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Increase in plasma Malondialdehyde-modified low-density lipoprotein in patients with atherothrombotic cerebral infarction

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Oxidized low-density lipoprotein cholesterol (Ox-LDL) is considered to play a critical role in the pathogenesis of atherosclerosis. We investigated the role of malondialdehyde-modified LDL (MDA-LDL), an indicator of Ox-LDL, in cerebral infarction. We also investigated the relationship between MDA-LDL and atherosclerotic change of the carotid artery. Subjects were 30 patients with lacunar infarction (LA), 19 patients with atherothrombotic infarction (AT) and 48 controls. Carotid arteries were evaluated with B-mode Doppler ultrasonography. The intima-media thickness (IMT) was considered to be increased if its value was more than 1.1 mm. The level of MDA-LDL concentration was significantly ($P<0.05$) elevated in AT patients (129.96±27.88 U/l) than in LA patients (99.35±34.06 U/l) and controls (97.65±32.61 U/l). Among AT patients, plasma level of MDA-LDL concentration was statistically significantly elevated in the group with increased IMT (139.7±24.5 U/l) than in the group without increased IMT (102.7±17.2 U/l). No statistically significant difference was found among LA patients. However, there was no difference in LDL-C concentration between the patients with and without IMT thickening among LA or AT patients. The concentration of MDA-LDL was significantly decreased ($P<0.05$) after statin administration for 5-6 months. MDA-LDL, namely degraded or qualitatively changed LDL-C, appears to be related to atherosclerotic change of the carotid artery in AT patients.

Key words: MDA-LDL, oxidized LDL, IMT, cerebral infarction, statin

INTRODUCTION

Ox-LDL is considered to play a critical role in the pathogenesis of arteriosclerosis [6, 11-13, 17]. Malondialdehyde-modified LDL (MDA-LDL) is measured as an indicator of ox-LDL, for which a direct assay system was recently established. MDA-LDL has been suggested to be a useful indicator for the identification of patients with coronary artery disease [2, 3, 10, 15, 16], but its role in cerebrovascular diseases is not fully understood. We examined the level of MDA-LDL in patients with lacunar infarction (LA) and atherothrombotic infarction (AT). We also investigated the relationship between MDA-LDL and atherosclerotic change of the carotid artery, using B-mode ultrasonography, in patients with cerebral infarction.

MATERIALS AND METHODS

Subjects

Informed consent was obtained before starting of this study.

We studied 30 patients with LA (male 17, female 13, age 68±11 y/o), 19 patients with AT (male 11, female 8, age 74±10 y/o), and 48 controls without any type of stroke (male 18, female 30, age 69±12 y/o). All of them had visited Tokai University Oiso Hospital from 2001 to 2002.

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Evaluation of Carotid Artery

IMT of the common carotid artery was examined with B-mode Doppler ultrasonography (GE Yokokawa, LOGIQ700, linear 7.5-MHz transducer), and the measurement was taken at the most thickened part of the distal wall. IMT of the carotid artery increases with aging, but never rises above 1.0 mm in normal Japanese adults. The presence of increased IMT (thickening) in the carotid artery has been defined as a thickness of more than 1.1 mm [7].

Measurement of MDA-LDL

MDA-LDL was measured with the enzyme-linked immunosorbent assay (ELISA) developed by Kotani *et al.* [8, 9]. Briefly, the monoclonal antibody against MDA-LDL, ML25, recognizes MDA-LDL as well as MDA-modified proteins in a solid-phase competitive enzyme immunoassay. Therefore, to detect MDA-LDL specifically, ML25 was combined with apoB-specific antibody (AB16) as the second antibody. In the sera of healthy individuals the concentration of MDA-LDL ranged from 20-80 units/l when the absorbance obtained with artificially prepared MDA-LDL at the concentration of 1 mg/l was defined as 1 unit/l.

Statin treatment

10 mg statin (pravastatin sodium)/day was administrated. Durations of administration were 5.1 ± 2.0 months for LA patients and 6.6 ± 3.1 months for AT patients.

