Correlation of Plasma Oxidized Low-Density Lipoprotein Levels to Vascular Complications and Human Serum Paraoxonase in Patients With Type 2 Diabetes

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The oxidative modification of low-density lipoprotein (LDL) plays a central role in the initiation and acceleration of atherosclerosis. Human serum paraoxonase (PON1) is associated with high-density lipoprotein (HDL) and has been shown to reduce the susceptibility of LDL to lipid peroxidation. We investigated whether circulating oxidized LDL (Ox-LDL) levels were associated with diabetic vascular complications, and whether the enzymatic activity and gene polymorphisms of PON1 influenced Ox-LDL concentrations in vivo. There was no difference in the plasma Ox-LDL concentrations between diabetic patients with and without macrovascular diseases. However, Ox-LDL concentrations corrected by LDL-cholesterol (OxLDL/LDL-C) or apolipoprotein B (apoB) concentrations (Ox-LDL/apoB), which probably reflect the proportion of oxidatively modified LDL to total LDL particles, were significantly higher in patients with macrovascular diseases than in those without. In addition, patients with peripheral neuropathy had a significantly higher Ox-LDL/apoB ratio than patients without this complication. The genotype TT of −108C/T polymorphism in the promoter region of the PON1 gene, which is associated with decreased PON1 expression, showed a significantly higher Ox-LDL/apoB ratio than genotypes TC or CC (TT: 0.60 ± 0.15, CT + CC: 0.55 ± 0.11, $P = .02$). Stepwise multiple regression analysis for Ox-LDL concentration revealed that the −108C/T polymorphism, subsequently to apoB concentration, was identified as a significant contributor. In summary, the Ox-LDL/apoB ratio was associated with macrovascular disease and peripheral neuropathy in Japanese patients with type 2 diabetes. Increased Ox-LDL/apoB may result, at least partly, from reduced serum antioxidant capacity in the diabetic state, including the attenuation of PON1 action. Increased Ox-LDL/apoB could be a significant marker for susceptibility to vascular complications in diabetic patients.

THE OXIDATIVE modification of low-density lipoprotein (LDL) plays a central role in the initiation and acceleration of atherosclerosis.[1,2] Oxidized LDL (Ox-LDL) exerts several proatherogenic effects, including the increased synthesis and secretion of adhesion molecules, monocyte chemotaxis and adhesion, cytotoxicity to endothelial cells, enhanced foam cell formation, and increased smooth muscle cell proliferation.[3,4] Oxidative modified lipoproteins have been isolated from atherosclerotic lesions, and the oxidation of lipoproteins is generally considered to occur in the vessel wall.[3,4] On the other hand, oxidized lipoproteins are also identified in the circulation, although it remains unclear whether the plasma oxidized lipoproteins originate in the arterial wall (back-diffusion) or are produced in the circulation.

Lipid oxidation is believed to occur in response to increased oxidative stress or deficiency of endogenous antioxidants. Human serum paraoxonase (PON1), an esterase, is associated with apolipoprotein A-I (apoA-I) and apo J in high-density lipoprotein (HDL).[5] Because both purified PON1 and HDL-associated PON1 inhibit LDL oxidation in vitro,[6,7] PON1 is believed to play an important role in LDL protection against oxidation in vivo. PON1 has 2 polymorphic sites in the coding region: Leu-Met (L/M) at position 55 of the amino acid sequence and Gln-Arg (Q/R) at position 192. The 192Q/R polymorphism is involved in serum paraoxonase activity and paraoxonase activity in subjects with the RR genotype is higher than in those with the QQ genotype.[8,9] The 55L/M polymorphism is associated with the PON1-expression level, and PON1 concentration is higher in the LL genotype than in the MM genotype.[10-13] Recently, we and other investigators[14-16] identified a −108C/T polymorphism in the promoter region of the PON1 gene and found that this polymorphism affects the promoter activity and serum concentration of PON1. The association of 55L/M polymorphism with PON1 expression level appears to be associated with linkage disequilibrium with the −108C/T polymorphism.[15,16]

Oxidative stress has been demonstrated to be increased in vivo in the diabetic state.[17,18] We and other investigators have shown that serum paraoxonase activity is lower in diabetic patients and is lower yet in those with diabetic complications, independent of PON1 gene polymorphisms.[19,20] Despite accumulated data on the effect of PON1 on LDL oxidation in vitro, there are no available data showing a direct correlation between plasma Ox-LDL and PON1 in vivo. We therefore measured plasma Ox-LDL concentrations in patients with type 2 diabetes and investigated the correlation of Ox-LDL concentration with diabetic vascular complications and with the enzymatic activity and genetic polymorphisms of PON1 in these patients.

