Reduced cholesteryl ester transfer in plasma of patients with lipoprotein lipase deficiency

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Abstract The net mass transfer of cholesteryl ester (CE) from high density lipoprotein (HDL) to the apolipoprotein (apo) B-containing lipoproteins, very low density lipoprotein (VLDL) and low density lipoprotein (LDL) in plasma (cholesteryl ester transfer (CET)) from three patients lacking lipoprotein lipase (LpL) activity was significantly lower (P < 0.001) than in plasma from fasting control subjects with comparable triglyceride levels. Chylomicrons isolated from LpL-deficient fasting plasma showed the same low level of CET activity as observed in the intact plasma when combined with HDL and cholesteryl ester transfer protein (CETP)-containing d 1.063 g/ml bottom fractions from control subjects. Preincubation of chylomicrons and large triglyceride-rich lipoproteins (Sf > 400) from LpL-deficient plasma with milk LpL, however, stimulated the capacity to engage in CET 4- to 5-fold to the same level as chylomicrons and VLDL from control subjects after a fat load. Consistent with these measurements of CET activity in plasma, chylomicrons obtained from the LpL-deficient subjects after a 14-h fast had higher TG/CE ratios than chylomicrons from controls 3 h after ingesting a fat load (LpL-deficient 26.3 ± 9.0 vs. controls 6.9 ± 2.1; mean ± SD). The mass of CETP did not differ in LpL-deficient and control subjects (LpL-deficient 1.03 ± 0.22 µg/ml vs. controls 1.58 ± 0.58 µg/ml). These studies are consistent with earlier in vitro studies showing that the actions of lipoprotein lipase and its lipolytic products are essential for maximal cholesteryl ester transfer protein activity.—Bagdade, J. D., M. C. Ritter, H. Lithell, D. Bassett, F. Mailly, P. Talmud, and M. R. Hayden. Reduced cholesteryl ester transfer in plasma of patients with lipoprotein lipase deficiency. J. Lipid Res. 1996. 37: 1696-1703.

Supplementary key words lipoprotein lipase • chylomicrons • VLDL • cholesteryl ester transfer protein

Castro and Fielding (1) first observed that lipoprotein remodeling is usually maximal during the postprandial state coincident with the peak of lipoprotein lipase (LpL) activity and the delipidation of the triglyceride-rich lipoproteins. At this time the esterification of free cholesterol on HDL by lecithin:cholesterol acyltransferase (LCAT) occurs closely associated with the hetero-exchange of cholesteryl ester and triglyceride between HDL and the apoB-containing lipoproteins (cholesteryl ester transfer (CET)). This shuttling of core and surface lipids is mediated by a 74 kDa protein that has been designated cholesteryl ester transfer protein (CETP) (2). Tall et al. (3) have shown that this reaction increased markedly when VLDL from fasting human plasma was exposed to LpL. These and subsequent studies by these workers showed that ionized non-esterified fatty acids (NEFA), generated by the delipidating actions of LpL, localized in the surface coat and promoted CET by enhancing the affinity of VLDL and, by implication, chylomicrons for CETP (4).

If LpL is an activator of CET, one would predict: first, that the level of CET in plasma from LpL-deficient patients would be low; and second that exposure of their TG-rich lipoproteins to LpL would generate lipolytic products that normalize their interaction with CETP. To test these possibilities, the CET activity of plasma, chylomicrons, and VLDL from LpL-deficient and control subjects was studied.

METHODS

Human subjects

For the CET-related experiments, three non-diabetic LpL-deficient patients and five normolipidemic controls were studied. The study protocol was approved by the Institutional Human Investigation Committee of each...
participating institution. Informed consent was obtained from subjects and the project was carried out according to the principles of the Declaration of Helsinki. Plasma was obtained after an overnight fast from the LpL-deficient subjects, normolipidemic controls, and from three otherwise healthy non-diabetic patients (two males, ages 54 and 57 yr; one female, age 62 yrs; mean group age 57.7 ± 4 yr) with hypertriglyceridemia (TG, 437 ± 227 mg/dl; cholesterol, 226 ± 26 mg/dl; HDL-C, 36.5 ± 7.5 mg/dl) prior to receiving treatment. Blood samples were also drawn from two healthy normolipidemic male control subjects 3 h after they had ingested a high-fat meal consisting of two cups of dairy cream with oatmeal, two eggs, and sausage.

