Oxidized LDL in Carotid Plaques and Plasma Associates With Plaque Instability

Kyoko Nishi, Hiroyuki Itabe, Masaaki Uno, Keiko T. Kitazato, Hidehisa Horiguchi, Kiyohito Shinno, Shinji Nagahiro

Objective—Oxidation of LDL plays a significant pathogenic role in atherosclerosis. In this study, we attempted to clarify the correlation between the morphology of human atherosclerotic plaques and the oxidized LDL (OxLDL) levels in plasma and carotid plaques.

Methods and Results—OxLDL levels (ng/µg apolipoprotein B) in plasma and carotid plaques from 44 patients undergoing carotid endarterectomy and OxLDL levels in 17 control plasma and 9 normal intima samples were determined by a sandwich ELISA by using specific antibodies against OxLDL (DLH3) and apolipoprotein B. The plaques were immunohistochemically classified as macrophage (Mφ)-rich and Mφ-poor. In paired samples from individual patients, plaque OxLDL was nearly 70 times higher than plasma OxLDL (mean±SEM, 11.9±1.7 vs 0.18±0.01 ng/µg apoB, P<0.0001). The OxLDL level was significantly higher in Mφ-rich- than Mφ-poor plaques (19.6±2.8 vs 5.50±0.77ng/µg apoB, P<0.0001) and corresponded with DLH3 antigen positivity of the plaques. In patients with Mφ-rich plaques, plasma OxLDL was significantly higher than in the controls (0.20±0.02 vs 0.13±0.01ng/µg apoB, P=0.02).

Conclusions—Our results suggest that LDL undergoes further oxidation in plaques, and that high plasma and plaque levels of OxLDL are correlated with the vulnerability to rupture of atherosclerotic lesions. (Arterioscler Thromb Vasc Biol. 2002;22:1649-1654.)

Key Words: oxidized LDL ■ plaque ■ plasma ■ carotid endarterectomy ■ histopathology

The oxidation of LDL plays a significant pathogenic role in atherosclerosis.1–4 Oxidized LDL (OxLDL) exhibits a number of specific biological properties in vitro and in vivo, including foam cell formation from macrophages. LDL oxidation is a complex process, and a number of oxidation products derived from lipids are generated through peroxidation and fragmentation. Apolipoprotein B (apoB) is exposed to oxidative degradation and modification by oxidized lipids; these processes result in substantially modified lipid and protein components. However, because OxLDL consists of a group of heterogeneously modified particles, it has been difficult to examine OxLDL epitopes in vivo. Itabe et al5 reported a sensitive method to quantify OxLDL using a specific antibody to OxLDL. Oxidized phosphatidylcholine (OxPC) molecules that form adducts with apoB in OxLDL are recognized by the anti-OxLDL monoclonal antibody (mAb), DLH3.6 Using this method, Toshima et al7 and Ehara et al8,9 demonstrated significant increases in the plasma OxLDL levels of patients with coronary heart disease. In addition, previous clinical and experimental studies detected oxidative epitopes in the arterial wall10–12; however, to our knowledge, no quantitative analysis of OxLDL in human atherosclerotic lesions has been reported to date.

To clarify how much OxLDL is present and how it is modified in plaques compared with plasma, we assayed the amount of OxLDL in plasma and carotid plaques from patients scheduled for carotid endarterectomy (CEA). We then investigated the relationship between plaque and plasma OxLDL levels and plaque morphology, neurological symptoms, risk factors, and plasma lipid parameters. In our previous study,13 we demonstrated that plaques in which more than 5% of the total area is occupied by macrophages manifested significant elevation of thiobarbituric acid-reactive substances, that they were prone to rupture or fibrous cap thinning, and that they had a large lipid core (≥10% of total area). Based on these observations, we considered macrophage infiltration a landmark for plaque instability and classified carotid plaques as macrophage (Mφ)-rich and (Mφ)-poor. We hypothesized that OxLDL level and the degree of macrophage infiltration play a role in whether atherosclerotic lesions are stable or unstable.