MATERIALS AND METHODS

Subjects

We recruited 155 patients with type 2 diabetes (79 men and 76 women, mean age ± SD; 64 ± 13 years) from among the outpatients and inpatients seen at our department. The diagnosis of diabetes mel-

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litus was made according to the criteria set out by the World Health Organization (WHO).[21] These patients had no ketoacidosis, renal failure (serum creatinine > 2.5 mg/dL), liver disorder, or recent history of cardiovascular disease. Subjects taking drugs known to affect lipoprotein oxidation (eg, vitamin E, vitamin C, or probucol) were excluded from the study.

Diabetic nephropathy was defined as a urine protein concentration of > 0.30 g/L at 2 or more consecutive measurements separated by an interval of 4 weeks or more. The concentration was measured by dipstick test (Uriflet II; Kyoto Daiichi Kagaku, Kyoto, Japan). A consultant ophthalmologist using direct ophthalmoscopy with dilated pupils assessed diabetic retinopathy. Patients with simple, preproliferative, or proliferative retinopathy were defined as having retinopathy. Diabetic neuropathy was defined as the presence of at least one of the symptoms of pain, tingling, burning, or loss of sensation, and at least one of the following objective signs on examination: decreased or absent patellar tendon reflex, decreased vibratory sensation, or impaired position sense. Coronary heart disease (CHD) was defined as the presence of any one of the following: (1) acute myocardial infarction or confirmed nonacute myocardial infarction based on serial readings of baseline and annual electrocardiograms; (2) coronary artery disease requiring bypass surgery or angioplasty; or (3) angina confirmed by angiography and/or by ischemic changes shown by noninvasive testing. Cerebral vascular disease (CVD) was defined as neurologic abnormalities evidenced on computed tomography or magnetic resonance imaging. Arteriosclerosis obliterans (ASO) was defined as evidence of claudication by objective findings including arteriography.

All subjects were Japanese and resided in the same area (Kochi Prefecture, Japan) and gave their informed consent to participate in the study.

Plasma-Oxidized LDL Concentration

Ox-LDL concentrations were measured using a sandwich enzyme-linked immunosorbent assay (ELISA) according to the manufacturer's instructions (Mercodia oxidized LDL ELISA; Mercodia AB, Uppsala, Sweden). In brief, diluted plasma (6,561-fold) and standards were incubated in the wells of a microtiter plate coated with murine monoclonal antioxidized LDL antibodies (mAb-4E6) for 2 hours at room temperature. After washing 6 times to remove nonreactive plasma components, a peroxidase-conjugated antiapolipoprotein B antibody was added to the wells with the Ox-LDL bound to the solid phase. After a second incubation for 1 hour at room temperature and a washing step to remove the unbound enzyme-labeled antibody, the bound conjugate was detected by a reaction with 3, 3′, 5, 5′-tetramethylbenzidine (TMB). This reaction was stopped by adding 1M H_2SO_4, and the optical density (OD) was measured at OD 450 nm using a microplate reader. The results were calculated using the computerized data reduction of absorbance for the standards versus the concentration using cubic spline regression.

Serum Paraoxonase and Arylesterase Activities

Serum was preincubated with 5 μmol/L eserine for 10 minutes at room temperature to inhibit serum butyrylcholinesterase activity. Paraoxonase activity was measured by adding serum to 1 mL glycine buffer (50 mmol/L, pH 10.5) containing 1 mmol/L $CaCl_2$ and 1 mmol/L paraoxon (Sigma, St Louis, MO). The rate of generation of *p*-nitrophenol was determined at 412 nm, 25°C, using a continuously recording spectrophotometer. Arylesterase activity was measured by adding serum to 1 mL Tris-HCl buffer (10 mmol/L, pH 8.0) containing 1 mmol/L $CaCl_2$ and 1 mmol/L phenylacetate (Sigma). The rate of hydrolysis of phenylacetate was determined spectrophotometrically at 270 nm, 25°C.

