Lipoprotein Lipase Deficiency (R243H) in a Type 2 Diabetes Patient with Multiple Arterial Aneurysms

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Abstract

Lipoprotein lipase (LPL) deficiency is a rare monogenic disorder that manifests as severe hypertriglyceridemia. Whether or not LPL deficiency accelerates the development of atherosclerosis remains controversial. We herein report a 66-year-old woman who was homozygous for the R243H LPL mutation. She had developed multiple arterial aneurysms and systemic atherosclerosis despite good control of other atherogenic risk factors, including diabetes. Furthermore, although intensive pharmaceutical therapies had been minimally effective, medium chain triglyceride (MCT) therapy reduced the serum triglyceride levels. Thus, this case suggests important role that mutated LPL protein plays in the progression of atherosclerosis and that MCT therapy is potentially effective, even for severe hypertriglyceridemia due to LPL deficiency.

Key words: lipoprotein lipase (LPL) deficiency, R243H, atherosclerosis, aneurysm, medium chain triglyceride (MCT) therapy

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Introduction

Lipoprotein lipase (LPL) is the key enzyme for hydrolysis and the removal of chylomicrons (CMs) from the circulation (1). A decreased LPL activity is responsible for most cases of type I hyperlipidemia and a portion of those with type V hyperlipidemia. Most notably, familial LPL deficiency caused by mutations of the LPL gene is a rare autosomal recessive disorder, which is estimated to affect about one in one million people (2). LPL deficiency causes a marked serum triglyceride (TG) elevation, with levels reaching 1,500-25,000 mg/dL, which frequently results in recurrent attacks of pancreatitis. On the other hand, it was believed that LPL deficiency does not lead to atherosclerosis development, because CM particles are too large to penetrate the endothelium of the arterial wall (3). However, in recent years, there have been several reports indicating that patients with LPL deficiency do, in fact, show accelerated atherosclerosis (4-8). Therefore, whether or not hypertriglyceridemia caused by LPL deficiency leads to atherosclerosis remains a controversial topic.

We herein report a 66-year-old patient with type V hyperlipidemia who was homozygous for a missense mutation of the LPL gene. She had hypertension and type 2 diabetes mellitus (T2DM), both of which had been well controlled since initially being diagnosed. To the best of our knowledge, only two cases with this homozygous missense mutation have been reported, but these patients were too young (23 days and 2 months after birth) for an evaluation of atherosclerosis (6, 9). Notably, our present case had developed multiple arterial aneurysms and several atherosclerotic lesions in the coronary and carotid arteries, suggesting that this mutation may thus play a role in atherosclerotic progression. It is also interesting to note that diet therapy with medium chain triglycerides (MCTs) was useful for reducing...
iliac arteries (Fig. 1A) with diffuse atherosclerosis. There was no family history of consanguinity, but her brother also had severe hypertriglyceridemia and had experienced recurrent episodes of acute pancreatitis. She had given birth three times, uneventfully. Severe hypertriglyceridemia was first diagnosed at 38 years of age. Drug therapies were started soon after the diagnosis, but her serum TG levels had remained within the range of 500-5,000 mg/dL for 28 years. During the course, hypertension and T2DM had been diagnosed at 38 and 62 years of age, respectively. Since the onsets of these diseases, her blood pressure and blood glucose levels had been under strict control with medications including 10 mg/day of cilnidipine, 2 mg/day of doxazosin mesilate and 2.5 mg/day of carvedilol to keep blood pressure below 130/60 mmHg, and 100 mg/day of vildagliptin to keep HbA1c below 5.7%. She was taking cilnidipine 10 mg/day, doxazosin mesilate 2 mg/day and carvedilol 2.5 mg/day to maintain blood pressure below 130/60 mmHg, and vildagliptin 100 mg/day to keep below HbA1c 5.7%. In fact, she had developed no diabetic microvascular complications. Furthermore, no other atherosclerogenic risk factors were present. Considering these observations together, severe hypertriglyceridemia due to LPL mutations together, severe hypertriglyceridemia due to LPL mutations.

