Insulin Treatment Prevents LDL from Accelerated Oxidation in Patients with Diabetes

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In a study population, we compared the level of malondialdehyde-modified LDL (MDA-LDL) with the concentrations of lipid parameters in serum and found a strong correlation between MDA-LDL and apolipoprotein B (apo B) concentrations. Their interrelations had a turning point at an apo B concentration of 1,150 mg/l. In diabetic patients, the ratio of MDA-LDL/apo B increased at apo B concentrations above 1,150 mg/l. This ratio represents the extent of modification of apo B by MDA. In the control subjects, this ratio remained stable. When we divided the patients into medication groups (statins and insulin), we found that the 1,150 mg/l threshold disappeared. At apo B concentrations above 1,150 mg/l, the ratio of MDA-LDL/apo B in the statin group was as high as that in the non-drug group. In the insulin group, the means of MDA-LDL/apo B in all ranges of apo B levels decreased to an extent statistically indistinguishable from those of the control group. In conclusion, insulin therapy represses LDL oxidation even at apo B concentrations > 1,150 mg/l and should be noted for its anti-oxidation properties.


Key words: Malondialdehyde modified LDL, Small dense LDL, Insulin, Statin

Introduction

LDL has been implicated in the initial events forming atherosclerotic lesions (1). LDL-cholesterol (LDL-C) lowering therapy has been considered essential for the clinical prevention of lesion development. Dietary therapy remains the first choice of treatment for high LDL-C. If necessary, drug therapy is recommended for patients considered at high risk for coronary heart disease (CHD).

3-Hydroxy-3-methylglutaryl CoA (HMG-CoA) reductase inhibitors, or statins, are one type of LDL-C lowering drug. The inhibition of HMG-CoA reductase, the rate-limiting enzyme of cholesterol synthesis, lowers hepatic cholesterol content and stimulates hepatic LDL receptor synthesis (2). As a consequence, LDL in the bloodstream is taken up by the hepatic LDL receptor.

In NIDDM, the risk of CHD increases with enhancing dyslipidemia (3,4). Usually, total and VLDL triglycerides (TG) are elevated, whereas concentrations of serum LDL cholesterol are normal or slightly elevated. It is known that insulin therapy induces a significant drop not only in total and VLDL TG but also in small, dense LDL with no change in LDL-C (5,6). The small, dense LDL particles are considered more atherogenic than normal ones.

Austin et al. (7) have shown that LDL subclass pattern B, characterized by a preponderance of small, dense LDL particles, is associated with an increased risk of myocardial infarction. Information has recently been accumulated on small, dense LDL. When different subpopulations of small, dense LDL are isolated from the same donor, the smallest LDL fraction shows the poorest LDL-receptor binding (8). Furthermore, small LDL is more susceptible than normal LDL to oxidation in the presence of copper ions (9,10).

These findings evoke great interest in the relation between drugs, oxidation, and LDL, especially small, dense LDL. In this study, we investigated small LDL and malondialdehyde (MDA)-modified LDL (MDA-LDL) in dia-
betic patients undergoing drug therapy with statins or insulin, using gradient gel electrophoresis and enzyme linked immunosolvent assay (ELISA), respectively.

Materials and Methods

Reagents
Colloidal gold particles (10 and 15 nm in diameter) were purchased from E-Y Laboratories, Inc. Carbonic anhydrase derived from bovine erythrocytes was obtained from Roche Diagnostics Inc. Monoclonal antibodies against MDA-LDL (ML25) and apolipoprotein B (apo B) (AB16), and 2-15% nondenaturating polyacrylamide gel were obtained from Daiichi Pure Chemicals Co.

Subjects
The study subjects included 63 patients with diabetes mellitus (DM) and 35 healthy controls. Diabetic patients were recruited from the Hamamatsu University School of Medicine Hospital. The control subjects and patients with DM were matched for age and sex. All patients (32 women and 31 men; age range, 33 through 72 years) were randomly selected from diabetic patients, with glycosylated hemoglobin (HbA1c) values ≥ 6% at the onset of participation in the study. DM was diagnosed according to the criteria of the World Health Organization (11). Fourteen subjects were IDDM patients (8 women and 6 men); 5 patients had received statins and 7 were already on insulin therapy. The other 49 subjects were NIDDM patients (24 women and 25 men); 7 patients were treated with statins and 10 were treated with insulin therapy. None of the patients were taking drugs, other than insulin or statins, known to influence lipid metabolism. Statin treatment consisted of pravastatin (11 subjects) and simvastatin (1 subject). No subjects received a combination of insulin and statins. The control subjects (18 women and 17 men; age range, 34 through 72 years) were selected from hospital staff, with serum total cholesterol (TC) concentrations ≤ 5.69 mmol/L and serum TG concentrations ≤ 1.69.

