Very-Low-Density Lipoprotein Subfraction Composition and Metabolism by Adipose Tissue

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Lipoprotein lipase (LPL) plays a pivotal role in very-low-density lipoprotein (VLDL) metabolism. Within the circulation, the VLDL population is heterogeneous with respect to both size and composition. Several studies have investigated the action of LPL in vitro on different VLDL subfractions, but little is known of the action of LPL in vivo. To investigate this, arterial and adipose tissue venous plasma samples were obtained from 16 normal male healthy volunteers (aged 24.4 ± 1.8 years; body mass index, 23.5 ± 0.7 kg · m^{-2}) following an overnight fast. VLDL subfractions were isolated ($VLDL_1$ of S_f 60 to 400 and $VLDL_2$ of S_f 20 to 60) and characterized in terms of triacylglycerol (TAG) and apolipoprotein (apo) B, E, CI, CII, and CIII content. The apolipoprotein content of $VLDL_1$ differed from that of $VLDL_2$: the $VLDL_2$ fraction contained significantly more apo B (0.018 ± 0.004 *v* 0.011 ± 0.003 μmol · L^{-1}, $P = .001$) but the ratios of TAG:apo B and apo CI:B, CII:B, and CIII:B were significantly higher in $VLDL_1$ (48,200 ± 7,960 *v* 13,860 ± 2,420, 22.7 ± 5.5 *v* 12.5 ± 2.2, 45.0 ± 6.3 *v* 14.9 ± 2.0, and 0.434 ± 0.077 *v* 0.357 ± 0.054, respectively, molar ratios, all $P < .05$). The venous blood draining an adipose tissue depot contained less $VLDL_1$-TAG than arterial blood (328 ± 68 *v* 381 ± 83 μmol · L^{-1}, respectively, $P < .01$), whereas $VLDL_2$-TAG exhibited an opposite tendency (199 ± 46 *v* 172 ± 31 μmol · L^{-1}, NS). Concentrations of $VLDL_1$-apo B, -apo CII, and -apo CIII were significantly less in adipose tissue venous blood compared with arterial blood (0.011 ± 0.004 *v* 0.013 ± 0.004, 0.38 ± 0.08 *v* 0.43 ± 0.10, and 1.33 ± 0.35 *v* 1.58 ± 0.38 μmol · L^{-1}, respectively, all $P < .05$). These studies demonstrated novel differences in $VLDL_1$ and $VLDL_2$ in terms of composition and metabolism by human adipose tissue LPL in vivo.

VERY-LOW-DENSITY LIPOPROTEINS (VLDLs) are the major carriers of triacylglycerol (TAG) in the blood and are a particularly important source of TAG in the postabsorptive state, during which few chylomicrons are present. VLDLs are synthesized mainly in the liver and secreted into the circulation, permitting transport of TAG to peripheral tissues.[1] It is now known that within VLDLs there are further divisions of the particles into metabolically distinct groups.[2,3] Hence VLDLs should be considered a heterogeneous population in at least some respects.

The metabolic fate of a lipoprotein is thought to be largely determined by its composition, in particular its apolipoprotein composition, since these proteins have several regulatory functions. Apolipoproteins regulate the metabolism of lipoproteins by modulating their interaction with specific receptors, or by regulating the activity of enzymes such as lipoprotein lipase (LPL).[4] Differences in the apolipoprotein composition of VLDL subfractions could determine their differing metabolism.[5,6]

Lipoprotein arteriovenous differences across muscle tissue were measured by Kiens and Lithell,[7] and Rössner et al[3] have measured the removal of an artificial lipid emulsion by both muscle and subcutaneous tissues. Such techniques provide an in vivo bioassay for LPL by measuring TAG clearance of the tissue in question.[9] In this study, arteriovenous differences across adipose tissue have been measured. LPL is found abundantly in the capillaries of adipose tissue,[10] which is rich in LPL action and is known to clear VLDL-TAG.[11,12] An advantage of studying adipose tissue versus muscle is that larger TAG arteriovenous differences are obtained across the former because of the greater LPL activity.[13] Adipose tissue is not thought to synthesize VLDL, and thus the clearance of VLDL particles across the adipose tissue capillary bed is a direct reflection of LPL action on these particles. We have previously reported that there is no net difference in whole plasma apolipoprotein (apo) A and B concentrations across adipose tissue.[14]

The aims of the present study were to investigate some of the differences in apolipoprotein composition and metabolism of VLDL subfractions in a group of normal healthy males following a fasting period of 11.5 to 14.5 hours. The composition was defined in terms of TAG and apo B, E, CI, CII, and CIII. Local clearance of VLDL subfractions was studied using an arteriovenous difference method.