RESULTS

Concerning the concentration of LDL-C, there was no statistically significant difference among the LA patients (121.4±24.7 U/l), AT patients (134.1±25.6 U/l) and controls (128.6±26.6 U/l) (Fig. 1). However, the concentration of MDA-LDL was significantly ($P<0.05$) elevated in AT patients (130.0± 27.9 U/l) than in LA patients (99.4±34.1 U/l) and controls (97.7±32.6 U/l) (Fig. 2).

Among LA and AT patients, there was no difference in LDL-C concentration (Fig. 3) or MDA-LDL concentration (Fig. 4) between those with and without increased IMT. Among AT patients, MDA-LDL was statistically significantly elevated in the group with IMT thickening (139.7±24.5 U/l) versus the group without IMT thickening (102.7± 17.2 U/l) (Fig. 5). No statistically significant difference was found among LA patients (101.8 ± 38.9 U/l vs. 96.2 ± 27.7 U/l) (Fig. 6).

Plasma level of MDA-LDL concentration was statistically significantly decreased ($P<0.05$) after statin (pravastatin sodium) administration. Before administration of statin, the concentrations of MDA-LDL were 98.5±40.4 U/l in LA patients and 141.9± 37.2 U/l in AT patients. After administration of statin they were decreased to 80.9±30.1 U/l and to 112.1±27.1 U/l, respectively (Figs. 7 and 8).

DISCUSSION

MDA-LDL is considered to play a critical role in the pathogenesis of atherosclerosis. The relationship between MDA-LDL and atherosclerotic disease has mainly been studied in relation to coronary artery disease, and little work has been done in the case cerebral infarction. Oxidized LDL is known to be present in atherosclerotic lesions of rabbit and man [18]. Holvoet *et al.* reported that uptake of MDA-LDL by macrophages resulted in foam cell generation, and that an elevated plasma level of MDA-LDL is associated with plaque instability [4, 5]. They concluded that oxidized LDL is a useful marker for identifying patients with coronary artery disease [3]. Our results also show that MDA-LDL is more elevated in AT patients than in the LA patients. However, in neither case was there any relationship between increased IMT of the carotid artery and the concentration of LDL-C or MDA-LDL. For patients with AT infarction only, the concentration of MDA-LDL was elevated in patients with increased IMT. This may suggest that AT has a stronger relationship to atherosclerosis than does LA, and MDA-LDL might be available as a marker for atherosclerosis of the carotid artery in patients with AT infarction.

Statin decreased the concentration of MDA-LDL. Tamura *et al.* reported that in hypercholesterolemic patients, 8-week treatment with atorvastatin or pravastatin produced significant reductions in LDL-C and MDA-LDL concentrations, with a significant increase of HDL-C concentration [14]. Bujoh *et al.* reported that in patients with hyperlipidemia who are taking statin, IMT and the concentration of plasma MDA-LDL were correlated ($r=0.41$, $P<0.01$), so the concentration of MDA-LDL may be related to the progression of thickening of the carotid

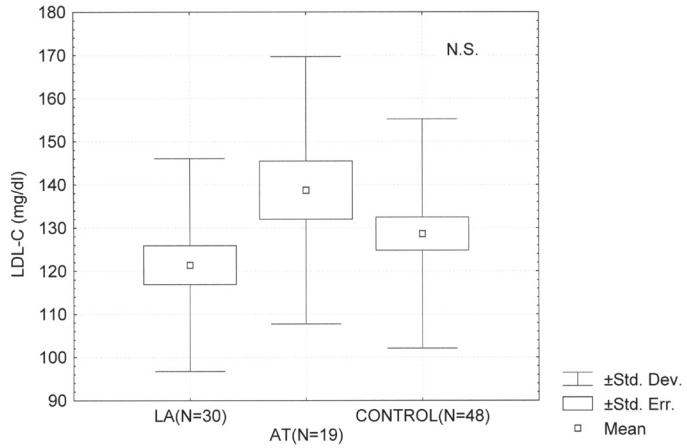

Fig. 1 Plasma LDL-C level in the 3 groups of LA (lacunar infarction), AT (atherothrombotic infarction) and control