The serum TG levels, even though intensive pharmaceutical therapy with four anti-hyperlipidemic drugs had only been minimally effective.

Case Report

A 66-year-old Japanese woman with multiple iliac arterial aneurysms was admitted to our hospital to undergo an endovascular aortic repair (EVAR) operation. She also had severe hypertriglyceridemia with a serum TG level of 2,942 mg/dL on admission (Table) despite taking four anti-hyperlipidemic drugs, including 160 mg/day of fenofibrate, 500 mg/day of nicotinol, 4 g/day of n-3 polyunsaturated fatty acids (PUFAs) and 10 mg/day of ezetimibe. She had a history of acute pancreatitis at age 19 years. She was 1.56 m in height, weighed 44.3 kg and had a body mass index of 17.8. She neither smoked nor drank alcohol. There was no family history of consanguinity, but her brother also had severe hypertriglyceridemia and had experienced recurrent episodes of acute pancreatitis.

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A three-dimensionally-reconstructed computed tomography (CT) scan revealed multiple arterial aneurysms in the iliac arteries (Fig. 1A) with diffuse atherosclerosis. There were no symptoms of infectious diseases, such as syphilis, or Marfan syndrome. In addition, coronary angiography showed 90% stenosis of the distal left anterior descending artery segment 8, and ultrasonography indicated multiple isoechoic and hyperechoic plaques in the common carotid arteries. She had a homozygous missense mutation of the LPL gene (Arg243→His: R243H) (Fig. 2), which resulted in very low plasma post-heparin LPL activity (Table). In contrast, a substantial LPL protein mass was detected in both the pre- and post-heparin plasma (Table). Polyacrylamide gel electrophoresis (Fig. 3A) and the overnight plasma standing test (Fig. 3B) revealed remarkable increases in CM and very low density lipoprotein (VLDL), indicating type V hyperlipidemia according to the Fredrickson classification (10). A low-fat diet (20 g/day) was started under supervision during hospitalization, leading to a decrease in her serum TG level (616 mg/dL). The EVAR operation was successfully performed on the left internal iliac artery aneurysm (Fig. 1B).

After discharge, due to discontinuation of the low-fat diet at home, the serum TG level again rose to levels similar to those prior to hospitalization. Therefore, MCT therapy, in which her usual cooking plant oil was replaced with 10 g of MCT enriched oil (Makuton oil™, Kissei Pharmaceutical Co., Nagano, Japan), was initiated. Her lipid profile thereafter improved as demonstrated by high performance liquid chromatography (Fig. 4). Furthermore, increasing the amount of MCT to 20 g gradually decreased the serum TG levels to less than 1,000 mg/dL (Fig. 5). As a result, we therefore consider MCT therapy to be effective for severe hypertriglyceridemia, even that due to a complete absence of LPL activity.

Discussion

This case with type V hyperlipidemia attributable to a homozygous missense mutation in the LPL gene had developed severe atherosclerosis including multiple arterial aneurysms and several lesions in the coronary and carotid arteries. The contributions of other atherogenic risk factors, such as gender, age, smoking, hypertension and T2DM should also be taken into consideration. She had been treated for hypertension and T2DM for 28 and 4 years, respectively. However, because her blood pressure and glucose levels had been regularly and carefully checked in connection with the management of hypertriglyceridemia, both of these diseases were detected immediately after their onsets and had been well controlled since initially being diagnosed. When she was admitted to our hospital for an aneurysm operation, she was taking cilnidipine 10 mg/day, doxazosin mesilate 2 mg/day and carvedilol 2.5 mg/day to maintain blood pressure below 130/60 mmHg, and vildagliptin 100 mg/day to keep below HbA1c 5.7%. In fact, she had developed no diabetic microvascular complications. Furthermore, no other atherosclerogenic risk factors were present. Considering these observations together, severe hypertriglyceridemia due to LPL mutation.
Figure 1. A three-dimensionally-reconstructed computed tomography scan. (A) A pre-operative figure shows multiple arterial aneurysms (arrows). (B) A post-operative figure shows a left internal iliac artery aneurysm treated with endovascular aortic repair (EVAR) (arrows).