PON1 Gene Polymorphisms

Genomic DNA was extracted from whole blood using a commercial kit (SMI test; Sumitomo, Tokyo, Japan). The PON1 coding region polymorphisms, 55L/M and 192Q/R, were analyzed using a polymerase chain reaction (PCR) and digestion of the amplified fragments with restriction enzymes as described previously.[9] The PON1 promoter polymorphism −108C/T was determined using a cycle sequencing method. The DNA fragments were amplified by a PCR method using a sense primer (5′-TGGACTAGGCACCTATTCTC-3′) and an antisense primer (5′-GACTGGTGGTTCCTGAAGAG-3′). A PCR fragment separated by electrophoresis on agarose gel was recovered and purified using a commercially available kit (QIAquick PCR Gel Extraction Kit; QIAGEN, Hilden, Germany). The sequence of the PCR fragment was detected using a commercial kit and analyzer (BigDye Terminator Cycle Sequencing FS Ready Reaction Kit and ABI PRISMTM310 Genetic Analyzer; PE Applied Biosystems, Foster City, CA), and the same sense or antisense primer used in the PCR was used as the sequencing primer.

The plasma concentrations of the total cholesterol and triglycerides were measured enzymatically using an autoanalyzer. The plasma HDL-cholesterol (HDL-C) concentration was determined using a kit based on the dextran sulfate, phosphotungstate, and magnesium precipitation method. The LDL-cholesterol (LDL-C) concentration was calculated using the Friedewald equation (LDL-C = total cholesterol − HDL-C − triglyceride/5). The apoA-I, apoA-II, and apoB concentrations were measured by turbidometric immunoassays (ApoA-I Auto2, ApoA-II Auto2, ApoB Auto2; Daiichi Pure Chemicals, Tokyo, Japan).

Statistical Analysis

All data are presented as the mean ± SD. A comparison of variables between 2 groups or among 3 groups was performed using Student's unpaired *t* test or 1-way analysis of variance (ANOVA), respectively. Single linear univariate correlations were evaluated by Pearson's correlation coefficient. Stepwise regression analysis was conducted using a stepwise method and a software program on a personal computer (Statview, version 4.5; Abacus Concepts, Berkeley, CA). Statistical significance was defined as $P < .05$.

RESULTS

Clinical Characteristics of Study Subjects

The clinical characteristics and prevalence of vascular complications of the diabetic patients in this study are shown in Table 1. Fifty-one male and 14 female patients were current smokers. Sixty-two patients were receiving treatment with insulin, 62 with sulfonylureas, 2 with thiazolidine derivates, 4 with biguanides, and 25 with diet alone.

Plasma-Oxidized LDL Concentrations

The Ox-LDL level in the diabetic patients was 51.9 ± 16.0 U/L (mean ± SD) and was not associated with age or gender. Ox-LDL concentrations positively correlated with total cholesterol and triglyceride concentrations (Fig 1), and negatively with HDL-C concentration ($r = -.198$, $P < .02$). Strong correlations were detected between Ox-LDL concentrations and LDL-C or apoB concentrations (Fig 2).

Plasma-Oxidized LDL Concentrations and Macrovascular Diseases

As shown in Table 2, no difference in Ox-LDL concentration was observed between diabetic patients with and without ma-

Table 1. Clinical Characteristics of Patients With Type 2 Diabetes

Variable	Mean ± SD
Gender (male/female)	79/76
Age (yr)	64 ± 13
BMI (kg/m^2)	24.7 ± 5.3
Smoking (n; male/female)	51/14
HbA_{1c} (%)	7.2 ± 1.3
Total cholesterol (mg/dL)	186 ± 39
Triglyceride (mg/dL)	129 ± 73
HDL-cholesterol (mg/dL)	50 ± 15
Therapy (n)	
Diet alone	25
Oral hypoglycemic agent	68
Insulin	62
Statin use (n)	47
Hypertension (n)	94
Retinopathy (n)	84
Persistent proteinuria (n)	32
Peripheral neuropathy (n)	76
Coronary heart disease (n)	34
Cerebral vascular disease (n)	22
Arteriosclerosis obliterans (n)	8