**Characterization of LpL deficiency**

The mutations in the gene for LpL were characterized in the first LpL-deficient patient in the laboratory of Dr. Steve Humphries in the Division of Cardiovascular Genetics at University College London Medical School. This Swedish patient, a compound heterozygote for LpL deficiency, had a new previously undescribed novel mutation in exon 55 193 R and a 2 kb insertion in exon 6/intron 6 as described by Langlois (5). Two American LpL-deficient brothers were studied by Drs. Michael Hayden at the University of British Columbia and John Brunzell at the University of Washington. They were compound heterozygotes for the Glu188 Gly missense mutation in exon 5 and a duplication in exon 6 of the LpL gene. The Glu188 Gly mutation results from a single base transition from guanine to adenine at cDNA position 818 in codon 188 that substitutes glutamic acid for glycine (6). The exon 6 duplication involves a 2 kb duplication that results from juxtaposition of intron 6 to a partially duplicated exon 6 (7).

**Lipoprotein isolation**

Chylomicrons were obtained from the LpL-deficient patients' fasting plasma and from control subjects after the fat load by ultracentrifugation at 30,000 RPM for 30 min at 4°C. For the recombination studies, VLDL (d < 1.006 g/ml), LDL (d 1.019–1.063 g/ml), and HDL (d 1.063–1.21 g/ml) were isolated in the absence of an LCAT inhibitor by sequential preparative ultracentrifugation at 10°C in a 40.3 rotor at 10,000 rpm for 18 h for VLDL and LDL and 44 h for HDL. The isolated lipoprotein fractions were then dialyzed against 0.15 M NaCl, 1 mM EDTA, pH 7.4, for 48 h at 4°C.

**LpL treatment of chylomicrons**

Chylomicrons from the LpL-deficient patients and controls fed the high fat meal were preincubated with 1000 units of bovine milk lipoprotein lipase (1 nmol FA/min per ng enzyme) (provided by Dr. P. H. Iverius) or without LpL in the presence of 1 mg/ml bovine serum albumin for 30 min at 37°C and the reaction was stopped with 1 mM phenylmethylsulfonyl fluoride (PMSF); aliquots of each treated preparation were added to plasma concentrations of the d > 1.063 g/ml fractions and the net mass transfer of CE was determined.

**Cholesterol ester transfer**

The net mass of preformed cholesterol ester transferred from HDL to VLDL + LDL in intact plasma was measured during incubation at 37°C in a metabolic shaker in the presence of 1.5 mM dithio-bis-dinitrobenzoic acid (DTNB) to inhibit LCAT as previously described (8). Briefly, 0.4 ml plasma or recombined fractions was incubated for 6 h at 37°C. Aliquots were removed at zero time and after 1, 2, 4, and 6 h, chilled on ice, and the apoB-containing lipoproteins were then precipitated with 0.1 vol of heparin/MnCl$_2$ and 1.3 mg/ml heparin. MnCl$_2$ at this concentration has been found not to precipitate a significant quantity of apoE-containing HDL. At each sampling interval, the mass of free and total cholesterol present in the supernatant was measured and the amount of CE was calculated from the difference between the two values. The mass of esterified cholesterol (CE) transferred into the apoB-containing lipoproteins from HDL at each time interval was determined by subtracting this value from zero-time CE in HDL. Cholesterol ester transfer protein (CETP) mass was measured by radioimmunoassay in the laboratory of Dr. Ruth McPherson at the Ottawa Heart Institute (9).

**Analytical methods**

Enzymatic kit procedures were used to quantitate cholesterol (Boehringer-Mannheim, Indianapolis, IN) and triglyceride (Sigma Chemical Co., St. Louis, MO). HDL-C was measured by a Standardized Lipid Research Clinic heparin/MnCl$_2$ precipitation procedure in whole plasma in controls and in the chylomicron-free fractions of the LpL-deficient subjects (10). Free cholesterol was estimated with the same components of the cholesterol kit, except that cholesteryl ester (CE) hydrolase was omitted. Esterified cholesterol was determined from the difference between total and free cholesterol. Lecithin was estimated in 0.3-ml aliquots of chylomicrons, VLDL, LDL, HDL$_2$, and HDL$_3$ after mixing with 0.1 ml of 0.15 M NaCl/1 mM EDTA solution in the following steps: extraction by the Bligh and Dyer (11) procedure; separation on thin-layer chromatography plates; scraping from the plate, and measurement by a modification of the Bartlett procedure (12). Controls for the lipoprotein composition studies were 10 healthy young adults (4 males; 6 females; age 28.6 ± 4.3 yr; mean ± SD).
RESULTS

Fasting plasma triglycerides were increased significantly and total and HDL cholesterol decreased in the LpL-deficient patients (Table 1). Despite triglyceride elevation, plasma CET activity of LpL-deficient patients was not elevated and did not differ from that of the normolipidemic control subjects (Fig. 1). In contrast, plasma from dyslipidemic patients with less marked hypertriglyceridemia showed a markedly higher capacity to transfer CE from HDL to VLDL + LDL.