| Patient | Control |
|---------|---------|
| A       | G       |
| C C A C G A G A C T C C A | C C A C G A G C C T C C A |
| His 241 Glu 242 His 243 Ser 244 | His 241 Glu Arg 243 Ser 244 |

Figure 2. Nucleotide sequence in Exon 6 of the LPL gene.

Figure 3. Polyacrylamide gel electrophoresis, followed by lipoprotein quantification with densitometric analysis (A) and the overnight plasma standing test (B).
deficiency, rather than other atherogenic risk factors, likely contributed to the progression of atherosclerosis in this case.

However, in contrast to heterozygosity for the LPL gene mutations (11), a complete absence of LPL activity due to homozygous or compound heterozygous mutations has generally been considered to be non-atherogenic (12), because the CM particles, in which triglycerides are not hydrolyzed, are thought not to penetrate the vessel walls due to their large sizes (3). On the other hand, in recent years, several reports have indicated that atherosclerotic diseases do, in fact, develop in patients with some forms of LPL deficiency (4-8). This issue thus remains controversial.

The present patient had a homozygous missense mutation of the LPL gene (R243H) (Fig. 2), and exhibited almost a complete absence of LPL activity (Table) with a substantial LPL protein mass (Table). Arg243 of LPL is among the strictly conserved amino acid residues. The sites of Ser132, Asp156 and His241 in LPL are reportedly essential for the formation of a catalytic triad with triglycerides (13). In addition, Gotoda et al. demonstrated R243H-mutated LPL expressed in COS-1 cells to have no enzymatic activity. Japanese (9) and Chinese (6) patients with the same homozygous missense mutation at this site have been reported, and both also showed a complete absence of LPL activity. However, the atherogenic effect of this mutation remains uncertain because these patients were too young (23 days and 2 months after birth) for any evaluation of atherosclerosis. Our observations in the present case may shed light on the atherogenic effect of this LPL mutation.

In contrast to the LPL activity, the LPL protein mass was
preserved (90.8 ng/mL, which is 45.5% of control reference). Consistently, COS-1 cells expressing R243H-mutated LPL reportedly secrete the mutated LPL protein (9). It has been proposed that LPL protein lacking any lipase activity may function as a bridge between apoB-containing lipoproteins and proteoglycans on vessel walls, thereby retaining atherogenic lipoproteins on endothelial cells, and leading to the progression of atherosclerosis (7, 14). In fact, patients with complete LPL deficiency, whereby both enzymatic activity and protein mass are absent, reportedly exhibit a non-atherogenic phenotype (15, 16), while patients with nonsense mutations which result in the absence of LPL activity with retention of the LPL protein are prone to atherogenesis (4, 6-8). Therefore, functionally inactive R243H-LPL protein may have played a role in the development of the severe atherosclerosis observed in this patient.

The present patient had developed both multiple aneurysms in the iliac arteries and systemic atherosclerosis. In general, arterial aneurysm formation has been regarded as a focal manifestation of advanced atherosclerosis (17). Indeed, aneurysm and atherosclerosis reportedly have common several risk factors, such as smoking, hypertension, hypertriglyceridemia and low HDL cholesterol levels (18). In our patient as well, a CT scan demonstrated multiple arterial aneurysms that were accompanied by diffuse atherosclerosis in the same arteries. As is the case with atherosclerosis, an arterial aneurysm is also a rare clinical feature of LPL deficiency, although hypertriglyceridemia is one of the risk factors for aneurysm development. However, there is one report describing a patient with LPL deficiency who had an arterial aneurysm (7). The reported patient had a homozygous mutation (L303F) in the LPL gene and suffered from both abdominal aortic aneurysm and multiple stenosis in the coronary, renal and femoral arteries. In addition, the homozygous L303F mutation caused the absence of LPL activity with a substantial LPL mass. These findings, which were also observed in our case, may support the notion that inactive LPL protein exerts an atherogenic effect.