NOTE. Data are presented in number or mean ± SD. Hypertension: diastolic blood pressure ≥ 90 mm Hg, systolic blood pressure ≥ 140 mm Hg, and/or use of antihypertensive medication. Five patients had both coronary heart disease and cerebral vascular disease.

crovascular diseases. However, Ox-LDL concentrations corrected by LDL-C (OxLDL/LDL-C) or apoB concentrations (Ox-LDL/apoB), which probably reflect the proportion of oxidatively modified LDL to total LDL particles, were significantly higher in patients with macrovascular diseases than in those without. The Ox-LDL/apoB ratio, but not uncorrected Ox-LDL or Ox-LDL/LDL-C ratio, was also significantly higher in patients with CHD than in those without this complication (Table 2).

Plasma-Oxidized LDL Concentrations and Diabetic Microangiopathies

Table 3 shows the relationship between Ox-LDL concentration or Ox-LDL/apoB ratio and diabetic microangiopathies. No differences in the Ox-LDL concentration or Ox-LDL/apoB ratio were detected between patients with and without retinopathy. The Ox-LDL/apoB, but not Ox-LDL concentration, tended to be higher in patients with nephropathy than in those without. Both the Ox-LDL concentration and Ox-LDL/apoB ratio were significantly higher in patients with peripheral neuropathy than in those without, although the difference in the Ox-LDL/apoB ratio was more evident than in the Ox-LDL concentration.

Because some patients had 2 or more complications, we performed stepwise multiple regression analysis using Ox-LDL/apoB as an independent variable and the presence or absence of retinopathy, nephropathy, peripheral neuropathy, and macrovascular disease as dependent variables. In this model, the existence of peripheral neuropathy ($\beta = 0.217$, $F = 7.673$) was identified as a significant contributor (total $R^2 = .108$, $P < .001$), as was macrovascular disease ($\beta = 0.204$, $F = 6.734$).

Plasma-Oxidized LDL Concentrations and PON1

Serum paraoxonase activity in patients was 251.3 ± 88.9 μmol · min^{-1} · L^{-1} (mean ± SD), and arylesterase activity was 60.7 ± 16.0 mmol · min^{-1} · L^{-1}. Paraoxonase activity in patients with macrovascular diseases tended to be lower than in those without such complications, but the difference did not reach statistical significance (238.1 ± 80.4 *v* 259.5 ± 93.2 μmol · min^{-1} · L^{-1}, respectively). Arylesterase activity in patients with macrovascular diseases was significantly lower than in those without (56.0 ± 16.9 *v* 63.6 ± 14.7 mmol · min^{-1} · L^{-1}, respectively, $P < .005$).

The frequency of the L and M alleles of the 55L/M polymorphism was 0.91 and 0.09, respectively, of the Q and R

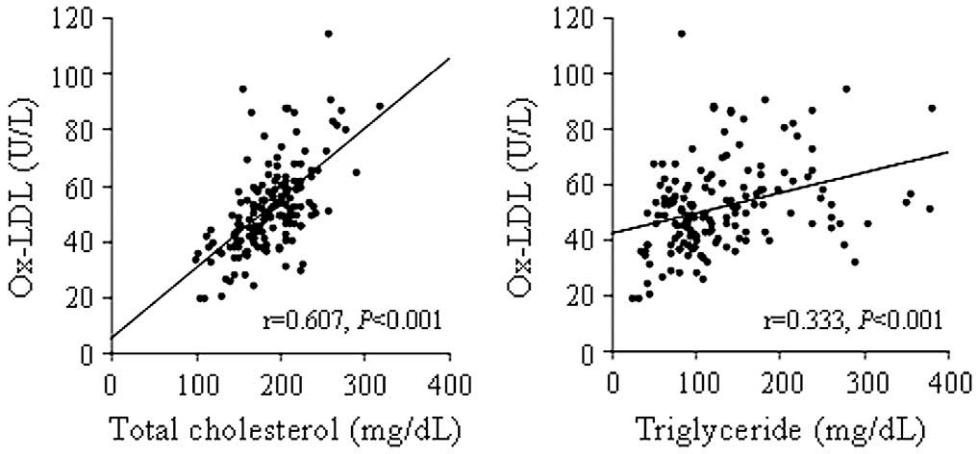

Fig 1. Correlations of Ox-LDL concentrations to total cholesterol and triglyceride concentrations in type 2 diabetic patients. Single linear univariate correlations were evaluated by Pearson's correlation coefficient.