Methods

Subjects
Informed consent was obtained from all study participants. Carotid plaque and plasma samples from 44 patients who underwent CEA
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heart disease or arteriosclerosis obliterans). Control plasma was dentedly on medical examination for other complaints (ischemic patients with carotid bruit, and patients who were diagnosed inci-
confirmed by magnetic resonance angiography or ultrasonography,
cluded patients with progression of ipsilateral carotid stenosis /H11005 more than one month after onset. The asymptomatic group (n =

ischemic attack, amaurosis fugax, or hemispheric stroke) within 3

Sixteen patients with hyperlipidemia had received lipid-lowering
summarizes demographic and clinical data on the study participants.

Yl-Herttuala et al.11 Briefly, plaques were cut into small pieces,
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Plaque LDL was extracted by the slightly modified method of
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dialyzed at 4
°d

mean of homogenate buffer. The tubes were then centrifuged at

overnight at 4
°

Fitzgerald), and the anti-OxPC mAb DLH3. Serial sections were
acted to quantitative analysis of OxLDL. To determine plasma
jected to quantitative analysis of OxLDL. To determine plasma

between June 8, 1999, and December 12, 2000, were examined. All
CEAs were performed according to established criteria.14–15 Table 1
summarizes demographic and clinical data on the study participants.
Sixteen patients with hyperlipidemia had received lipid-lowering
therapy. The side ipsilateral to the operated side was involved in all
patients (n = 19) who manifested ischemic symptoms (transient ischemic attack, amaurosis fugax, or hemispheric stroke) within 3
months of onset. All operations were performed at the chronic stage, more than one month after onset. The asymptomatic group (n = 25)
included patients with progression of ipsilateral carotid stenosis confirmed by magnetic resonance angiography or ultrasonography,
patients with carotid bruit, and patients who were diagnosed inci-

dently on medical examination for other complaints (ischemic heart disease or arteriosclerosis obliterans). Control plasma was
obtained from 17 age-matched normal volunteers, they were 9 men
and 8 women aged 49 to 74 years (mean ± SD, 63.3 ± 8.2) without a
history of ischemic vascular disease or risk factors. In addition, 9
carotid arteries were removed at autopsy manifested no macroscopic
evidence of atherosclerosis. The intima was dissected carefully for examination. CEA specimens were removed en bloc. Immediately after removal, the plaques were examined macroscopically.
The thickest part of each plaque was cut transversely to define histomorphological characteristics. The remainder of each plaque was
subjected to quantitative analysis of OxLDL. To determine plasma

XLDL, venous blood was drawn into a test tube containing

EDTA-Na on the morning of the operation from patients who had
been fasting for at least 12 hours. The plasma was separated, stored
at 4°C, and used within 6 days. Total cholesterol, triglycerides, HDL,
and LDL-cholesterol were determined in patients as well as in

As amounts of OxLDL per microgram of apoB protein.


either the plaque OxLDL levels or the patients

data.

Histopathological Characteristics of Plaques
Fresh plaque specimens, some of which required decalcification,
were promptly fixed in methanol-Carnoy’s fluid (Ishizu) and em-
bedded in paraffin, and 4-μm-thick sections were cut. Histopatho-
logical characteristics were examined after staining with hematoxy-
lin and eosin and elastica van Gieson. Adjacent serial sections were
stained immunohistochemically. An experienced pathologist ana-
yzed the histological features. The level of M6 infiltration and the
lipid core size were measured by using an image analysis program
(Mac SCOPE). Intraplaque inflammation plays a key role in plaque
destabilization.17 Based on the extent of M6 infiltration, the plaques
were classified as M6-poor (infiltration: <5% of total area, n = 24)
and M6-rich (infiltration: ≥5% of total area, n = 20). All results were
cross-checked to investigate possible relationships with diverse
histopathological and clinical findings (eg, the grade of angioGraphi-
cal stenosis [%], ultrasound echogenicity). The pathologist was not
cognizant of either the plaque OxLDL levels or the patients’ clinical
data.

Immunohistochemical Analysis
Antibodies used for immunohistochemical study were anti-macro-
phage mAb (kp-1, Dako; HAM-56, Dako), anti-α-smooth muscle actin mAb (Sigma), anti-αPOB polyclonal antibody (anti-αPOB;
Fitzgerald), and the anti-OxPC mAb DLH3. Serial sections were
deparaffinized in xylene and dehydrated in a graded series of ethanol and
immunostained by using LSAB kit (Dako). They were treated
overnight at 4°C with each antibody and anti-mouse IgG1 mAb
(Dako) as a negative control. Some sections were treated with proteinase K (Dako) for 9 minutes at room temperature. Nuclei in all
sections were stained with methylgreen.

