Familial Hypercholesterolemia with Cholesteryl Ester Transfer Protein Deficiency

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A 21-year-old male was clinically diagnosed with familial hypercholesterolemia (FH) by the manifestations of hypercholesterolemia, tendon xanthoma and family history of premature coronary heart disease. Low density lipoprotein receptor gene was analyzed in attempt to determine a possible point mutation. The normal sequence was partially preserved, and the patient was genetically diagnosed as a heterozygote of FH. In addition, screening for two cholesteryl ester transfer protein (CETP) gene mutations common to Japanese revealed the patient to be a heterozygote of CETP deficiency. A complication of two influential mutations for atherosclerotic ailments was genetically ascertained.

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Key words: low density lipoprotein receptor, gene, mutation, cholesteryl ester, carrier protein

Introduction

Familial hypercholesterolemia (FH) may be the most important disorder of lipid metabolism because of its high frequency and tendency to cause death due to premature coronary artery disease. High levels of plasma low density lipoprotein cholesterol (LDL-C) in FH patients are often resistant to medical treatment. To diagnose FH early and to treat patients appropriately, it is important to investigate the family of FH patients, to prevent the development of atherosclerotic ailments. With more than 150 mutations of the low density lipoprotein (LDL) receptor gene (1), the diagnosis of FH has often been made from clinical manifestations: hypercholesterolemia, tendon xanthoma and a family history of premature coronary heart disease. It is also important to determine particular genetic disorders related to FH to establish FH in other family members.

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Cholesteryl ester transfer protein (CETP) deficiency has almost only been identified in Koreans, Chinese, and Japanese (2, 3), and is characterized by increased high density lipoprotein cholesterol (HDL-C) levels. It is suggested that CETP deficiency may prevent atherosclerotic disease due to its high HDL-C level (4, 5), but recent studies demonstrate atherogenic profiles of CETP deficiency in some conditions (6, 7). We treated a young FH patient with CETP deficiency, which may beneficially affect the prognosis of FH.

Case Report

A 21-year-old male was referred to the hospital due to resistance to treatment with normal doses of simvastatin. The serum LDL cholesterol level was considerably increased after discontinuing medical treatment (Table 1). The family history showed that the father and father’s brother had hypercholesterolemia and the father had experienced a coronary event at the age of 51 (Fig. 1). A roentgenogram of the Achilles tendon certified apparent Achilles tylosis: the thickness was 12 millimeters on the right and 11 millimeters on the left. Thus, the patient was clinically diagnosed with familial hypercholesterolemia.

Signs of arterial atherosclerosis were examined. Maximal treadmill testing showed no evidence of coronary atherosclerosis and echo cardiography showed no plaque just under the aortic valves. This is characteristic of FH (8).

Identification of the mutation of the LDL receptor gene was attempted. The 18 exons of the LDL receptor protein gene, which is located on chromosome 19, were amplified with the polymerase chain reaction method. Each exon was investigated...
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Figure 1. Family tree. Lca: lung carcinoma, D442G: cholesteryl ester transfer protein gene mutation D442G, FH: familial hypercholesterolemia, ys: years old. The proband is indicated by an arrow. The same LDL receptor gene mutation of the patient was demonstrated in the father and the same CETP gene mutation was found in the mother.

Table 1. Serum Lipids and Genetic Mutations in the Family Members

<table>
<thead>
<tr>
<th></th>
<th>Patient</th>
<th>Father</th>
<th>Mother</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cholesterol</td>
<td>332</td>
<td>299</td>
<td>234</td>
</tr>
<tr>
<td>High density lipoprotein-cholesterol</td>
<td>47</td>
<td>74</td>
<td>44</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>58</td>
<td>87</td>
<td>97</td>
</tr>
<tr>
<td>Low density lipoprotein-cholesterol</td>
<td>273</td>
<td>208</td>
<td>166</td>
</tr>
<tr>
<td>Apolipoprotein A-I</td>
<td>119</td>
<td>167</td>
<td>115</td>
</tr>
<tr>
<td>Apolipoprotein B</td>
<td>229</td>
<td>168</td>
<td>131</td>
</tr>
<tr>
<td>Apolipoprotein E</td>
<td>6.2</td>
<td>6.9</td>
<td>5.1</td>
</tr>
<tr>
<td>K790X / D442G</td>
<td>++</td>
<td>++</td>
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</tbody>
</table>

Values are in milligrams per deciliter. The patient’s was measured a month after discontinuing medication, the father’s during medication, the mother’s with no medication. Values of LDL-cholesterol were calculated using the Friedewald formula.

for point mutations with the single strand conformation polymorphism method. The single strand moiety of exon 17 from this patient migrated at a different position from the control, exhibiting a mutation in exon 17 (Fig. 2). The exon 17 sequence was compared with a control by the direct sequence method (Fig. 3). The base sequence AAG of 790 lysine was partially altered to TAG which created a stop codon and it was confirmed to be a heterozygote of K790X, which is a common mutation of FH (9). Since exon 17 constitutes the genetic code of the protein of the transmembrane site of the LDL receptor, it indicated that the moiety of the LDL receptor was lacking after this site.