Quantitation of MDA-LDL by ELISA
The ELISA was based on the method previously reported by Kondo et al. (12). Briefly, serum samples were diluted 2400-fold in a dilution buffer. After the diluted sample was preincubated, it was added (in duplicate) to the wells of plates coated with a monoclonal antibody against MDA-LDL (ML25). The reaction stood for 1 h at room temperature, and then the plates were washed. β-Galactosidase-conjugated monoclonal antibody against apo B (AB16) was added, and the mixture was incubated for 30 min at room temperature. The excess enzyme-labeled antibody was washed away, and o-nitrophenyl-β-galactopyranoside as a substrate was pipetted into the wells. After 2 h, the reaction was stopped by the addition of 0.2 mol/L sodium carbonate (pH 12). Absorbance in the individual wells was determined at 415 nm with a microplate reader. In each assay, an aliquot of reference sera was applied as a secondary standard.

Procedure for nonnaturating gradient-gel electrophoresis
The major peak of LDL diameter was estimated with nondenaturating polyacrylamide gradient-gel electrophoresis according to our previous method (12). Briefly, 5 µL of serum from each subject was diluted 2-fold with 40% sucrose and electrophoresed for 24 h at 10°C in 2% to 15% polyacrylamide gradient gels. The gels were stained with oil red O. The standard lane was stained with Coomassie blue R250, and a standard curve was constructed on the migration distances of five known marker diameters: ferritin (12.2 nm), thyroglobulin (17.0 nm), thyroglobulin dimmer (23.6 nm, Pharmacia), and protein-coated gold particles (21.1 and 29.2 nm, our preparations for the previously reported technique). As a reference standard, a control serum was run with each gel. The location of individual bands was compared to that of the control serum in the scanning of each gel. The predominant LDL size in each sample lane was estimated from the migration distance of the major LDL peak and calculated from the equation shown in the next section. Each gel lane was scanned with a densitometer interfaced with a PC computer.

Calculation of the predominant LDL size in a sample
Using the method of Williams et al. (13) with minor modifications (12), we calculated the equation for converting molecular diameter (nm) to migration distance (RF). The equation was derived from the distances between size markers. Each particle’s RF, measured relative to apoferritin, was plotted against the particle diameter (nm).

Analytical methods for lipids and other parameters
Serum TC, TG, and HDL-C levels were determined enzymatically (Daiichi). Apo B and apolipoprotein A-I (apo A-I) in serum were measured with a commercial immunoturbidimetric assay (Daiichi). HbA1c levels in whole blood were determined with high-performance liquid chromatography. The normal range of HbA1c levels was 4.0%-5.8%. LDL-C content was calculated according to the method of Friedewald et al. (14), except in subjects with serum TG at a concentration of > 4.52 mmol/L.

Statistical analysis
Statistical analysis was performed using the Statistical Package (SPSS 10.0.J for Windows). All data are expressed as the mean ± SD. The Spearman rank correlation coefficients were computed to identify significant associations between the levels of MDA-LDL and either the levels of lipid parameters or the predominant LDL size. Comparisons of the mean values of lipid parameters between the diabetic patient groups were made with Student’s t test. Multiple comparisons with variables
between diabetic patients in the no-treatment group and the medication groups were performed with one-way ANOVA followed by the Bonferroni test. Linear regression analysis and Pearson correlation coefficients were performed to examine the relation between MDA-LDL and apo B in the control and patient groups. To define the relation between the levels of MDA-LDL and predominant LDL size in the control subjects and diabetic patients, when limited to apo B levels, multiple comparisons were also performed with one-way ANOVA. The association between variables was evaluated by linear regression analysis and Pearson correlation coefficients. A probability (P) less than 0.05 was considered statistically significant.

**Results**

**Lipid parameters in control subjects and diabetic patients**

Table 1 shows the mean values for the lipid parameters in the control subjects and patients with DM. HDL-C and apo A-I concentrations, and the predominant LDL size in subjects with DM were lower (p < 0.01, p < 0.001 and p < 0.01, respectively) than those in the control subjects. Levels of TC, LDL-C, TG, MDA-LDL, and apo B were significantly higher (p < 0.01) in subjects with DM than in the control subjects, except for the difference (p < 0.001) of MDA-LDL and apo B levels between those.