Determination of OxLDL Levels
By using the method described previously,3 microtiter wells
precoated with DLH3 (5 μg/mL in PBS, 100 μL/well) were
blocked with 1% BSA in 0.05 mol/L Tris-buffered saline, pH 8.0.
In each assay, we added to the wells 100 μL of appropriately
diluted plasma and plaque LDL fractions (plasma LDL, 2 to 10
μg protein/well; plaque LDL, 0.5 to 5 μg protein/well), and 0.2
to 8.0 ng/well of standard OxLDL. The plates were incubated
overnight at 4°C. After an overnight incubation, OxLDL was
detected with 100 μL of sheep anti-human apoB IgG antibody
(Boehringer) and 100 μL of alkaline phosphatase-conjugated
donkey anti-sheep IgG antibody (Chemicon). The reactivity of
alkaline phosphatase was measured at 405 nm after incubation for
an appropriate length of time at 37°C with 100 μL substrate
solution containing 1 mg/mL of p-nitrophenylphosphate hexahy-
drate (Wako). OxLDL per microgram of protein was calculated
from the standard curve. Data from samples in the saturation
range were discarded. Simultaneously, a parallel set of ELISA
was run to determine the amounts of apoB in the same lipoprotein
fractions by using anti-apoB mAb (OEM Concepts). For apoB
measurement, 0.5 to 15 ng/well native LDL was added to each
ELISA plate as the standard. The OxLDL levels were expressed
as amounts of OxLDL per microgram of apoB protein.

Immunoblot Analysis
The lipoprotein fractions from carotid plaques and plasma LDL
fractions were submitted to immunoblot analysis. Aliquots (1 μg
protein/lane) were subjected to 12% SDS-PAGE (Bio-Rad, Japan)
and electrotransferred to polyvinylidene difluoride membranes.
We also used silver-stain SDS-PAGE. The membranes were
blotted with anti-apoB polyclonal antibody (Binding Site Inc) and
visualized with an HRP-conjugated second antibody (Chemicon)
and an enhanced chemiluminescence reagent (Amersham).


table 1. Patient Characteristics

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>n</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>38–80</td>
<td></td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>66.3 ± 7.7</td>
<td></td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>35</td>
<td>79.5</td>
</tr>
<tr>
<td>Female</td>
<td>9</td>
<td>20.5</td>
</tr>
<tr>
<td>Symptoms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Symptomatic</td>
<td>19</td>
<td>43.2</td>
</tr>
<tr>
<td>Asymptomatic</td>
<td>25</td>
<td>56.8</td>
</tr>
<tr>
<td>Risk factors</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertension</td>
<td>36</td>
<td>81.8</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>16</td>
<td>36.4</td>
</tr>
<tr>
<td>Smoking</td>
<td>21</td>
<td>47.7</td>
</tr>
<tr>
<td>Hyperlipidemia</td>
<td>16</td>
<td>36.4</td>
</tr>
<tr>
<td>Ischemic heart disease</td>
<td>16</td>
<td>36.4</td>
</tr>
</tbody>
</table>
times lower than in LDL fractions from plaques (P<0.001), and the concentration of apoB in the LDL fraction recovered from normal intima was less than 40% of that of plaques. The OxLDL content in LDL fractions from normal intima was about one-tenth of that in LDL fractions from plaques when normalized by amounts of total protein (0.06±0.02 vs 0.84±0.13 ng/μg protein, P<0.0001). ApoB-containing lipoproteins in plaques correlated with plaque OxLDL levels (n=53, r=0.555, P<0.0001). These findings suggest that plaque LDL is enriched with oxidative modification.