Screening for two CETP gene mutations common in Japanese, a missense mutation of Asp 442 to Gly in exon 15

Figure 2. The single strand conformation polymorphism. Exon 17 of the patient’s LDL gene is indicated by an arrow. It migrates at a different position from the control and demonstrates a mutation.
Figure 3. DNA sequences of exon 17 of the LDL receptor gene of a control and the case. The arrow indicates a nonsense mutation of A to T in codon 790 (Lysine to stop) in exon 17.

Figure 4. Agarose gel electrophoresis of PCR products after Map I digestion. MW: molecular size marker, N: Normal type, Ho: homozygote. The He is the patient’s and it reveals the patient to be a heterozygote of CETP deficiency D442G.

Discussion

Several population variances in lipoprotein levels can probably be attributed to genetic factors (12, 13). Some facilitate atherosclerosis, and others inhibit it.

Increased levels of LDL-C is the most important change in lipid metabolic disorders since it is a preventable cause of acute coronary syndrome. A number of genetic conditions have been identified that affect LDL-C levels: FH is a relatively common cause of elevated LDL-C levels and is present in about 5% of patients with myocardial infarction (14); it is an autosomal-dominant disorder due to a defective LDL receptor gene on chromosome 19 and the incidence of heterozygotes is about one per 500 individuals (15). A genetic diagnosis of FH is very laborious as there are more than 150 mutations of the LDL receptor gene (1). A large number of these mutations have already been described in Japan, and K790X is a common mutation of the LDL receptor gene, which has been reported from different parts of Japan (9, 16, 17). The hospital has identified this mutation in seven unrelated families in Hokkaido, and identification of gene mutations is useful in determining other affected family members.
The CETP facilitates the transfer of cholesteryl ester from high density lipoprotein to very low density lipoprotein, intermediate density lipoprotein (IDL), and low density lipoprotein (18). The IDL and LDL are then catabolized in the liver by LDL receptors. The transfer of cholesteryl ester occurs bidirectionally and opposite to that of the triglycerides, with an equimolar cholesteryl ester to triglyceride ratio. The HDL becomes enriched with triglycerides after the transfer of cholesteryl ester and is then hydrolyzed by hepatic triglyceride lipase.

CETP deficiency has been attracting attention due to its localization, high frequency in a limited population, and especially its high HDL-C level. Gene mutations of CETP on chromosome 16 are often found in Japan, but are rare in Europe and North America. There are two common mutations, D442G and I14A. The present investigation has shown that the frequency of I14A is 0.8% and D442G 4.6% (11). The CETP deficiency results in hyperalphalipoproteinemia and epidemiological studies have indicated that HDL-C is a negative risk factor for atherosclerotic cardiovascular ailments (4). But it has not been established whether or not elevated HDL-C levels due to CETP deficiency are anti-atherogenic. Some animal studies suggest that CETP deficiency prevents atherosclerosis which results from cholesterol diet (19), whereas recent studies such as in Omagari, Japan, and in Honolulu, USA, suggest the possibility of atherogenicity of CETP deficiency (6, 7).

Calculation from the incidence of CETP deficiency and FH shows that the occurrence of this complication may be about one per 10,000 individuals in Japan. Twenty double heterozygotes of FH and CETP deficiency have already been reported in Kanazawa, Japan (20). But genetic identification of this double heterozygote has never been reported elsewhere. Coexistence of the two genetic factors, that affect atherosclerotic ailments, brings a new point of view to the consideration of the pathogenesis and dynamics of serum cholesterol.

The clinical appearance of the present case is similar to that of FH and CETP deficiency seems to have little effect. However, this case shows a similar HDL-C level as the mother in spite of having FH. Investigation from clinical studies shows that increased levels of LDL-C accelerate CETP activity and lead to a decrease in HDL-C (21, 22). The CETP deficiency of this case may have prevented the decrease of HDL-C. Meanwhile, HDL-C level of the mother is too low to determine her CETP deficiency from the clinical profile. The lipid profile of this family, especially that of this case and the mother, may have been severely modified by environmental factors such as cholesterol diet and smoking, or other genetic factors.

Cholesteryl ester transported by CETP is processed via hepatic LDL receptors. Because of decreased expression of LDL receptors, increased CETP activities may result in an accumulation of unprocessed LDL cholesteryl ester in the case of FH. Accumulation of circulating LDL cholesteryl ester makes it easily oxidized and taken up by macrophages in the subendothelial space. Therefore, the CETP deficiency in FH is thought to be a beneficial metabolic state for atherosclerotic ailments.

Patients with heterozygous FH exhibit a striking diversity in the age at which they develop coronary artery complaints, and even patients with homozygous FH, in whom plasma cholesterol levels exceed 800 mg/dL, may differ by as much as 30 years in the age at which coronary artery disease is expressed clinically (23). The concomitant genetic mutation demonstrated in the present case may help to explain such variation.

References

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