To determine why hyperlipidemic plasma from the LpL-deficient patients demonstrated such a low level of CET, a series of recombination experiments were performed with isolated lipoprotein fractions from the patients and control subjects. When the chylomicron-containing (Sf > 400) fractions of fasting plasma from two LpL-deficient patients were briefly treated with LpL and then added to the HDL and CETP-containing d 1.063 bottom fraction of control plasma, their capacity to engage in CET increased 6-fold (Fig. 2). In contrast, when chylomicrons from control subjects were treated in an identical manner, CET increased slightly above the stimulation observed with untreated chylomicrons (Fig. 3).

To determine whether other apoB-containing lipoproteins might also contribute to the reduced CET activity in LpL-deficient whole plasma, VLDL and LDL from patients and control subjects were isolated and added in separate experiments to the control d 1.063 bottom fractions (Fig. 4 and Fig. 5). In contrast to VLDL from normal subjects, which displayed a curvilinear increase in CET similar to that observed in the whole control plasma, VLDL from an LpL-deficient subject lacked the capacity to stimulate CE transfer from HDL. No difference in CET was found when LDL from an LpL-deficient and this normolipidemic control subject were combined with the control's d 1.063 g/ml fraction (data not shown).

Further experiments were carried out to assess the CET-promoting capacity of the HDL and the CETP-containing d 1.063 bottom fraction of LpL-deficient subjects (Fig. 5). Chylomicrons from a control subject stimulated the transfer of CE in a roughly dose-dependent manner from both his own and the LpL-deficient patient's d 1.063 bottom fractions at two different HDL concentrations, indicating that the functional capacity of this fraction was intact in LpL deficiency. CETP mass measurements in two of the LpL-deficient patients were slightly lower than those of the controls (LpL-deficient 1.03 ± 0.23 µg/mL vs. control 1.58 ± 0.58 µg/mL; mean ± SD).

The lipid composition of chylomicrons from the three LpL-deficient subjects and two healthy control subjects after a high-fat meal are shown in Table 2. Table 3 shows the lipid composition of lipoproteins from two of the LpL-deficient subjects and three hypertriglyceridemic subjects. Consistent with their LpL deficiency, the chylomicron fraction of LpL-deficient subjects contained higher triglyceride and cholesterol concentrations after an overnight fast than plasma from controls 3 h after a fat load. Chylomicrons from LpL-deficient subjects had higher TG/CE ratios (P < 0.1) and a lower proportion of esterified cholesterol to total cholesterol compared to control values but they also showed a trend toward a lower proportion of their total cholesterol to be present as esterified cholesterol (i.e., reduced CE/TG ratios). Measurements performed on the lipoprotein fractions

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**TABLE 1.** Plasma lipids in control subjects and LpL-deficient patients

<table>
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<tr>
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<th>Control</th>
<th>LpL-Deficient</th>
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<tbody>
<tr>
<td>TC mg/dl</td>
<td>80.6 ± 15.2</td>
<td>941 ± 88*</td>
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<tr>
<td>TG mg/dl</td>
<td>154 ± 20.1</td>
<td>155 ± 80.4</td>
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<tr>
<td>HDL-C mg/dl</td>
<td>47.2 ± 1.6</td>
<td>22.1 ± 3.3*</td>
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*P < 0.025.

*P < 0.001.
varied considerably. Nevertheless, a trend was present toward lower unesterified cholesterol/lecithin ratios in VLDL, LDL, and HDL₂ and higher TG/CE ratios in LDL and in the HDL subfractions in the LpL-deficient compared to either normolipidemic controls or the hypertriglyceridemic subjects.

