Hereditary Serum Cholinesterase Deficiency Associated with Severe Lipid Deposition in the Kidney

Kazuho Honda, Wako Yumura, Junko Arai, Tsutomu Sanaka, Hiroshi Nihei and Nobuhiro Sugino

A 47-year-old woman who was homozygous for a silent cholinesterase gene (hereditary serum cholinesterase deficiency) presented with nephrotic syndrome and hyperlipidemia. Renal biopsy performed in 1986 demonstrated mesangial proliferative glomerulonephritis. Four years later, a second biopsy revealed progression with mesangial interpositions and severe lipid deposition in the glomeruli, tubules and interstitium. This is the first case of hereditary serum cholinesterase deficiency accompanied by renal disease. Serum cholinesterase deficiency may be related to hyperlipidemia and abnormal lipid deposition in the kidney, which promotes the progression of renal disease.

Key words: butyryl cholinesterase (pseudocholinesterase), hyperlipidemia, nephrotic syndrome, low density lipoprotein (LDL), LDL apheresis

Introduction

Hereditary serum cholinesterase (ChE) deficiency is a rare disease that was first described by Forbat and Lehmann in 1953 (1). Affected patients show a prolonged response to succinylcholine used in anesthesia due to their reduced ability to catabolize this agent. Except for problems with anesthesia, no clinical abnormalities have been reported in individuals with hereditary serum ChE deficiency. While ChE activity has been detected in various organs (2) including the kidney (3, 4), its biologic function has not been clarified. Recently, ChE activity has been suggested to play a role in lipid metabolism (5, 6) and atherosclerosis (7).

We describe a patient who had hereditary serum ChE deficiency accompanied by chronic glomerulonephritis with severe lipid deposition. The relationship between serum ChE deficiency and abnormal lipid metabolism in the kidney will be discussed.

Case Report

This 47-year-old woman had an eight-year history of proteinuria. She had been admitted to the Kidney Center of Tokyo Women's Medical College in 1986 when she was 43 years old because of proteinuria (1.5 g/day), microhematuria (1–2 RBC/HPF), hyperlipidemia (serum total cholesterol: 326 mg/dl, serum low density lipoprotein: 1,027 mg/dl) and undetectable serum ChE activity (<0.01 ApH). Renal biopsy performed at the time demonstrated mesangial proliferative glomerulonephritis. Despite corticosteroid treatment (prednisolone, 40 mg/day), the proteinuria gradually worsened over the next four years, reaching a level of 8.6 g/day early in 1990. She was readmitted to our hospital for further examination in May 1990.

Physical examination revealed mild edema of the lower extremities. There was no xanthoma of the skin. Blood pressure was 140/85 mmHg. Proteinuria (4.7 g/day) and slight microscopic hematuria (RBC 3/HPF) with occasional lipid droplets and lipid-storing cells in the urine were present. Glycosuria was absent. Serum total protein (4.9 g/dl) and albumin (2.8 g/dl) were decreased, and serum creatinine (0.8 mg/dl) and blood urea nitrogen (10 mg/dl) were within the normal range. Serum total cholesterol (307 mg/dl) and LDL (1,120 mg/dl) were increased. Details of the serum lipid analysis appear in Table 1. Lecithin cholesterol acyltransferase (LCAT) was within the normal range (56.1 nmol/ml/h). Serum immunoglobulin levels and complement activity were normal. Tests for serum autoantibodies and cryoglobulin were all negative, and serum immune complexes showed no elevation. The glomerular filtration rate (GFR) measured by inulin clearance was mildly decreased.
No abnormalities were detected on the PSP test (15 minutes: 34.7%, 120 minutes: 78.5%) or Fishberg's test (1.032 in specific gravity, 927 mOsm/l in maximum). Renal biopsy demonstrated the progression of the glomerular lesion with frequent mesangial interposition and severe lipid deposition in the kidney.

The administration of pravastatin, probucol and cholestyramine and treatment with LDL apheresis were begun in an attempt to reduce the hyperlipidemia and thus to halt the progression of the renal disease. After 6 months of LDL apheresis (a total of 15 sessions), serum LDL remained within the normal range; however, the proteinuria worsened, as summarized in Fig. 1.

The family of the patient is shown in Fig. 2. There were no consanguineous marriages in her family. Neither renal disease nor hyperlipidemia was found in her family history.

### Materials and Methods

1) **Measurement of serum ChE activity**

Sera obtained from the patient, her four sons and her husband were analyzed. Serum ChE activity was determined by the enzymatic method using a Determiner ChE-S® assay kit (Kyowa Hakko Kogyo Co. Ltd., Tokyo) (8). Choline was liberated from the substrates (o-toluoylcholine) by ChE and oxidized by choline oxidase to betaine with the simultaneous production of hydrogen peroxide, which was determined using 4-aminoantipyrine and phenol in the presence of peroxidase. After additional incubation at 37°C for 20 minutes, the resultant red color was measured at 500 nm against a reagent blank by photoabsorptometry. The reference range of healthy subjects is from 300 to 720 IU/l using this method.