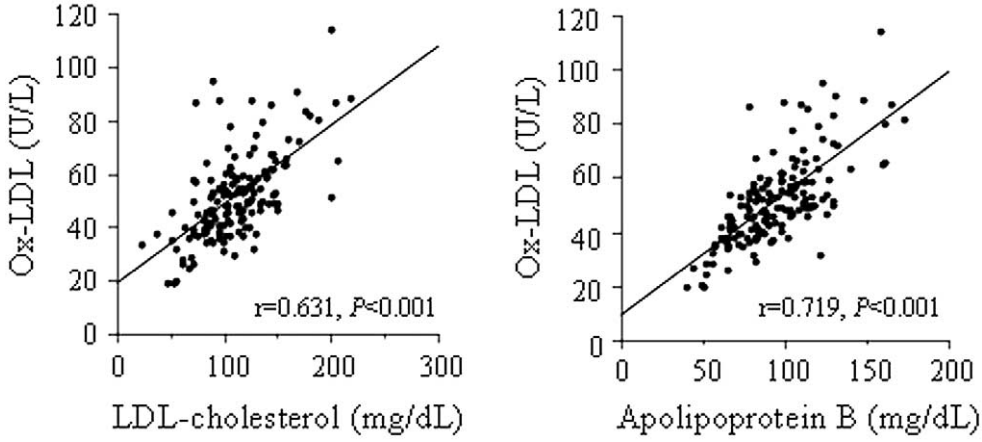

Fig 2. Correlations of Ox-LDL concentrations to LDL-C and apoB concentrations in type 2 diabetic patients. Single linear univariate correlations were evaluated by Pearson's correlation coefficient.

alleles of the 192Q/R polymorphism, 0.34 and 0.66, respectively, and of the C and T alleles of the −108C/T polymorphism, 0.56 and 0.44, respectively. Table 4 shows the association of the PON1 polymorphisms with the Ox-LDL concentration and Ox-LDL/apoB ratio. No association was observed between the 55L/M or 192Q/R polymorphism and the Ox-LDL/apoB ratio. However, the genotype TT of the −108C/T polymorphism showed a significantly higher Ox-LDL/apoB ratio compared with other genotypes with the C allele, which are associated with increased PON1 concentrations (TT: 0.60 ± 0.15, CT + CC: 0.55 ± 0.11, $P = .02$).

Table 5 shows a model of the stepwise multiple regression analysis for Ox-LDL. The −108C/T polymorphism of PON1 was identified as a significant contributor, subsequent to the apoB concentration.

DISCUSSION

In this study, Ox-LDL concentrations measured using sandwich ELISA (Mercodia oxidized-LDL ELISA) were closely correlated with LDL components, such as LDL-C and apoB concentrations. However, these correlations contradicted the findings of a report by Holvoet et al.[22] They used the same antioxidized LDL antibodies (mAb-4E6), but measured Ox-LDL concentration using a competitive ELISA, therefore, a methodologic difference may have caused the discrepancy. The oxidization of lipoproteins is generally considered to occur in the vessel wall.[3,4] However, the strong association observed between Ox-LDL concentrations and circulating LDL mass suggests that, in the Ox-LDL detected using this ELISA, it is not only back-diffusion that originates from the vessel wall; a reflection of lipoproteins modified in the blood may be reflected. In this study, plasma Ox-LDL concentrations did not differ between diabetic patients with and without macrovascular diseases. On the other hand, the Ox-LDL/apoB ratio, which may reflect the proportion of oxidatively modified LDL to total LDL particles, was significantly higher in patients with macrovascular diseases. Because the oxidative modification of LDL plays a central role in the initiation and acceleration of atherosclerosis, absolute Ox-LDL mass is considered a more important risk factor for the development of vascular diseases than the Ox-LDL/apoB ratio, at least, in a prospective study. However, in a cross-sectional study, a low level of Ox-LDL concentration does not always indicate a slight degree of atherosclerosis, because interventions for serum LDL-C level are usually more intensive in patients who already have macrovascular diseases than in those without, and the Ox-LDL level decreases along with a decrease in LDL mass as shown in Fig 2. In fact, the frequency of statin use was 42.4% in patients with macrovascular diseases, significantly higher than in patients without such complications (22.9%). Even in patients with advanced atherosclerosis, the Ox-LDL level can decrease after LDL-C lowering therapy. We therefore propose that an Ox-