**DISCUSSION**

Recent evidence suggests that the relationship between the actions of CETP and atherosclerosis is complex (13). Nevertheless, it appears that in a broad range of clinical conditions in which CET activity is increased, such as hypercholesterolemia (8), hypertriglyceridemia (14), dyslipidemia (15), and both insulin-dependent (16) and non-insulin-dependent diabetes mellitus (17), cardiovascular disease develops prematurely. Because of this association and the fact that Japanese families (18) and animal species that lack CETP (19) are resistant to atherosclerosis, the factors that regulate CETP have become a topic of major investigative interest.

If CET is linked to atherogenesis, we reasoned that it may be reduced in clinical conditions in which atherosclerosis is uncommon. Subjects with LpL deficiency have been observed to be relatively free of vascular disease despite the presence of abnormally high plasma triglycerides and often markedly reduced HDL levels (20). Their relative immunity to CHD has been ascribed to low LDL levels that result from their reduced capacity to convert VLDL to LDL. Low LDL levels may be a factor that reduces cardiovascular risk in LpL-deficient patients but, because of their elevated TG, low HDL, and recent evidence implicating families of TG-rich lipoproteins rather than LDL in the progression of atherosclerosis (21), they might be expected to be at high risk.

A number of lines of evidence demonstrate a direct association between CET and LpL, suggesting that CET might be reduced in LpL-deficient patients. First, Tall, Sammett, and Granot (22) showed that in vitro exposure of TG-rich lipoproteins to LpL promoted their interaction with CETP. Second, we have recently reported that when basal LpL activity was abnormally increased in the

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**Fig. 2.** Changes in the mass of esterified cholesterol in HDL present in the d > 1.063 g/ml fraction isolated from the fasting plasma from two normolipidemic subjects during a 6-h incubation after combination with chylomicrons in fasting plasma from two LpL-deficient patients treated with and without bovine milk lipase (1 nmol FA/min per ng enzyme for 30 min at 37°C).

**Fig. 3.** Changes in the mass of esterified cholesterol in HDL obtained from two normolipidemic controls after incubation of their d > 1.063 g/ml fractions with their post-fat load chylomicrons untreated or treated with LpL (see legend for Fig. 2).
plasma of conventionally treated IDDM patients as a consequence of systemic hyperinsulinemia, their CET was also elevated (23) as measured with the same in vitro method used in the present study. When their LpL levels were reduced by lowering insulin levels with intraperitoneal insulin delivery, CET activity fell to normal. Third, studies of Castro and Fielding (1) and Marcel et al. (9) together indicate that CETP action is maximal when LpL is active postprandially.

The fact that CET has been shown by Mann et al. (14) to correlate closely with the VLDL-TG concentration in subjects over a wide range of TG levels would lead one to expect that CET in LpL-deficient patients would be increased based on their elevated TG. However, we found that the net mass transfer of CE in their intact plasma was decreased. The combination of studies we performed with isolated lipoprotein fractions documents the central role played by LpL shown earlier in the activation of CETP. First, we found that CET activity in the plasma of these patients was reduced in the presence of elevated TG and normal CETP concentrations. Second, their d 1.063 bottom fraction containing HDL and CETP demonstrated a normal capacity to stimulate CET when combined with VLDL from control subjects, effectively ruling out these factors as Contributors to the attenuated CET response found in their plasma. Even though their total HDL levels were markedly reduced, we found that at the same cholesterol concentration their HDL transferred to control chylomicrons an amount of CE comparable to that transferred by control HDL. These observations indicated that their CE donor lipoproteins (i.e., HDL) and CETP functioned normally.

Our finding that VLDL from the LpL-deficient and control subjects showed a similar low-level capacity to stimulate net CE mass transfer implied that their apoB-containing TG-rich lipoproteins in general were dysfunctional. Additionally, the TG/CE ratio was higher in

Fig. 4. Changes in the mass of esterified cholesterol in HDL present in the d > 1.063 g/ml fraction of plasma from a normolipidemic subject during a 6-h incubation with his own VLDL (d < 1.006 g/ml) and VLDL obtained from an LpL-deficient subject as the d < 1.006 g/ml fraction after removal of chylomicron fraction by centrifugation at 30,000 rpm for 30 min at 8°C.