Another important finding in our LPL-deficient patient is that MCT therapy was effective for hypertriglyceridemia, which had been resistant to multi-drug therapies. MCTs consist of a mixture of C6:0-C12:0 medium-chain fatty acids. Since absorbed MCTs are carried to the liver via the portal vein without forming CM lipoprotein, the replacement of dietary long chain triglycerides with MCTs has been proposed as a potentially useful treatment for severe hypertriglyceridemia (19). However, the efficacy of this therapy for mild hypertriglyceridemia has not as yet been established (20, 21). Tremblay et al. (22) demonstrated that MCT therapy has no effect on the serum TG levels in insulin-resistant patients. The TG elevation in patients with insulin resistance mainly results from an increased production of VLDL lipoprotein in the liver and its decreased clearance, rather than an increased production of CM lipoprotein. In contrast, in our case, MCT therapy was markedly useful for improving severe hypertriglyceridemia with LPL deficiency.

To prevent the development and exacerbation of acute pancreatitis in LPL-deficient patients, fat restriction (20 g/day or 15% of total calories) is highly recommended (23, 24). When fat restriction is difficult to achieve, then MCT therapy is sometimes attempted (25). In our case, the serum TG levels worsened by 5,000 mg/dL, after hospital discharge due to discontinuation of the fat restricted diet at home. We thus initiated MCT therapy, resulting in the serum TG decreasing to levels below 1,000 mg/dL, in an ambulatory setting (Fig. 5). These results suggest that the inhibition of CM formation by MCT therapy may therefore be an effective treatment alternative in the management of severe hypertriglyceridemia due to LPL deficiency.

The authors state that they have no Conflict of Interest (COI).

References
1. Eckel RH. Lipoprotein lipase. A multifunctional enzyme relevant to common metabolic diseases. N Engl J Med 320: 1060-1068, 1989.
2. Havel RJ. Triglyceride-rich lipoproteins and plasma lipid transport. Arterioscler, Thromb Vasc Biol 30: 9-19, 2010.
3. Ebara T, Ramakrishnan R, Steiner G, Shachter NS. Chylomicronemia due to apolipoprotein CIII overexpression in apolipoprotein E-null mice. Apolipoprotein CIII-induced hypertriglyceridemia is not mediated by effects on apolipoprotein E. J Clin Invest 99: 2672-2681, 1997.
4. Benlian P, De Gennes JL, Foubert L, Zhang H, Gagne SE, Hayden M. Premature atherosclerosis in patients with familial chylomicronemia caused by mutations in the lipoprotein lipase gene. N Engl J Med 335: 848-854, 1996.
5. Hoeg JM, Osborne JC Jr, Gregg RE, Brewer HB Jr. Initial diagnosis of lipoprotein lipase deficiency in a 75-year-old man. Am J Med 75: 889-892, 1983.
6. Ma Y, Liu MS, Chitayat D, et al. Recurrent missense mutations at the first and second base of codon Arg243 in human lipoprotein lipase in patients of different ancestries. Hum Mutat 3: 52-58, 1994.
7. Saika Y, Sakai N, Takahashi M, et al. Novel LPL mutation (L303F) found in a patient associated with coronary artery disease and severe systemic atherosclerosis. Eur J Clin Investigat 33: 216-222, 2003.
8. Shrama F, Puntoni M, Bigazzi F, et al. Cognitive impairment and polidistrectal atherosclerotic disease in chylomicronemia syndrome: a case report. Transfus Apher Sci 49: 323-327, 2013.
9. Gotoda T, Yamada N, Kawamura M, et al. Heterogeneous mutations in the human lipoprotein lipase gene in patients with familial lipoprotein lipase deficiency. J Clin Invest 88: 1856-1864, 1991.
10. Fredrickson DS, Lees RS. A system for phenotyping hyperlipoproteinemia. Circulation 31: 321-327, 1965.
11. Witstrup HH, Tybjerg-Hansen A, Ribaldgaard S, Steffensen R, Schnohr P, Nordestgaard BG. A common substitution (Asn291Ser) in lipoprotein lipase is associated with increased risk of ischemic heart disease. J Clin Invest 99: 1606-1613, 1997.
12. Havel RJ, Gordon RS Jr. Idiopathic hyperlipemia: metabolic studies in an affected family. J Clin Invest 39: 1777-1790, 1960.
13. Winkler FK, D’Arcy A, Hunziker W. Structure of human pancreatic lipase. Nature 343: 771-774, 1990.
14. Clee SM, Bissada N, Miao F, et al. Plasma and vessel wall lipoprotein lipase have different roles in atherosclerosis. J Lipid Res 41: 521-531, 2000.
15. Ebara T, Okubo M, Horinishi A, Adachi M, Murase T, Hirano T.
No evidence of accelerated atherosclerosis in a 66-yr-old chylomiconemia patient homozygous for the nonsense mutation (Tyr61-stop) in the lipoprotein lipase gene. Atherosclerosis 159: 375-379, 2001.