SUBJECTS AND METHODS

Subjects

Sixteen healthy male subjects were studied (aged 24.4 ± 1.8 years; body mass index, 23.5 ± 0.7 kg · m^{-2}; body fat, 17.8% ± 1.8%). The mean fasting plasma TAG concentration was 1,090 ± 116 μmol · L^{-1} (this represents the mean concentration of samples drawn at 9:30 and 10:30 AM). Two subjects habitually smoked 15 to 20 cigarettes per day before the study. The results were recalculated excluding these two smokers, but were found to be similar. Therefore, results from these individuals were included in the final analyses.

This study was approved by the Institutional Review Board of the Mayo Clinic, and all subjects provided informed consent.

Experimental Design

All subjects were admitted to the Mayo Clinic General Clinical Research Center on the afternoon before the study. Body composition was determined using a dual-energy x-ray absorptiometer (Lunar, Madison, WI). At 6:00 PM, the subjects ate a standard meal of 20 kcal · kg lean body mass $(LBM)^{-1}$ with 50% of calories as carbohydrate, 30% as fat, and 20% as protein. At 10:00 PM, the subjects ate a fat-free snack containing 5 kcal · kg LBM^{-1}. From the time of

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admission, subjects refrained from smoking and drank only water or caffeine-free diet drinks.

Beginning at 7:00 AM the following morning, blood sampling cannulae were inserted (using local anesthesia) into a radial artery in all subjects and a superficial abdominal vein[15] in 11 subjects, which were kept patent by a slow infusion of isotonic saline (150 mmol · L^{-1} NaCl).

Postabsorptive blood samples were drawn from both lines at 9:30 and 10:30 AM (ie, at 11.5 and 12.5 hours of fasting and at 15.5 and 16.5 hours after the last fat intake). Arterial samples alone were taken at 11:30 AM and 12:30 PM (ie, at 13.5 and 14.5 hours of fasting).

Analytical Methods

Blood samples were taken into syringes containing EDTA 1 mg/mL blood (Monovette; Sarstedt, Princeton, NJ).

Lipoprotein isolations were performed by cumulative flotation ultracentrifugation using a method previously described.[6] In brief lipoproteins were isolated from 1 mL plasma (adjusted to a density of 1.118 kg · L^{-1} by the addition of solid NaCl) in a Beckman SW 55Ti rotor (r_{max}, 108.5 mm; r_{min}, 60.8 mm) using a discontinuous salt gradient from density 1.0988 to 1.0588 kg · L^{-1} (NaBr/NaCl solutions). Successive centrifugations were performed at 23°C, the first to remove any chylomicron-like particles ($S_f > 400$) and the second and third to isolate $VLDL_1$ (S_f 60 to 400) and $VLDL_2$ (S_f 20 to 60), respectively. The conditions of isolation were calculated in terms of ω^2t values. Successive ω^2t values were 8.8×10^9, 4.7×10^{10}, and 9.8×10^{10} for S_f greater than 400, S_f 60 to 400, and S_f 20 to 60, respectively. These values are for the start of rotor rotation to the rotor coming to rest (deceleration without braking), and the correction for previous centrifugations was taken into account when the conditions were calculated for S_f 60 to 400 and S_f 20 to 60. Each fraction was harvested by aspiration from the top of the tube.

TAG concentrations were measured by an enzymatic colorimetric method with correction for free glycerol, adapted to a COBAS Mira Chemistry System (Roche Diagnostic Systems, Montclair, NJ).[16] Apo E, CI, CII, and CIII concentrations were estimated by radioimmunoassay,[17,18] and apo B by a particle concentration fluorescence immunoassay.[19] Apo CI estimates were available in 11 subjects only.