2) **Pathological analysis of renal biopsy specimens**

The renal tissue specimens obtained at biopsy were

![Fig. 1. Clinical course of the patient. Her proteinuria increased and her serum total protein gradually decreased. LDL apheresis combined with lipid-lowering drug therapy was not effective in halting disease progression although serum LDL was maintained within the normal range.](image-url)
Hereditary ChE Deficiency with Renal Disease

1) Serum ChE activity
The serum ChE activity of the patient, all of her sons and her husband is shown in Table 2. The serum ChE activity of the patient was below 1 IU/l (undetectable). The serum ChE activity of her first and second sons was decreased (258 and 271 IU/l, respectively) but that of her third and fourth sons (twins) was within the normal range (526 and 451 IU/l, respectively). The serum ChE activity of her husband was within the normal range (451 IU/l). Serum total cholesterol, HDL cholesterol and triglyceride values are also shown in Table 2. The total cholesterol, HDL cholesterol and triglyceride levels of her family were almost normal.

2) Pathology of renal biopsy specimens
(a) Light microscopic findings
The biopsy specimen obtained in June 1986 contained five glomeruli. Each showed mild proliferation of the mesangial cells and an increase in the mesangial matrix with focal and segmental accentuation. Mesangial interposition was partially seen (Fig. 3). No abnormalities of the tubules, interstitium and vessels were found.

Fig. 3. Glomerular changes present at the first biopsy in June 1986. Mild mesangial proliferation with focal and segmental mesangial interposition was seen (PAS Stain, x 100).

Table 2. Serum ChE Activity and Total Cholesterol (T. Chol), HDL Cholesterol (HDL Chol) and Triglyceride (TG) Levels of the Patient and Her Family

<table>
<thead>
<tr>
<th></th>
<th>Normal range</th>
<th>Patient</th>
<th>First son</th>
<th>Second son</th>
<th>Third son</th>
<th>Fourth son</th>
<th>Husband</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td></td>
<td>47</td>
<td>24</td>
<td>22</td>
<td>17</td>
<td>17</td>
<td>52</td>
</tr>
<tr>
<td>ChE (IU/l)</td>
<td>300–720</td>
<td>&lt;1 IU/l</td>
<td>258</td>
<td>271</td>
<td>526</td>
<td>451</td>
<td>451</td>
</tr>
<tr>
<td>T. Chol (mg/dl)</td>
<td>130–250</td>
<td>307</td>
<td>134</td>
<td>136</td>
<td>134</td>
<td>139</td>
<td>171</td>
</tr>
<tr>
<td>HDL Chol (mg/dl)</td>
<td>42–72</td>
<td>32</td>
<td>37</td>
<td>33</td>
<td>62</td>
<td>63</td>
<td>n.e.</td>
</tr>
<tr>
<td>TG (mg/dl)</td>
<td>40–150</td>
<td>107</td>
<td>105</td>
<td>156</td>
<td>69</td>
<td>116</td>
<td>n.e.</td>
</tr>
</tbody>
</table>

n.e.: not examined.
No apparent accumulation of lipids was observed. The biopsy specimen obtained in May 1990 contained eight glomeruli with one hyalinized glomerulus. The mesangial area was severely expanded, with an increase in the mesangial matrix and mild proliferation of the mesangial cells. Foam cells were seen within the glomerular capillary lumina and fine vacuolar changes were observed in the mesangium. Mesangial interpositions were frequently seen and the glomerular capillary lumina were generally obscured. Exudative changes such as fibrin caps were occasionally seen in the paramesangial area and the glomerular capillary tufts (Fig. 4). Sudan Black B staining of the frozen sections revealed that these foam cells contained neutral lipid substances and, furthermore, that the fine vacuolar changes in the mesangium were caused by lipid deposition (Fig. 5). Foamy degeneration of the tubular cells and foam cells in the interstitium were frequent findings (Fig. 6).

(b) Immunohistochemical findings

Immunofluorescent examination of the frozen sections demonstrated that granular and lumpy staining of IgG (Fig. 7), IgA, IgM, Clq and C3 was present predominantly along the capillary walls and partially in the mesangium. These findings were also observed at the first biopsy, but the lumpy staining along the capillary wall was not as conspicuous as that seen at the second biopsy. At the second biopsy, intense and granular staining of apolipoprotein (apo) B was present both in the mesangium and along the capillary walls (Fig. 8). The deposition of apo E followed the same pattern as that of apo B.

Fig. 4. Glomerular changes present at the second biopsy in May 1990. Mesangial interposition became frequent and the glomerular capillary lumina were generally obscured. Foamy cells within the capillary lumina and fine vacuolar changes in the mesangium were frequently seen. Exudative changes in the paramesangium and capillary loops were also observed (PAS Stain, ×100).

Fig. 5. In the second biopsy specimen, Sudan Black B staining of a frozen section demonstrated lipid deposition in the foamy cells and the vacuolar lesion in the mesangium (Sudan Black B, ×160).

Fig. 6. Foamy degeneration of the tubular cells and foamy cells in the interstitium were occasionally seen at the second biopsy (PAS Stain, ×100).