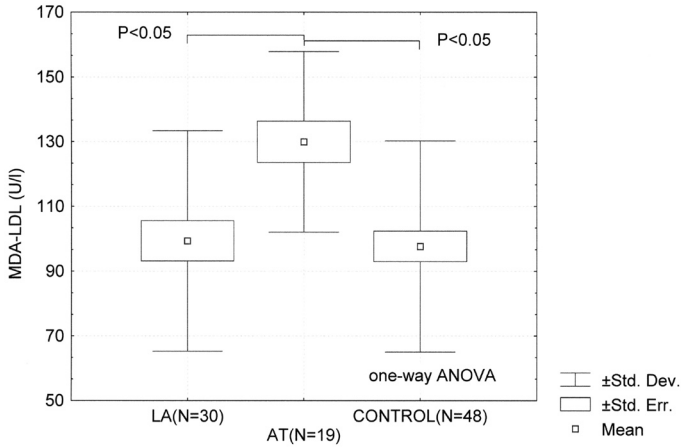

Fig. 2 MDA-LDL level in the 3 groups of LA (lacunar infarction), AT (atherothrombotic infarction) and control

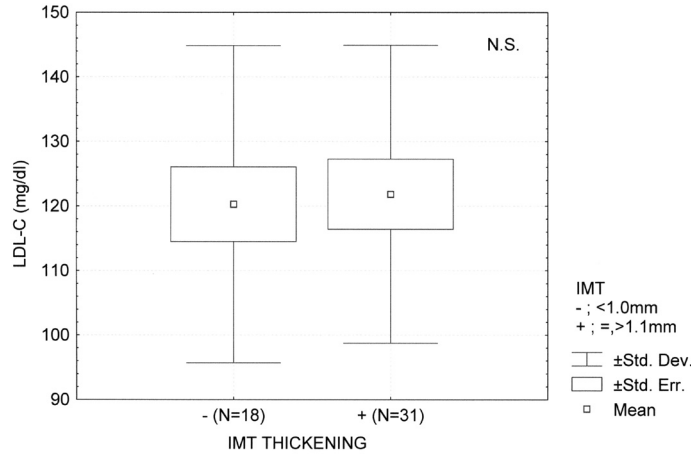

Fig. 3 Correlation between IMT and LDL-C (lacunar infarction and atherothrombotic infarction)

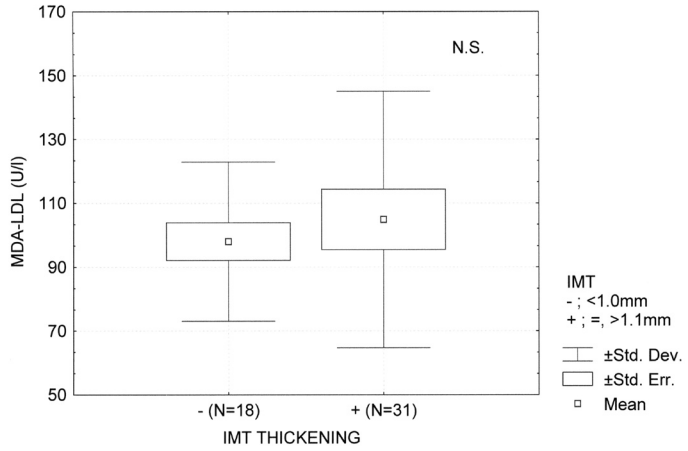

Fig. 4 Correlation between IMT and MDA-LDL (lacunar infarction and atherothrombotic infarction)

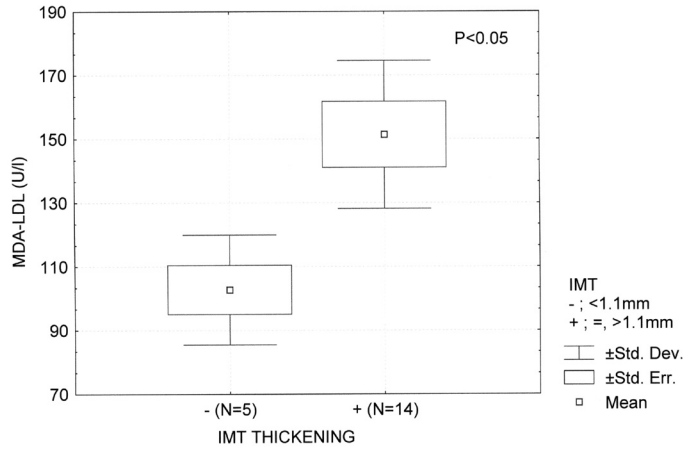

Fig. 5 Correlation between IMT and MDA-LDL (atherothrombotic infarction)