Intraassay coefficients of variation (CVs) for the TAG, apo B, and apo E assays were 2.2%, 3.9% to 7.6%, and 8.9%, respectively. For apo CI, apo CII, and apo CIII, intraassay CVs were 2.4% to 13.5%. Sensitivities of the TAG, apo B, and apo E assays were 10 μmol · L^{-1}, 0.27 mg · L^{-1}, and 17 μg · L^{-1}, respectively. The sensitivity for apo CI and apo CII assays was 6.25 μg · L^{-1}, and for apo CIII 13.2 μg · L^{-1}. All values in Tables 1 and 2 represent the mean of two samples, each of which was assayed in duplicate.

Calculations and Statistical Analysis

Each VLDL fraction was weighed and the volume estimated by assuming a density of 1.0588 kg · L^{-1}. The fraction volume was then used to calculate TAG and apolipoprotein equivalent concentrations in plasma. Apolipoprotein molar concentrations were calculated from the measured concentrations in milligrams per deciliter using molecular masses of 549,000 for apo B-100,[20] 6,600 for apo CI, 8,820 for apo CII, 8,750 for apo CIII, and 34,150 for apo E.[1]

Data are presented as the mean ± SEM. Statistical tests of significance to compare different groups were performed using nonparametric statistical methods (Wilcoxon's test). Changes with time were assessed by ANOVA.

RESULTS

Changes in TAG Concentration of VLDL Subfractions

The arterial $VLDL_1$-TAG concentration decreased by 20% ± 5% as the subjects' fasting period increased from 11.5 to 14.5 hours ($P < .005$), but there was no significant change in the arterial $VLDL_2$-TAG concentration (Fig 1). Insulin concentrations were 45.6 ± 4.6 and 40.2 ± 5.8 pmol · L^{-1} after 11.5 and 14.5 hours of fasting, respectively.

Composition of VLDL Subfractions

In arterial plasma, concentrations of TAG and apo CII and CIII within the whole $VLDL_1$ subfraction were greater than those within the $VLDL_2$ subfraction, whereas apo B and E concentrations were higher within the $VLDL_2$ fraction. There was no difference in the amount of apo CI in the two subfractions. Assuming there is a single molecule of apo B per VLDL particle,[21] the apo B results confirmed that there were greater numbers of $VLDL_2$ than $VLDL_1$ particles.

The ratios of TAG and other apolipoproteins to apo B are a measure of the relative composition of individual particles of $VLDL_1$ and $VLDL_2$. $VLDL_1$ particles were shown to be more TAG-rich than $VLDL_2$ because they had a higher TAG:apo B ratio. There was no significant difference between the apo E content of $VLDL_1$ and $VLDL_2$, but per particle, $VLDL_1$ contained more apo CI, CII, and CIII than $VLDL_2$. Apo CII:CIII and CIII:E ratios were significantly higher in $VLDL_1$ (Table 1).

Arteriovenous Differences for VLDL Subfractions Across Adipose Tissue

There was significant removal of TAG and apo B, CII, and CIII from the $VLDL_1$ subfraction by adipose tissue, but there were no changes in these concentrations in the $VLDL_2$ subfraction. In a single passage of the adipose tissue, 13.2% ± 2.6% and 18.8% ± 4.5% of TAG and apo B, respectively, were removed from arterial $VLDL_1$. Apo E increased in $VLDL_2$ during passage through the adipose tissue, but did not change significantly in $VLDL_1$. Apo CI concentrations showed no change in either subfraction.

Apo CI:B and CII:B ratios increased significantly in $VLDL_1$ and the apo CII:CIII ratio increased in both $VLDL_1$ and $VLDL_2$ in adipose tissue venous plasma compared with arterial samples. The apo CIII:E ratio in $VLDL_2$ decreased on passage through the adipose tissue (Table 2).

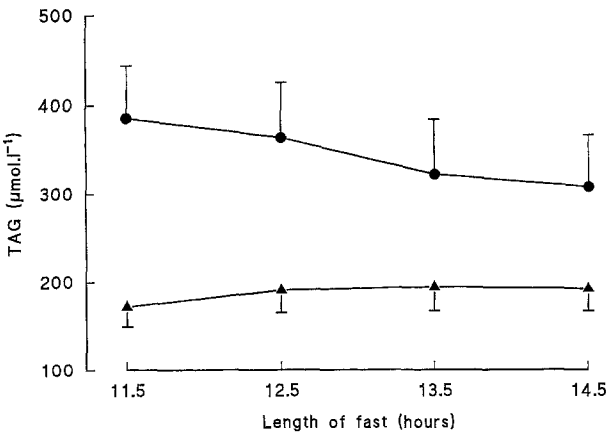

Fig 1. Arterial TAG concentrations in plasma of $VLDL_1$ (●) and $VLDL_2$ (▲) by length of fast. Results are the mean ± SEM for 16 subjects.

