis is detectable. Clinical trials such as the present rosvastatin long-term cIMT trial should focus on treatment of these younger children to explore the efficacy and safety of statin treatment. At the same time, extensive follow-up studies are needed to establish the long-term efficacy, safety, and tolerability of statin therapy initiated in childhood and to further address the question of the appropriate age to start statin therapy.

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References
3. McCrindle BW, Urbina EM, Dennison BA, Jacobson MS, Steinberger J, Rocchini AP, Hayman LL, Daniels SR. Drug therapy of high-risk lipid abnormalities in children and adolescents: a scientific statement from the American Heart Association Atherosclerosis, Hypertension, and Obesity in Youth Committee, Council of Cardiovascular Disease

**Novelty and Significance**

**What Is Known?**
- Familial hypercholesterolemia (FH) is a common disorder, characterized by severely elevated cholesterol levels from birth onwards.
- High cholesterol levels lead to atherosclerosis, which can be visualized in the early stage by measuring the carotid intima-media thickness (cIMT).

**What New Information Does This Article Contribute?**
- This study shows that young children with FH already have a significant greater cIMT compared with their siblings without FH.
- This study underlines the importance of treatment for children with FH to lower cholesterol levels and prevent atherosclerosis and cardiovascular disease.

FH is characterized by severely elevated cholesterol levels from birth onwards. If left untreated, patients are at high risk of premature cardiovascular disease. Subclinical atherosclerosis can be visualized by measuring cIMT. In this study, we found that children with FH may have a significant greater cIMT compared with their unaffected siblings from about the age of 8 years. Our present results reaffirm the findings of a previous study and extend them to a younger age. This study underlines the importance of lipid-lowering treatment in children with FH, and future trials should focus on the efficacy and safety of starting treatment before atherosclerosis is detectable.
Carotid Intima-Media Thickness in Children With Familial Hypercholesterolemia
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