Comparison of OxLDL Levels and Plaque Morphology
There was a strong correlation between the plaque OxLDL level and the extent of macrophage infiltration (r=0.63, P<0.0001, Figure 1). The characteristics of the Mφ-rich and Mφ-poor carotid atherosclerotic lesions are summarized in Table 2. The Mφ-rich plaques were associated with plaque rupture, fibrous cap thinning, and a large-sized lipid core and correlated with ultrasonographic echolucency but not with angiographical carotid stenosis. The plasma OxLDL level in patients with Mφ-rich plaques was significantly higher than in control plasma (0.20±0.02 vs 0.13±0.01 ng/μg apoB, P=0.02) (Figure 2A) and slightly higher than in patients with Mφ-poor plaques (0.16±0.01 ng/μg apoB); the difference in the plasma OxLDL level between patients with Mφ-rich- and Mφ-poor plaques was not significant. The OxLDL level in Mφ-rich plaques was 3.6 times higher than in Mφ-poor plaques (19.6±2.8 vs 5.50±0.77 ng/μg apoB, P<0.0001) (Figure 2B). There was no significant difference between the OxLDL level in normal intima and Mφ-poor plaques and between the plasma OxLDL level in the patients with Mφ-poor plaque and the controls.

ApoB Oxidation Analysis by Western Blot
As shown in Figure 3, Western blot analysis of LDL fractions from control and patient plasma and from carotid plaques

**TABLE 2. Characteristics of the Plaques**

<table>
<thead>
<tr>
<th>Histopathological findings</th>
<th>Macrophage-Rich (n=20)</th>
<th>Macrophage-Poor (n=24)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macrophage infiltration, % range</td>
<td>5.0–15.3</td>
<td>0.4–4.7</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Mean±SEM</td>
<td>7.8±0.6</td>
<td>2.6±0.2</td>
<td></td>
</tr>
<tr>
<td>Lipid core, % range</td>
<td>14.0–52.2</td>
<td>0.1–21.5</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Mean±SEM</td>
<td>28.4±2.7</td>
<td>7.2±1.4</td>
<td></td>
</tr>
<tr>
<td>Plaque rupture, n (%)</td>
<td>12 (60.0)</td>
<td>1 (4.2)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Fibrous cap thinning, n (%)</td>
<td>18 (90.0)</td>
<td>1 (4.2)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Intraplaque hemorrhage, n (%)</td>
<td>15 (75.0)</td>
<td>9 (37.5)</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Calcification, n (%)</td>
<td>5 (25.0)</td>
<td>11 (45.8)</td>
<td>NS</td>
</tr>
<tr>
<td>Ulcer, n (%)</td>
<td>17 (85.0)</td>
<td>14 (58.3)</td>
<td>NS</td>
</tr>
<tr>
<td>Clinical findings</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Echogenicity: echolucent, n (%)</td>
<td>8/10 (80.0)</td>
<td>2/18 (11.1)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Angiographical stenosis, % range</td>
<td>30–99</td>
<td>60–99</td>
<td>NS</td>
</tr>
<tr>
<td>Mean±SEM</td>
<td>87.2±4.7</td>
<td>82.1±2.6</td>
<td></td>
</tr>
</tbody>
</table>

*P* by Fisher test or Mann-Whitney *U* test between macrophage-rich- and macrophage-poor plaque. NS indicates not significant.
manifested that, in the controls, native apoB had a molecular weight of 500 kDa; there was no 500-kDa apoB band in plaque LDL fractions. Smear-staining was noted in the lower molecular weight range. However, in plasma LDL from the same patient, the apoB band was almost the same as in the controls, suggesting that apoB in plaques was partially degraded. Comparison of the degree of apoB degradation in MΦ-poor and MΦ-rich plaques revealed a loss of native-sized apoB and the appearance of low molecular weight (up to 120 kDa) apoB in LDL from MΦ-rich plaques. However, the changes in apoB were moderate in LDL fractions from MΦ-poor plaques; the size of low molecular weight bands from degraded apoB was slightly larger (up to 200 kDa).

Itabe et al.18 reported that partially degraded apoB accumulated in lysosomes after OxLDL was taken up by cultured macrophages. The observation that apoB in MΦ-rich plaques tended to be of smaller molecular size may be reflective of metabolic activity acting on OxLDL in macrophages present in these plaques.