**Correlation coefficients between lipid parameters**

Table 2 shows the correlation coefficients comparing the levels of MDA-LDL and other lipid parameters for the control and DM patient groups combined. In the study population, MDA-LDL concentration was inversely correlated with the level of HDL-C and LDL size, and positively correlated with TG, TC, LDL-C, and apo B levels. Apo A-I level had no association with the concentration of MDA-LDL. Overall, interrelations closely existed between the concentrations of MDA-LDL and apo B.

**Correlation studies between MDA-LDL and apo B in control and diabetic subjects**

We analyzed the relation of MDA-LDL and apo B by dividing the population into control and diabetic patients (Fig. 1). In the control subjects, the linear regression analysis and Pearson correlation coefficients were y = 0.65x - 0.9, r = 0.73. In the diabetic patients, there seemed to be a turning point in that relation at an apo B concentration of approximately 1,150 mg/l. We dichotomized the groups into apo B concentrations of 1,150 mg/l or less and more than 1,150 mg/l. The former exhibited a correlation equation as follows: y = 0.81x - 4.6, r = 0.64, and the latter was y = 3.08x-274.0, r = 0.67.

**Comparison of lipid parameters in diabetic patients**

We investigated the influence of insulin and statins on the lipid parameters in subjects with DM (insulin group and statins group). Table 3 shows the mean values of lipid parameters in diabetic subjects. The concentration of TG in the statins group was significantly higher (p < 0.05 and p < 0.05, respectively) than in the no-treatment group and the insulin group. The insulin group tended to have lower MDA-LDL and apo B levels than the group without medication. No significant differences between these groups existed in HbA1c and fasting blood glucose levels. The other lipid parameters showed no significant differences among these groups.

**Influence of drugs on the relation between MDA-LDL and apo B**

We also studied the effects of insulin and statins on the relation between levels of MDA-LDL and apo B (Fig. 2). The turning point in that relation disappeared in each drug treatment group, unlike the total patients. The regression equations and Pearson correlation coefficients were y = 0.86x-13.9 and r = 0.81 for the insulin group, and y = 1.41x-62.8 and r = 0.62 for the statins group. The slope of the regression line in the insulin group was less than that in the statin group.

**Comparison of MDA-LDL/apo B and LDL size between the control and drug treatment groups**

Comparisons between the control subjects and patient groups were performed in relation to the ratios of MDA-

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**Table 1. Lipid parameters in control subjects and patients with DM**

<table>
<thead>
<tr>
<th></th>
<th>Control (n = 35)</th>
<th>DM (n = 63)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>year</td>
<td></td>
</tr>
<tr>
<td></td>
<td>54.1 ± 10.1</td>
<td>54.9 ± 8.8*</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>4.84 ± 0.47</td>
<td>5.40 ± 1.06*</td>
</tr>
<tr>
<td>HDL-C</td>
<td>mmol/l</td>
<td>1.63 ± 0.41</td>
</tr>
<tr>
<td>LDL-C</td>
<td>mmol/l</td>
<td>2.77 ± 0.47</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>mmol/l</td>
<td>0.94 ± 0.33</td>
</tr>
<tr>
<td>LDL size</td>
<td>nm</td>
<td>26.2 ± 0.8</td>
</tr>
<tr>
<td>MDA-LDL</td>
<td>units/l</td>
<td>52 ± 12</td>
</tr>
<tr>
<td>Apo A-I</td>
<td>mg/l</td>
<td>1470 ± 170</td>
</tr>
<tr>
<td>Apo B</td>
<td>mg/l</td>
<td>820 ± 140</td>
</tr>
</tbody>
</table>

Abbreviations: DM - diabetes mellitus; HDL-C - high-density lipoprotein-cholesterol; LDL - low-density lipoprotein; LDL-C - LDL-cholesterol; MDA-LDL - malondialdehyde modified LDL; apo B - apolipoprotein B; apo A-I - apolipoprotein A-I Values are mean ± SD. Statistical differences between control subjects and diabetic patients are shown by p values; **p < 0.01, ***p < 0.001.