16. Kawashiri MA, Higashikata T, Mizuno M, et al. Long-term course of lipoprotein lipase (LPL) deficiency due to homozygous LPL (Arita) in a patient with recurrent pancreatitis, retained glucose tolerance, and atherosclerosis. J Clin Endocrinol Metab 90: 6541-6544, 2005.

17. Patel MI, Hardman DT, Fisher CM, Appleberg M. Current views on the pathogenesis of abdominal aortic aneurysms. J Am Coll Surg 181: 371-382, 1995.

18. Stather PW, Sidloff DA, Dattani N, et al. Meta-analysis and meta-regression analysis of biomarkers for abdominal aortic aneurysm. Br J Surg 101: 1358-1372, 2014.

19. Furman RH, Howard RP, Brusco OJ, Alaupovic P. Effects of medium chain length triglyceride (MCT) on serum lipids and lipoproteins in familial hyperchylomicronemia (dietary fat-induced lipemia) and dietary carbohydrate-accentuated lipemia. J Lab Clin Med 66: 912-926, 1965.

20. Bach AC, Ingenbleek Y, Frey A. The usefulness of dietary medium-chain triglycerides in body weight control: fact or fancy? J Lipid Res 37: 708-726, 1996.

21. Greenberger NJ, Skillman TG. Medium-chain triglycerides. N Engl J Med 280: 1045-1058, 1969.

22. Tremblay AJ, Lamarche B, Labonté ME, Lépine MC, Lemelin V, Couture P. Dietary medium-chain triglyceride supplementation has no effect on apolipoprotein B-48 and apolipoprotein B-100 kinetics in insulin-resistant men. Am J Clin Nutr 99: 54-61, 2014.

23. Gotoda T, Shirai K, Ohta T, et al. Diagnosis and management of type I and type V hyperlipoproteinemia. J Atheroscler Thromb 19: 1-12, 2012.

24. Miller M, Stone NJ, Ballantyne C, et al. Triglycerides and cardiovascular disease: a scientific statement from the American Heart Association. Circulation 123: 2292-2333, 2011.

25. Shirai K, Kobayashi J, Inadera H, et al. Type I hyperlipoproteinemia caused by lipoprotein lipase defect in lipid-interface recognition was relieved by administration of medium-chain triglyceride. Metabolism 41: 1161-1164, 1992.