**Immunohistopathology**

Figure 4 shows representative cases of MΦ-rich- and MΦ-poor plaques, including examples of intraplaque hemorrhage (Figure 4A) and plaque rupture (Figure 4B). In MΦ-rich, but not in MΦ-poor plaques, accumulations of macrophage-derived foam cells and macrophages were noted (Figure 4C).
The localization pattern of DLH3-antigens (Figure 4D) was consistent with that of macrophages detected by anti-macrophage antibodies (Figure 4C). DLH3-positive antigens were detected more in \( \text{M}\phi\)-rich than \( \text{M}\phi\)-poor plaques (Figure 4D, 4F, and 4H). This corresponds with the finding that the OxLDL level was 3.6 times higher in \( \text{M}\phi\)-rich than \( \text{M}\phi\)-poor plaques (Figure 2B). Foamy and macrophage-like cells showed strong staining with DLH3 (Figure 4D). The intracellular space between relatively small foam cells tended to be more strongly stained than that of giant and expanded foam cells (Figure 4F), suggesting that small foam cells are more active and that large foam cells are prone to rupture. The necrotic core was immunostained with DLH3, but the degree of staining was variable. The distribution of smooth muscle cells differed from that of DLH3 antigen. ApoB was deposited mainly in the extracellular matrix and occasionally colocalized with OxLDL in macrophage-derived foam cells.

**Comparison of OxLDL Levels, Lipid Parameters, Patient Symptoms and Risk Factors**

There was no significant correlation between plasma lipid parameters and OxLDL levels in either plasma or plaques (data not shown). Plasma OxLDL was not correlated with plasma LDL \( (r=-0.07, P=0.63) \). There was also no association between plasma and plaque OxLDL levels \( (r=0.20, P=0.20) \). In symptomatic patients \( (n=19) \), the plaque OxLDL levels were higher than in asymptomatic patients \( (n=25) \), however, the difference was not significant \( (15.0\pm 3.5\text{ versus } 9.6\pm 1.4\text{ ng/\mu g apoB}) \). Furthermore, macrophage infiltration was not significantly different between symptomatic and asymptomatic patients \( (5.6\pm 0.8\text{ versus } 4.4\pm 0.6\%\text{ of total area}) \). There was also no significant difference between symptomatic and asymptomatic patients with respect to their plasma OxLDL levels. Patient risk factors appeared to have no effect on plasma and plaque OxLDL levels (data not shown).

**Discussion**

OxLDL is present in atherosclerotic lesions.\(^1\)\(^-\)\(^4\) Immunohistochemical studies using antibodies to OxLDL revealed the presence of OxLDL-related epitopes in atherosclerotic lesions.\(^6\)\(^,\)\(^8\)\(^,\)\(^10\)\(^,\)\(^12\) Ylä-Herttuala et al\(^11\)\(^,\)\(^19\) isolated the LDL fraction from human atherosclerotic lesions and compared the biological properties of OxLDL with those in normal intima. Although it is widely accepted that the atherogenic oxidation of LDL occurs predominantly in the arterial wall, to our knowledge, no quantitative analyses of OxLDL in human atherosclerotic lesions have been reported. We first determined both plasma and plaque OxLDL levels by a sandwich ELISA using DLH3 and anti-apoB antibody. Then we investigated the correlation between these levels and the morphology of atherosclerotic carotid plaques. The results showed that in carotid plaques the OxLDL level was nearly 70 times higher than in plasma from the same patient. The OxLDL level in control plasma suggests that approximately 1 in 7500 LDL particles was oxidatively modified. This is consistent with values reported previously.\(^7\) Our sandwich ELISA procedure used anti-OxPC and anti-apoB antibodies, thus facilitating the determination of preformed OxLDL in biological samples. OxLDL detected by this method can be assumed to be OxPC-apoB adduct-containing LDL particles; the results do not reflect OxLDL concentration but rather the ratio of oxidatively modified LDL particles in total LDL. Because OxPC is one of the oxidative products formed in OxLDL, it should be noted that this method might not be able to detect the total amount of OxLDL.