1The number of patients, except for 3 patients with triglyceride levels of > 4.52 mmol/l because LDL-C content was calculated by the method of Friedewald et al. (14).
Table 2. Spearman correlation coefficients between lipid parameters

<table>
<thead>
<tr>
<th></th>
<th>TG</th>
<th>TC</th>
<th>HDL-C</th>
<th>LDL-C</th>
<th>LDL size</th>
<th>Apo A-I</th>
<th>Apo B</th>
</tr>
</thead>
<tbody>
<tr>
<td>MDA-LDL</td>
<td>0.629</td>
<td>0.656</td>
<td>-0.415</td>
<td>0.669</td>
<td>-0.545</td>
<td>-0.192</td>
<td>0.803</td>
</tr>
</tbody>
</table>

Abbreviations: HDL-C, high-density lipoprotein-cholesterol; LDL, low-density lipoprotein; LDL-C, LDL-cholesterol; MDA-LDL, malondialdehyde modified LDL; apo B, apolipoprotein B; apo A-I, apolipoprotein A-I.

In comparison with LDL-C level, 3 patients with triglyceride levels of > 4.52 mmol/l were subtracted from the whole number of patients according to the method of Friedewald et al. (14). All P values of correlation coefficients were less than 0.001, and did not reach significant difference between the levels of MDA-LDL and apo A-I.

Table 3. Comparison of lipid parameters in diabetic subjects

<table>
<thead>
<tr>
<th></th>
<th>No-drug (n = 34)</th>
<th>Insulin (n = 17)</th>
<th>Statins (n = 12)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>55.3 ± 8.4</td>
<td>53.9 ± 10.1</td>
<td>55.0 ± 9.1</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>5.48 ± 1.16</td>
<td>5.07 ± 0.93</td>
<td>5.69 ± 0.96</td>
</tr>
<tr>
<td>HDL-C</td>
<td>1.29 ± 0.31</td>
<td>1.42 ± 0.54</td>
<td>1.29 ± 0.41</td>
</tr>
<tr>
<td>LDL-C</td>
<td>3.49 ± 0.93 (34)</td>
<td>3.03 ± 0.83 (16)</td>
<td>3.26 ± 0.65 (10)</td>
</tr>
<tr>
<td>Triglyceride</td>
<td>1.46 ± 0.85</td>
<td>1.42 ± 1.49</td>
<td>2.66 ± 2.04 *</td>
</tr>
<tr>
<td>LDL size</td>
<td>25.6 ± 1.3</td>
<td>25.7 ± 1.2</td>
<td>25.1 ± 1.0</td>
</tr>
<tr>
<td>MDA-LDL</td>
<td>92 ± 48</td>
<td>64 ± 28</td>
<td>85 ± 41</td>
</tr>
<tr>
<td>Apo A-I</td>
<td>1270 ± 220</td>
<td>1250 ± 230</td>
<td>1280 ± 220</td>
</tr>
<tr>
<td>Apo B</td>
<td>1040 ± 280</td>
<td>910 ± 270</td>
<td>1050 ± 190</td>
</tr>
<tr>
<td>HbA1c %</td>
<td>8.2 ± 1.5</td>
<td>8.8 ± 1.4</td>
<td>8.9 ± 1.3</td>
</tr>
<tr>
<td>FBG</td>
<td>9.94 ± 3.77</td>
<td>11.55 ± 4.94</td>
<td>10.55 ± 3.89</td>
</tr>
</tbody>
</table>

Abbreviations: HDL-C, high-density lipoprotein-cholesterol; LDL, low-density lipoprotein; LDL-C, LDL-cholesterol; MDA-LDL, malondialdehyde modified LDL; apo B, apolipoprotein B; apo A-I, apolipoprotein A-I; HbA1c, glycosylated hemoglobin; FBG, fasting blood glucose.

Values are mean ± SD.

Statistical differences between diabetic patients not taking a drug and those taking statins, or those taking insulin and statins are shown by P values; *p < 0.05, **p < 0.01, ***p < 0.001, respectively.

The number of subjects, except for patients with triglyceride levels of > 4.52 mmol/L because LDL-C content was calculated by the method of Friedewald et al. (14).

Table 4. MDA-LDL/apo B and LDL size in control and DM medication groups

<table>
<thead>
<tr>
<th></th>
<th>MDA-LDL/apo B</th>
<th>LDL size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apo B level (mg/l)</td>
<td>≤ 1,150</td>
<td>&gt; 1,150</td>
</tr>
<tr>
<td>Control</td>
<td>0.064 ± 0.009 (35)</td>
<td>—</td>
</tr>
<tr>
<td>DM medication</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No-drug</td>
<td>0.083 ± 0.021 (19) *</td>
<td>0.090 ± 0.034 (15) **</td>
</tr>
<tr>
<td>Insulin</td>
<td>0.068 ± 0.018 (13)</td>
<td>0.078 ± 0.023 (4)</td>
</tr>
<tr>
<td>Statins</td>
<td>0.071 ± 0.024 (9)</td>
<td>0.106 ± 0.035 (3) *</td>
</tr>
</tbody>
</table>

Abbreviations: DM, diabetes mellitus; LDL, low-density lipoprotein; MDA-LDL, malondialdehyde modified LDL; apo B, apolipoprotein B.