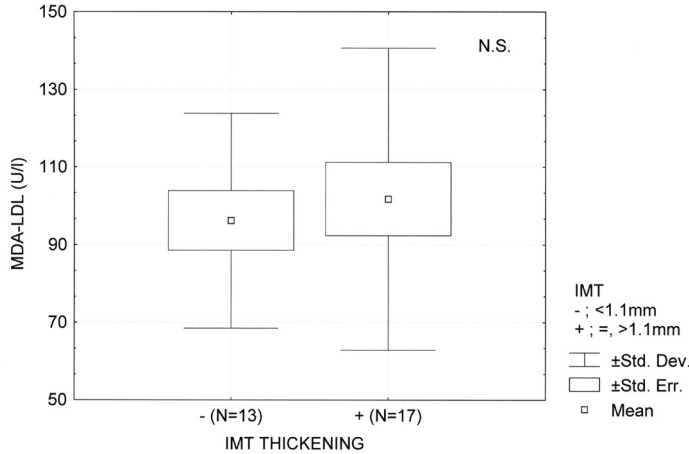

Fig. 6 Correlation between IMT and MDA-LDL (lacunar infarction)

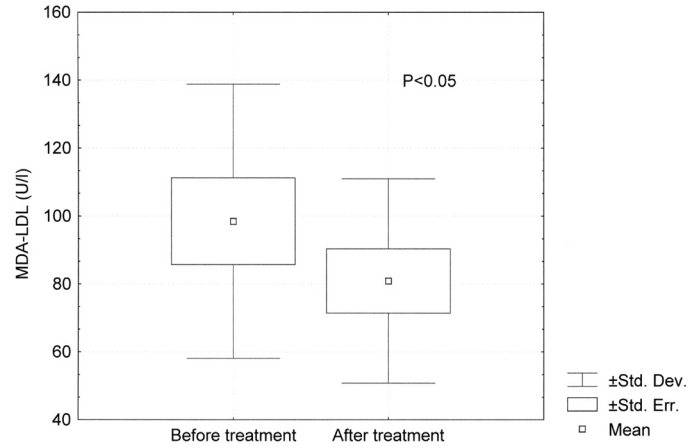

Fig. 7 Plasma level of MDA-LDL after statin treatment (lacunar infarction, N=10)

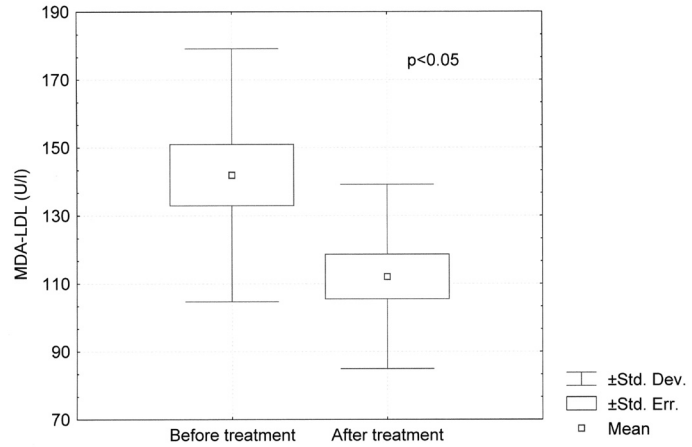

Fig. 8 Plasma level of MDA-LDL after statin treatment (atherothrombotic infarction, N=17)

artery wall [1]. MDA-LDL, namely degraded or qualitatively changed LDL-C, thus appears to be related to atherosclerotic change of the carotid artery in AT patients. In this study, the duration of statin treatment was just 5-6 months, and in order to evaluate fully the relationship between statin treatment and MDA-LDL concentration, further studies with larger populations and longer administration times will be needed.