Fig. 7. Granular and lumpy staining of IgG, predominantly in the capillary wall, at the second biopsy (IgG, ×100). Similar results were seen for IgA, IgM, Clq and C3. The immunofluorescence findings of the first biopsy resembled those of the second biopsy, although the lumpy staining was not so conspicuous.
(c) Electron microscopic findings

Electron microscopic examination of the second biopsy specimen revealed frequent electron-dense deposits of various sizes in the capillary walls and mesangium of the glomerulus. The deposits in the capillary walls, seen more frequently, were usually present in the subendothelial spaces, but sometimes extended into the intramembranous and subepithelial spaces of the GBM. Myelin-like figures were sometimes seen in the mesangial cells (Fig. 9). Vacuolar formation was observed in the mesangial cells (Fig. 10). At the first renal biopsy, similar electron deposits were seen in the capillary walls and mesangium, but not as frequently as at the second biopsy. Vacuolar changes in the mesangial cells and infiltration of the foamy macrophages were not seen in the first biopsy specimen.

Discussion

Serum cholinesterase (nonspecific or pseudocholinesterase, butyryl cholinesterase, EC3.1.1.8.: BChE) differs from acetyl cholinesterase (specific or true cholinesterase, EC3.1.1.7.: AChE), which is a neurotransmitter. Hereditary deficiency of serum cholinesterase is caused by a genetic variant of BChE. Two genetic loci (E1 and E2) determining BChE activity in plasma have been reported (9). The silent BChE gene is a variant gene of the E1 locus, and homozygotes for the silent BChE gene have no BCh-E activity in plasma or the liver, as in the present case.

Clinically, serum ChE activity is used as an indicator of the biosynthetic activity of the liver; however, the function of ChE is not yet fully understood. An increase in serum ChE activity is usually observed in conditions associated with an abnormal lipid metabolism such as hyperlipidemia or nephrotic syndrome (10, 11). Kutty et al reported that serum LDL shows ChE activity, and suggested that LDL is formed from VLDL in the presence of ChE (5). These observations suggest that ChE activity influences the metabolism of serum lipo-
proteins. Although we cannot determine whether the hyperlipidemia seen in the present case should be ascribed to the secondary effects of nephrotic syndrome or serum ChE deficiency, the presence of hyperlipidemia during the early period before the degree of proteinuria reached the nephrotic range suggests the latter possibility.

Hyperlipidemia is thought to exacerbate the progression of glomerular sclerosis by the accumulation of lipids in the glomerular tissue (12). This hypothesis is supported by various clinical observations and experimental models. In the kidney, serum LDL can be taken up by the LDL receptors that exist in glomerular endothelial cells (13), epithelial cells (14) and mesangial cells (15). The ligands for the LDL receptors are apo B and apo E, which were intensely stained in the glomeruli of the present patient. Modified (oxidized) LDL is also taken up by macrophages and mesangial cells via scavenger receptors (16). The uptake of lipids by scavenger receptors causes intracellular accumulation of lipids in the form of vacuoles, leading to foam cell formation (17). Here, we observed vacuolar changes in mesangial cells and foamy macrophages in glomerular capillaries. This suggests that modified LDL was also taken up in the glomeruli in the present patient. While the function of ChE in the kidney has not been elucidated, the present case emphasizes the fact that ChE may regulate the uptake or intracellular metabolism of lipids and a ChE deficiency may promote the accumulation of lipids as seen in this case. Further studies must be performed before conclusions about the relationship between ChE and lipid metabolism can be drawn.

Abnormal accumulation of lipids occurs in the kidneys of patients with several hereditary enzymatic disorders, such as lipid storage disease and a deficiency of lecithin cholesterol acyltransferase (LCAT) (18). In the present patient, the LCAT activity was normal, and no abnormal deposition of lipids was detected in various organs such as the liver, spleen and heart.

Recently, an unusual glomerulopathy with lipid deposition, termed "lipoprotein glomerulopathy", was reported as a new renal disease that was presumably induced by abnormal lipid metabolism (19). Histologically, the above-mentioned condition is characterized by marked dilatation of the glomerular capillary lumina and the presence of lipoprotein thrombi within the lumina. The serum apolipoprotein profile revealed a high level of apo E. In the present case however, neither marked dilatation of the capillary lumina nor lipoprotein thrombi were observed, and serum apo E was not elevated, thus excluding the diagnosis of lipoprotein glomerulopathy.

LDL apheresis removes the serum LDLs by extracorporeal circulation and is useful in patients with familial hypercholesterolemia (20). Recently, this treatment has also been given to patients with focal glomerulosclerosis (FGS), and it has proved effective in some cases (21). In the present patient, LDL apheresis was performed 15 times combined with the administration of pravastatin, a 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitor (HMG-CoA inhibitor), probucol and cholesteryramine for 6 months. Although the serum LDL level of the patient fell and was maintained within the normal
range, her proteinuria worsened. This suggests that the normalization of serum LDL for a short period of time did not lead to the remission of renal disease, and that other factors in addition to serum LDL regulate the pathogenesis of lipid-induced glomerular injury.

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References