Values are mean ± SD.

Statistical differences between parameters (MDA-LDL/apo B or LDL size) in control (apo B ≤ 1,150) and patient groups with DM are shown by P values; *p < 0.05, **p < 0.01, ***p < 0.001.

The number of subjects

LDL to apo B levels and distributions of LDL size (Table 4). Each patient group was divided into two groups on the basis of apo B concentrations (≤ 1,150 mg/l and > 1,150).

Regardless of the concentration group, the MDA-LDL/apo B ratio of no-drug patients was significantly greater (p < 0.05 and p < 0.01, respectively) than that of the control.
subjects. There was no statistically significant difference in the MDA-LDL/apo B ratio between the control and insulin groups. The statin group significantly differed ($p < 0.05$) from the control subjects at an apo B concentration of $> 1,150 \, \text{mg/l}$. In regards to LDL size (at an apo B concentration of $> 1,150 \, \text{mg/l}$), all patient groups were statistically smaller LDL ($p < 0.05$ for the no-drug and insulin groups, and $p < 0.001$ for the statin group) than the control.

**Correlations between LDL size and MDA-LDL level**

A correlation study (Fig. 3) between LDL size distribution and MDA-LDL level in the control subjects revealed that LDL modification by MDA is depressed in the presence of small and normal-sized LDL. In the no-drug patient group, LDL modification was accelerated in small and normal-sized LDL. Small LDL did not repress modification in the statin group, although the MDA-LDL levels tended to decrease in the presence of normal-sized LDL. In the insulin group, a depression of LDL modification by

![Graphs showing correlations](image)
Insulin Prevents LDL from Oxidation

In general, small dense LDL is associated with an increased risk of myocardial infarction (7), and the synergy between plasma apo B level and the distribution of small dense LDL produces a possible risk of ischemic heart disease (21, 22). This synergistic effect occurs as apo B concentration reaches 1,100 to 1,200 mg/l, consistent with our findings of a turning point at an apo B concentration of 1,150 mg/l. Small LDL has a low binding affinity for the cellular LDL receptor (23, 24), resulting in a prolonged presence in the plasma. Small LDL, compared to normal-sized LDL, is exposed to oxidative stress for a sufficient time to easily generate oxidized LDL (9). Using density gradient ultracentrifugation, we previously observed that distributions of MDA-LDL were localized in fractions of small dense LDL (25). These findings suggest that high levels of apo B lead to the promotion of MDA-LDL generation through a high distribution of small dense LDL. The significant correlation between MDA-LDL level and LDL size distribution (p < 0.001) supports such claims.

We then examined the correlations between MDA-LDL level and LDL size distribution in distinctive therapeutic groups. In the statin group, as well as the no-drug group, at an apo B concentration > 1,150 mg/l, the mean LDL size was significantly lower (p < 0.001 and p < 0.05, respectively) than in the control group. This may explain why the ratio of MDA-LDL/apo B in the statin group remained high in this range. LDL oxidation easily occurs because LDL is converted into a small size. At a concentration of apo B ≤ 1,150 mg/l, the MDA-LDL/apo B ratio diminished, possibly explained by the LDL size. In this range, LDL size increased at the same rate in the statin and control groups. However, we failed to determine why the mean of the MDA-LDL/apo B ratio in the statin group was lower than that in patients without drugs despite the same LDL size.

Generally, by inhibiting HMG-CoA reductase, statins decrease the LDL-C level in plasma. This reduction of the LDL-C level occurs more markedly in light LDL particles than in heavy LDL particles (26). Electrophoresis showed that heavy LDL coincides with small LDL (25, 27). Accordingly, in our study, in the statin group there was a more rapid metabolism of normal-sized LDL than of small LDL, resulting in a decreased MDA-LDL level in the case of normal-sized LDL. As a consequence of the rapid metabolism of LDL, MDA-LDL is replaced by unmodified LDL. In large LDL, the values of MDA-LDL in patients treated with statins became lower than in patients without drug treatment. This may explain why the MDA-LDL/apo B (apo B ≤ 1,150 mg/l) ratio in the statin group was reduced despite the same LDL size. In contrast, in the apo B range > 1,150 mg/l, MDA-LDL/apo B was unchanged. These results are consistent with a previous finding that the catabolism of LDL apo B associated with pravastatin increased in the light fraction with no effect.