REFERENCES

1) Bujoh H, Kotani K, Saitoh K *et al*. MDA-LDL blood levels and progression of carotid hypertrophy during therapy with statins. J. Japan Atherosclerosis Society 29 suppl: 268, 2001.
2) Ehara S, Ueda M, Naruto T, *et al*. Elevated levels of oxidized low density lipoprotein show a positive relationship with the severity of acute coronary syndromes. Circulation 103: 1955-1960, 2001.
3) Holvoet P, Mertens A, Verhamme P, *et al*. Circulating oxidized LDL is a useful marker for identifying patients with coronary artery disease. Arterioscler Thromb Vasc Biol 21: 844-848, 2001.
4) Holvoet P, Perez G, Zhao Z *et al*. Malondialdehyde-modified low density lipoproteins in patients with atherosclerotic disease. J Clin Invest 95: 2611-2619, 1995.
5) Holvoet P, Vanhaecke J, Janssens S *et al*. Oxidized LDL and malondialdehyde-modified LDL in patients with acute coronary syndromes and stable coronary artery disease. Circulation 98: 1487-1494, 1998.
6) Jessup W, Kritharides L. Metabolism of oxidized LDL by macrophages. Curr Opin Lipidol 11: 473-481, 2000.
7) Joint committee with the Guidelines Subcommittee of the Japan Academy of Neurosonology for

Ultrasonic Assessment of Carotid Artery Disease and the Subcommittee for Research into Methods of Screening Atherosclerotic Lesions. Guidelines for ultrasonic assessment of carotid artery disease: Preliminary report. Neurosonology 15: 20-33, 2002.

8) Kotani K, Kondo A, Manabe M, *et al*. Determination of malondialdehyde-modified LDL (MDA-LDL) and its potential usefulness. Jpn J Clin Pathol 45: 47-54, 1997.
9) Kotani K, Maekawa M, Kanno T, *et al*. Distribution of immunoreactive malondialdehyde-modified low-density lipoprotein in human serum. Biochim Biophys Acta 1215: 121-125, 1994.
10) Miyazaki T, Shimada K, Sato O, *et al*. Circulating malondialdehyde-modified LDL and atherogenic lipoprotein profiles measured by nuclear magnetic resonance spectroscopy in patients with coronary artery disease. Atherosclerosis 179: 139-145, 2005.
11) Ross R. Atherosclerosis is an inflammatory disease. Am Heart J 138: S419-20, 1999.
12) Steinberg D, Parthasarathy S, Carew TE, Khoo JC, Witztum JL. Beyond cholesterol. Modification of low-density lipoprotein that increase its atherogenicity. N Engl J Med 320: 915-24, 1989.
13) Steinberg D. Role of oxidized LDL and antioxidants in atherosclerosis. Adv Exp Med Biol 369: 39-48, 1995.
14) Tamura A, Watanabe T, Nasu M. Effects of atorvaststin and pravastatin on malondialdehyde-modified LDL in hypercholesterolemic patients. Circulation Journal 10: 816-820, 2003.
15) Tanaga K, Bujo H, Inoue M, *et al*. Increased circulating malondialdehyde-modified LDL levels in patients with coronary artery disease and their association with peak sizes of LDL particles. Arterioscler Thromb Vasc Biol 22: 662-6, 2002.
16) Toshima S, Hasegawa A, Kurabayashi M, *et al*. Circulating oxidized low density lipoprotein levels: a biochemical risk marker for coronary heart disease. Arterioscler Thromb Vasc Biol 20: 2243-2247, 2000.
17) Witztum JL. Immunological response to oxidized LDL. Atherosclerosis 131(suppl): S9-S11, 1997.
18) Ylä-Herttuala S, Palinski W, Rothenfeld ME, *et al*. Evidence for the presence of oxidatively modified low density lipoprotein in atherosclerotic lesions of rabbit and man. J Clin Invest 84: 1086-1095, 1989.