Fig. 5. Changes in the mass of esterified cholesterol in HDL present in d > 1.063 g/ml fractions of a control (top panel) and an LpL-deficient patient (bottom panel) at the same concentration of total HDL cholesterol (TC 15 mg/dl and 30 mg/dl) during a 6-h incubation with chylomicrons (6.4 mg/ml) from a normal subject after ingesting a high-fat meal.
TABLE 2. Lipid composition of chylomicrons from three LpL-deficient patients and two healthy controls after a high-fat meal

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Triglyceride</th>
<th>Cholesterol</th>
<th>Unesterified Cholesterol</th>
<th>Esterified Cholesterol</th>
<th>UC/Lecithin</th>
<th>TG/CE</th>
<th>EC/TC</th>
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<td>mg/dl</td>
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<td>LpL-deficient</td>
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<td>528</td>
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<td>50</td>
<td>19</td>
<td>0.86</td>
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<td>0.28</td>
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<td>435</td>
<td>54.6</td>
<td>42.0</td>
<td>12.6</td>
<td>0.47</td>
<td>34.5</td>
<td>0.23</td>
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<td>3</td>
<td>870</td>
<td>120</td>
<td>68</td>
<td>52</td>
<td>0.17</td>
<td>16.7</td>
<td>0.43</td>
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<tr>
<td>Mean ± SD</td>
<td>611 ± 229</td>
<td>81.2 ± 34.4</td>
<td>59.3 ± 13.3</td>
<td>27.9 ± 21.1</td>
<td>0.50 ± 0.35</td>
<td>26.3 ± 9.0</td>
<td>0.51 ± 0.10</td>
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<tr>
<td>Control</td>
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<td>142.5</td>
<td>47.2</td>
<td>21.0</td>
<td>26.2</td>
<td>0.05</td>
<td>5.4</td>
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<td>2</td>
<td>146.3</td>
<td>33.5</td>
<td>16.0</td>
<td>17.5</td>
<td>0.08</td>
<td>8.4</td>
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<tr>
<td>Mean ± SD</td>
<td>144.4 ± 2.7</td>
<td>40.4 ± 9.7</td>
<td>18.5 ± 3.5</td>
<td>21.9 ± 6.2</td>
<td>0.07 ± 0.02</td>
<td>6.9 ± 2.1</td>
<td>0.54 ± 0.03</td>
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chylomicrons obtained after an overnight fast from the LpL-deficient subjects than in chylomicrons from controls isolated only 3 h after ingestion of a fat load. This is precisely the type of difference to be expected in LpL-deficient chylomicrons when CET is low compared to chylomicrons obtained from controls when LpL and CET are both normally maximally active in the postprandial state. Moreover, the proportion of total cholesterol present as CE tended to be lower in LpL-deficient chylomicrons than in those of the controls. As CE in chylomicrons is derived primarily as a consequence of CET, the reduced content of CE in LpL-deficient chylomicrons is also consistent with decreased CET having taken place in vivo.

If failure of TG-rich lipoproteins to undergo LpL-mediated lipolysis in vivo was a factor contributing to low CET levels in LpL deficiency, then one would predict that exposing them to LpL in vitro would stimulate CET. Conversely, incubation of postprandial lipoproteins from controls under the same conditions should have little additional impact on their transfer activity because they had already been lipolyzed by LpL in vivo and, as a result, presumably contained sufficient concentrations of lipolytic products such as free fatty acids on their surface.

TABLE 3. Lipoprotein lipid composition in LpL-deficient patients and hypertriglyceridemic (HTG) subjects