MDA was observed in all LDL sizes.

Discussion

Numerous studies (15-18) have shown that an increased risk of atherosclerosis is associated with abnormalities in lipid metabolism, including elevated levels of plasma TC, TG, LDL-C, Lp(a), and apo B, and reduced levels of HDL-C and apo A-1. However, the value of apo B in plasma has not been practically used as a marker of atherogenic risk. In our study, there was a high correlation between concentrations of MDA-LDL and apo B (Table 2). MDA-LDL, an oxidized LDL, has been considered as another indicator of the direct formation of atherogenic lesions (19, 20). We found that there exists a turning point in the relation between levels of MDA-LDL and apo B (Fig. 1). Our data indicate that concentrations of apo B above 1,150 mg/l promote the modification of LDL by MDA. The reference range of plasma apo B was 690 to 1,050 mg/l, implying a tolerance range which is capable of maintaining a constant ratio of modification in apo B or LDL.

We investigated the influence of medication on the relation between levels of MDA-LDL and apo B, and found a significant difference between diabetic patients treated with statins and those treated with insulin (Fig. 2). This relation changes when analyzing data by medication group. The MDA-LDL/apo B ratio, which signifies the extent of modification of apo B by MDA, was higher in the statin group than in the insulin group at apo B concentrations > 1,150 mg/l (Table 4).

Fig. 3. Correlations studies between LDL size distribution and MDA-LDL level in each group.
on the very dense fraction (28). Overall, the MDA-LDL level in the statin group rapidly decreases with declining apo B concentrations along a linear regression formula between both levels (y = 1.41x - 62.8). Judging statin medication from the MDA-LDL value, the apo B concentration in patients should fall to within the reference range due to a sufficient reduction of the MDA-LDL level. Some epidemiological studies suggest that a reduction in coronary events in patients treated with statins is achieved by lowering the LDL-C level, such as to below 3.36 mmol/l (29). Considering the correlation between the levels of apo B and MDA-LDL, a reduction of LDL-C level means decreasing the MDA-LDL level through lowering the apo B concentration. In the case of the insulin group, the mean MDA-LDL/apo B ratio decreased in all ranges of apo B levels and was statistically similar to those in the control group. Recently, it was shown that insulin therapy decreases the proportion of small dense LDL particles in diabetic patients (6, 30, 31). Therefore, it is possible that the LDL size distribution in our insulin group was localized in the larger LDL. The mean LDL size in the insulin group tended to be larger than that in the no-drug patients (Table 3). However, when divided into two groups by apo B level, the LDL size remained small at apo B > 1.150 mg/dl while remaining normal-sized at ≤ 1.150 mg/dl, as in previous reports. Furthermore, since the HbaA₁ and fasting blood glucose levels in the insulin group were the same as in other groups, LDL glycation in this group, which could promote auto-oxidation of the particle, may not be depressed. This failed to explain the reduction of MDA-LDL/ apo B in the insulin group. Inversely, Quijones-Galvan et al. (32) reported that an acute effect of insulin administration is the susceptibility to oxidation of LDL particles. They considered that as large buoyant LDL is easily taken up by the LDL receptor rather than small dense LDL, insulin treatment by upregulating the LDL receptor (33) is associated with an enrichment of small dense LDL in plasma at the initial stage. In particular, the effect in hypertriglyceridemic subjects is enhanced in the presence of a predominance of small dense LDL. If this hypothesis is correct, LDL size and MDA-LDL/ apo B ratio at apo B ≤ 1.150 mg/dl that become normal and lower, respectively, could be explained from the chronic effect of insulin administration. However, at apo B > 1.150 mg/dl, the insulin effect is not enough to normalize the LDL size for a further influx of LDL apo B in circulation. Nevertheless, in our study, the MDA-LDL/apo B ratio became lower. In conclusion, in diabetic patients, insulin prevents LDL from accelerating oxidation by a different mechanism from that of statins. Insulin therapy has the characteristic of suppressing LDL oxidation even at apo B levels of > 1.150 mg/dl. Therefore, insulin therapy should be noted for its anti-oxidative properties.

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References

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