Table 2. Comparison of Ox-LDL Concentration, Ox-LDL/LDL-C, and Ox-LDL/apoB Ratios Between Patients With and Without Macrovascular Diseases or CHD

	Macrovascular Diseases			CHD		
Indexes	Without (n = 96)	With (n = 59)	*P*	Without (n = 121)	With (n = 34)	*P*
Ox-LDL	51.7 ± 16.4	52.2 ± 15.5	NS	52.1 ± 15.9	51.3 ± 16.6	NS
Ox-LDL/LDL-C	0.47 ± 0.13	0.54 ± 0.21	<.01	0.49 ± 0.17	0.53 ± 0.18	NS
Ox-LDL/apoB	0.54 ± 0.11	0.60 ± 0.12	<.002	0.55 ± 0.11	0.60 ± 0.15	<.05

NOTE. Macrovascular disease was defined as any one or more of the following: CHD, CVD, or ASO. Definitions of these diseases are included in Materials and Methods. Of 59 patients with macrovascular diseases, 29 had CHD, 17 had CVD, 5 had both CHD and CVD, and 8 had ASO.

Abbreviations: CHD, coronary heart disease; CVD, cerebral vascular disease; ASO, arteriosclerosis obliterans; NS, not significant.

Table 3. Comparison of Ox-LDL Concentration and Ox-LDL/apoB Ratio Between Patients With and Without Diabetic Microangiopathies

	Retinopathy		Nephropathy		Neuropathy	
Indexes	Without (n = 71)	With (n = 84)	Without (n = 123)	With (n = 32)	Without (n = 79)	With (n = 76)
Ox-LDL	50.6 ± 15.6	53.0 ± 16.4	51.2 ± 16.0	54.6 ± 16.1	49.3 ± 15.1	54.6 ± 16.6*
Ox-LDL/apoB	0.55 ± 0.13	0.57 ± 0.11	0.55 ± 0.12	0.59 ± 0.09†	0.53 ± 0.10	0.59 ± 0.12‡

NOTE. Definitions of complications are included in Materials and Methods.
*$P < .05$, †$P = .06$, ‡$P < .001$.

LDL level corrected by apoB (Ox-LDL/apoB, probably reflects the proportion of oxidatively modified LDL to total LDL particles) could be available to such patients. Patients with diabetic polyneuropathy also showed a significantly higher Ox-LDL/apoB ratio compared with those without this complication, which may support the hypothesis that increased oxidative stress in the diabetic state may be involved in the development of diabetic complications.

The Ox-LDL concentration also showed a significant positive correlation with the serum triglyceride concentration. It is known that the triglyceride concentration is inversely correlated with LDL size, and that small dense LDL particles are more susceptible to lipid oxidation than larger particles.[23,24] A significant negative correlation between LDL particle size and malondialdehyde-modified LDL (MDA-LDL) has been reported.[25] Therefore, the observed relationship between Ox-LDL concentrations and triglyceride concentrations in our population may be mediated by LDL particle size. However, we cannot rule out the possibility of cross reactivity of the Ox-LDL assay against other apoB-containing lipoproteins with oxidative modification. Further work is required to clarify this.

Both increased oxidative stress and decreased antioxidant capacity can promote lipid peroxidation. We found a negative correlation between Ox-LDL concentration and serum HDL-C. HDL has an antiatherogenic effect that occurs through the inhibition of lipid peroxidation, as well as reverse-cholesterol transport.[26-28] PON1 is associated with HDL and is believed to play a central role in the inhibitory effect of HDL on lipid peroxidation. Therefore, we next examined the gene polymorphisms and enzymatic activity of PON1. It has been reported that glycation of PON1 impairs its enzymatic activity,[29] and serum paraoxonase activity is lower in diabetic patients and is lower yet in those with diabetic complications, independent of the PON1 gene polymorphisms.[19,20] The attenuation of PON1 action could result in high susceptibility to LDL oxidation, especially in diabetic patients suffering oxidative stress.