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Triglyceride</th>
<th>Cholesterol</th>
<th>Unesterified Cholesterol</th>
<th>Esterified Cholesterol</th>
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<tr>
<td>Mean ± SD</td>
<td>297 ± 156</td>
<td>51.4 ± 8.9</td>
<td>27.7 ± 19</td>
<td>23.7 ± 10</td>
<td>0.88 ± 0.02</td>
<td>15.3 ± 15.1</td>
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<tr>
<td>HTG group</td>
<td>380 ± 244</td>
<td>84 ± 41</td>
<td>56.5 ± 24</td>
<td>28.1 ± 19.2</td>
<td>1.35 ± 0.18</td>
<td>16.3 ± 8.4</td>
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<td>Controls</td>
<td>44.6 ± 26.1</td>
<td>7.1 ± 6.8</td>
<td>4.8 ± 3.3</td>
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<tr>
<td>Mean ± SD</td>
<td>42.8 ± 6.7</td>
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<td>8.9 ± 2.1</td>
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<tr>
<td>HTG group</td>
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<td>117.2 ± 37.5</td>
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<td>22.5</td>
<td>18.8</td>
<td>13.8</td>
<td>13.8</td>
<td>0.99</td>
<td>14.7</td>
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<tr>
<td>Mean ± SD</td>
<td>20.5 ± 2.8</td>
<td>17.0 ± 2.6</td>
<td>10.6 ± 4.6</td>
<td>6.4 ± 7.2</td>
<td>0.85 ± 0.23</td>
<td>8.3 ± 9.0</td>
<td></td>
</tr>
<tr>
<td>HTG group</td>
<td>24.8 ± 10.3</td>
<td>44.0 ± 13.7</td>
<td>13.6 ± 5.1</td>
<td>30.4 ± 9.1</td>
<td>0.61 ± 0.32</td>
<td>0.80 ± 0.13</td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>9.4 ± 3.6</td>
<td>35.2 ± 8.4</td>
<td>9.0 ± 3.4</td>
<td>26.2 ± 8.9</td>
<td>0.43 ± 0.07</td>
<td>0.44 ± 0.31</td>
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</table>
surface to enhance their affinity for CETP. Indeed, as predicted, our recombination experiments demonstrated that chylomicrons and VLDL from LpL-deficient patients had a reduced capacity to engage in CET and to accept CE from HDL compared to these same lipoprotein fractions from controls. Moreover, this defect was fully corrected by contact with LpL.

Another possible interpretation of our finding low CET in the plasma of LpL-deficient subjects is that equilibration of core lipids between HDL and the apoB-containing lipoproteins VLDL and LDL had already taken place in vivo because of the prolonged residence time of the TG-rich lipoproteins. If this were true, then low levels of CET would be expected when lipoprotein-containing plasma from these subjects was examined in vitro. Our incubation studies with LpL, however, provided some evidence against this possibility. Specifically, if the core lipids of the lipoprotein particles present in the LpL-deficient chylomicron fractions had already equilibrated, one would not expect to find the increase we observed in CET after incubation with LpL. Rather, a negligible change would be anticipated as was found when chylomicrons from controls were treated similarly. While the compositional differences demonstrated between LpL-deficient and control chylomicrons were consistent with reduced CET, comparison of the relative amounts of TG and CE in lipoprotein core lipids of two of these subjects with those of hypertriglyceremic subjects was less conclusive because of the variability in the results observed. Patient 2, for example, showed a somewhat higher VLDL TG/CE ratio than that of the hypertriglyceremic subjects as might be expected if lipolysis and CET were decreased. In contrast, however, the other patient’s (patient 1) VLDL TG/CE was lower than that of the reference group suggesting that equilibration of VLDL and LDL core lipids had taken place, perhaps as a consequence of extended residence time. These findings indicate that lipid composition may vary among patients with LpL deficiency because of differences in other systems that regulate the content of lipoproteins, such as hepatic lipase, lecithin:cholesterol acyltransferase, and other transfer proteins. In patients with CETP deficiency, HDL-C levels are higher than normal (18). That HDL-C levels are low rather than high in the LpL-deficient patients in the present study probably reflects the fact that the delipidation and catabolism of TG-rich lipoproteins with attendant translocation of apoproteins and phospholipids is a prerequisite for maintaining normal HDL levels and transport.

Our finding that the mass of TG present in HDL3 in the two LpL-deficient patients exceeded that of CE is consistent with CE–TG reciprocal exchange having already occurred in vivo during the prolonged residence time of the TG-rich lipoproteins in the plasma compartment in the absence of LpL. While the elevated TG/CE ratios in LDL are consistent with this possibility, one would expect to have evidence of reciprocal exchange in their VLDL and this was not observed.

Our findings in intact plasma from patients with LpL deficiency complement a series of earlier observations by Castro and Fielding (1) and from Tall’s laboratory (4, 22) in which the central role of LpL in the activation of CET was first demonstrated in a defined system containing treated and untreated VLDL, HDL, and CETP. Data in the present study from patients with complete LpL deficiency provide evidence that supports the functional relationship between CET and LpL observed in these previous in vitro studies. Moreover, our findings implying that LpL functions as an activator of CET in vivo is consistent with physiologic observations reported earlier by Castro and Fielding (1) showing that CET was stimulated during the postprandial state at a time when the activity of LpL would be maximal. It remains to be determined, however, whether variations in LpL activity that occur under a variety of metabolic conditions (24) are associated with disordered CET. Such information would be required in order to determine whether LpL has a true regulatory role or functions in a more limited way as an activator of CET.

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