Table 1. Comparison of the Composition of $VLDL_1$ and $VLDL_2$

Parameter	$VLDL_1$	$VLDL_2$	P
TAG	375 ± 60	181 ± 24	<.001
Apo B	0.011 ± 0.003	0.018 ± 0.004	.001
Apo E	0.124 ± 0.019	0.176 ± 0.022	<.05
Apo CI	0.220 ± 0.051	0.218 ± 0.055	NS
Apo CII	0.383 ± 0.068	0.213 ± 0.030	<.001
Apo CIII	1.302 ± 0.287	0.882 ± 0.215	<.005
TAG:apo B	48,200 ± 7,960	13,860 ± 2,420	<.001
Apo E:B	16.7 ± 2.9	13.0 ± 1.7	NS
Apo CI:B	22.7 ± 5.5	12.5 ± 2.2	<.05
Apo CII:B	45.0 ± 6.3	14.9 ± 2.0	<.001
Apo CIII:B	150.3 ± 43.9	54.4 ± 13.4	<.001
Apo CII:CIII	0.434 ± 0.077	0.357 ± 0.054	<.05
Apo CIII:E	10.7 ± 1.5	4.9 ± 0.8	<.001

NOTE. Results are the mean ± SEM of paired arterial samples taken after 11.5 and 12.5 hours of fasting from 16 subjects (results only available from 11 subjects for apo CI). Concentrations are expressed as μmol · L^{-1}, and ratios are molar ratios.

DISCUSSION

VLDLs can be subfractionated on the basis of size and density, but functional differences between large and small VLDLs have not been clearly defined. It has been suggested that VLDLs of different sizes have different metabolic roles and fates, although it is unclear how these differences are achieved. Large VLDL ($VLDL_1$) particles are hydrolyzed rapidly by LPL to produce remnant particles,[3] many of which are removed directly from the circulation and make no contribution to intermediate-density lipoprotein (IDL) and low-density lipoprotein (LDL).[22] The fraction of smaller VLDL ($VLDL_2$) particles contains both $VLDL_1$ remnants and particles synthesized de novo by the liver.[3] Some $VLDL_2$ particles are converted to IDL and LDL, whereas others are removed by receptor-mediated uptake.[2]

In this study, $VLDL_1$-TAG concentration decreased as the length of the fast increased from 11.5 to 14.5 hours, but there was no equivalent change in $VLDL_2$-TAG concentration. This implies either that $VLDL_1$ was the favored TAG supply to tissues or that there was a decrease in $VLDL_1$ production relative to that of $VLDL_2$. Total VLDL-TAG has been shown to decrease over an equivalent period, [23] but this decrease has not previously been attributed to the large VLDL particle fraction.[24] The continued decrease in TAG has relevance to clinical assessments made following a 12- to 16-hour fast, as recommended by the Lipid Research Clinics Program.[25]

The distinct chemical properties of large and small VLDLs were investigated in the present study by characterizing apo B, CI, CII, CIII, and E in $VLDL_1$ and $VLDL_2$. The C apolipoproteins of VLDL are transferred to high-density lipoprotein (HDL) particles as TAG hydrolysis of the lipoprotein core takes place,[26,27] along with apo E,[28,29] but the apo B moiety is integral to the lipoprotein particle and cannot be transferred.[30] During alimentary lipemia, there is transfer of the C apolipoproteins from HDL into newly secreted chylomicrons.[31] Although the present data do not allow determination of the relative rates of clearance of $VLDL_1$ and $VLDL_2$, other reports have shown large VLDLs to be hydrolyzed more rapidly by LPL than smaller VLDLs both in vitro[32,33] and in vivo.[34-36]