In the course of atherogenesis, foam cells may play a critical role in the progression of lesions. Monocyte-derived macrophages can produce cytokines, proteolytic enzymes (particular metalloproteases), and growth factors. We posit that the atherogenicity of macrophages increases with their ability to take up OxLDL and leads to plaque instability. Based on the degree of macrophage infiltration, we classified the plaques as \( \text{M}\phi\)-rich and \( \text{M}\phi\)-poor. The former had significantly higher rates of rupture, fibrous cap thinning, intraplaque hemorrhage, and a bigger lipid core than did \( \text{M}\phi\)-poor plaques. This suggests that \( \text{M}\phi\)-rich plaques tend to be unstable. Moreover, plaque OxLDL levels exhibited a strong correlation with macrophage infiltration \( (P<0.0001) \). In patients with \( \text{M}\phi\)-rich plaques, plasma OxLDL levels were significantly higher than in the controls, suggesting that the elevation of plasma OxLDL is associated with plaque instability. Our findings support the hypothesis that the OxLDL level and the degree of macrophage infiltration determines whether atherosclerotic lesions are stable or unstable. Immunohistochemically, in \( \text{M}\phi\)-rich plaques, macrophage infiltration was 3 times higher than in \( \text{M}\phi\)-poor plaques and foam cells, and macrophages stained strongly for the DLH3-antigen. In \( \text{M}\phi\)-poor plaques, on the other hand, foam cells and macrophages stained only faintly. These findings also reflect the results of direct quantitative analyses. The distribution of DLH3- and apoB-positive areas was consistent with that reported previously.\(^5\)\(^,\)\(^9\)\(^,\)\(^20\)\(^,\)\(^21\)

Western blots using apoB antibody showed that apoB in plaques was partially degraded and that the extent of apoB degradation was related to the OxLDL level. ApoB was more extensively fragmented in \( \text{M}\phi\)-rich (high OxLDL levels) than \( \text{M}\phi\)-poor plaques (lower OxLDL levels). The accumulation of partially degraded apoB in lysosomes after exposure of murine macrophages to OxLDL has been reported\(^16\) and the size of degraded apoB in \( \text{M}\phi\)-rich plaques (up to 120kDa) corresponded to the size reported for murine macrophages. We posit that the uptake and degradation of OxLDL is enhanced in \( \text{M}\phi\)-rich plaques. It should be noted that plaque LDL would be extracted from both intracellular and extracellular spaces under our experimental conditions, because the plaques were homogenized with a polytron homogenizer. Degradation of apoB in plaque LDL was less prominent in a previous study by Ylä-Herttuala et al\(^11\) presumably because their milder extraction condition allowed them to extract extracellular LDL particles. In addition, there might be a contribution from extracellular proteases that degrade OxLDL in plaques because macrophages are an important source of extracellular proteases. Western blots detected little apoB degradation in plasma LDL fractions, presumably because the content of OxLDL in these fractions was very low. ApoB may be more susceptible to oxidative modifica-
tion in plaques than in plasma; alternatively, plaques may have greater ability to retain circulation-derived OxLDL.

Smith et al.²² showed that the interior of advanced human atherosclerotic lesions is a highly pro-oxidant environment containing transition metal ions. Plaque instability may induce a focal imbalance between pro-oxidants and anti-oxidant defense- and repair systems and contribute partly to high levels of plasma OxLDL. Our study did not address the origin of circulating OxLDL or the mechanisms underlying the increase in plasma OxLDL. We found no significant correlation between patient symptoms and their plasma Ox-

LDDL levels. Furthermore, the difference in the plasma Ox-LDL between patients with Mφ-poor- and Mφ-rich plaques was not significant. The timing of the CEA operation may affect the plasma OxLDL level. All of our patients had either suffered a stroke more than one month earlier or were asymptomatic at the time of the operation. We posit that the plasma OxLDL level during the chronic phase may be the stable level for the individual and that the higher plasma OxLDL level in patients with Mφ-rich plaques may reflect the susceptibility of their LDL to oxidation. To address these issues, we are currently performing studies in which we are obtaining measurements from a large patient population in the acute stroke phase.

Ours is the first report on OxLDL levels in paired plasma and plaque samples from CEA patients with Mφ-rich and Mφ-poor plaques. Our results suggest that LDL undergoes further oxidation in plaques, that plasma OxLDL levels reflect oxidative conditions, and that high plasma and plaque OxLDL levels are associated with the vulnerability to rupture of carotid atherosclerotic lesions.

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