The site at which PON1 acts against LDL oxidization has not yet been established, although the cystein in position 284 of the amino acid sequence is highly suspected as an active site for antioxidization.[30] The antioxidation site appears to differ from the enzyme-active site.[31] In this study, the hydrolysis rates for paraoxon (paraoxonase) and phenylacetate (arylesterase) were not significant contributors to Ox-LDL concentration in the stepwise regression analysis, and the 192Q/R polymorphism that influences serum paraoxonase activity was not associated with the Ox-LDL/apoB ratio. On the other hand, patients with the low expresser genotype TT of the −108C/T polymorphism revealed a higher Ox-LDL/apoB ratio than patients with the CT or CC genotype. In addition, the stepwise regression analysis revealed that the −108C/T polymorphism was a significant contributor to Ox-LDL concentration. The 55L/M polymorphism has also been associated with the PON1-expression level,[10-13] but its association appears to be through linkage disequilibrium with the −108C/T polymorphism[15,16] and is weaker than that of the −108C/T. This may result in a negative

Table 4. Association of PON1 Gene Polymorphisms With Ox-LDL Concentration and Ox-LDL/apoB Ratio

	Genotypes (n)	Ox-LDL	Ox-LDL/apoB
55L/M	LL (129)	52.8 ± 16.4	0.57 ± 0.12
	LM (23)	46.9 ± 13.3	0.53 ± 0.09
	MM (3)	53.1 ± 18.1	0.55 ± 0.05
192Q/R	QQ (18)	51.2 ± 18.4	0.53 ± 0.12
	QR (70)	50.6 ± 15.5	0.56 ± 0.11
	RR (67)	53.5 ± 16.0	0.57 ± 0.12
−108C/T	TT (30)	55.5 ± 18.3	0.60 ± 0.15*
	CT (75)	51.8 ± 15.9	0.55 ± 0.11
	CC (50)	49.9 ± 14.7	0.54 ± 0.11

*$P = .02$ *v* CT + CC genotype (n = 125, 0.55 ± 0.11).

Table 5. Stepwise Regression Analysis for Ox-LDL Concentration

Variables	β	F
Apolipoprotein B	0.721	164.796
PON1 −108C/T	−0.116	4.299
Body mass index	0.155	3.616
Hypertension	0.129	2.494
HDL-cholesterol	−0.109	1.756
Diabetes duration	0.091	1.216
PON1 55L/M	−0.088	1.155
HbA_{1c}	0.077	.874
Triglyceride	0.077	.868
Statin use	0.058	.500
Age	0.048	.341
LDL-cholesterol	0.036	.192
Gender	0.032	.149
Arylesterase activity	−0.024	.086
PON1 192Q/R	0.021	.065
Paraoxonase activity	−0.020	.057
Smoking	0.015	.033
Total R^2	.534	$P < .001$

NOTE. Stepwise multiple regression analysis was performed. The F values for the inclusion and exclusion of variables was set at 4.0 at each step. Gender: male = 1, female = 2; hypertension: absent = 1, present = 2; statin use: no = 1, yes = 2; PON1 55L/M: MM = 1, LM = 2, LL = 3; PON1 192Q/R: QQ = 1, QR = 2, RR = 3; PON1 −108C/T: TT = 1, CT = 2, CC = 3. β: Partial regression coefficient.

association between the 55L/M polymorphism and the Ox-LDL/apoB ratio. However, a larger study population is necessary to confirm such a correlation.

In summary, the Ox-LDL/apoB ratio was associated with macrovascular diseases and diabetic neuropathy in patients with type 2 diabetes. An increased Ox-LDL/apoB ratio may result from reduced serum antioxidant capacity in the diabetic state, including the attenuation of PON1 action. The Ox-LDL/apoB ratio may be a significant marker for susceptibility to vascular complications in diabetic patients.

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