Several differences in the composition of $VLDL_1$ and $VLDL_2$ were observed that could explain a higher clearance rate of $VLDL_1$ by LPL (Table 1). The TAG:apo B ratio was greater in $VLDL_1$ than in $VLDL_2$, reflecting the larger size of $VLDL_1$ particles. Some suggest that it is the ratio of surface to core components of VLDL particles rather than the apolipoprotein composition or lipoprotein surface area that accounts for the preference of LPL for larger $VLDL_1$ particles as substrates.[37] Apo CII is an essential cofactor for LPL.[38] The larger number of apo CII molecules found in $VLDL_1$ particles compared with $VLDL_2$ would promote the action of LPL on $VLDL_1$. The role of apo CIII is not quite so clear, but it has been proposed to have an inhibitory effect on the action of LPL.[39,40] Although $VLDL_1$ particles contained more CIII than $VLDL_2$, the apo CII:CIII ratio was higher in $VLDL_1$. Diet-induced changes in plasma TAG concentrations have been associated with changes in the VLDL apo CII:CIII ratio, suggesting that these changes may account, in part, for the regulation of plasma TAG concentrations.[41] Furthermore, the VLDL apo CII:CIII ratio has been found to decrease as both plasma TAG[42] and VLDL-TAG[43,44] concentrations increase, suggesting LPL action may be reduced as the apo CII:CIII ratio decreases. However, the importance of

Table 2. Arteriovenous Differences for $VLDL_1$ and $VLDL_2$

Parameter	$VLDL_1$			$VLDL_2$		
	Arterial	Adipose Tissue Venous	P	Arterial	Adipose Tissue Venous	P
TAG	381 ± 83	328 ± 68	<.01	172 ± 31	199 ± 46	NS
Apo B	0.013 ± 0.004	0.011 ± 0.004	<.005	0.022 ± 0.006	0.022 ± 0.006	NS
Apo E	0.140 ± 0.026	0.137 ± 0.024	NS	0.192 ± 0.031	0.224 ± 0.040	<.01
Apo CI	0.244 ± 0.064	0.227 ± 0.057	NS	0.246 ± 0.071	0.255 ± 0.071	NS
Apo CII	0.425 ± 0.096	0.378 ± 0.081	<.05	0.222 ± 0.042	0.231 ± 0.058	NS
Apo CIII	1.579 ± 0.382	1.329 ± 0.348	<.05	1.059 ± 0.295	0.971 ± 0.316	NS
TAG:apo B	41,500 ± 7,230	43,510 ± 7,160	NS	10,920 ± 2,350	11,920 ± 2,180	NS
Apo E:B	17.7 ± 4.2	21.5 ± 4.6	NS	12.1 ± 2.0	14.3 ± 2.4	<.05
Apo CI:B	24.2 ± 7.3	29.5 ± 9.3	<.05	12.7 ± 2.9	13.7 ± 3.5	NS
Apo CII:B	44.4 ± 9.0	49.8 ± 9.5	<.05	13.4 ± 2.8	12.9 ± 2.0	NS
Apo CIII:B	172.4 ± 62.6	177.2 ± 56.0	NS	58.2 ± 19.0	52.1 ± 16.9	NS
Apo CII:CIII	0.343 ± 0.060	0.377 ± 0.065	<.05	0.293 ± 0.049	0.335 ± 0.054	<.05
Apo CIII:E	11.9 ± 1.9	10.5 ± 2.1	NS	5.4 ± 1.1	4.2 ± 0.9	.01

NOTE. Results are the mean ± SEM of paired samples taken after 11.5 and 12.5 hours of fasting from 11 subjects (results only available from 8 subjects for apo CI). Concentrations are expressed as μmol · L^{-1}, and ratios are molar ratios.

the apo CII:CIII ratio in regulating the lipolysis of different VLDL particles has been called into question in an in vitro incubation system.[45] Apo CIII may also decrease the receptor-mediated removal of lipoprotein particles from the circulation by reducing their apo E content.[46] Our results showed the apo CIII:E ratio to be higher in $VLDL_1$ than in $VLDL_2$ particles. The amount of apo CI per particle was greater in $VLDL_1$ than in $VLDL_2$; apo CI has been proposed to inhibit the binding of VLDL remnants to the LDL receptor.[47] These apolipoprotein differences would favor a more efficient removal of (at least some) $VLDL_2$ particles by receptor-mediated mechanisms, while $VLDL_1$ particles remain in the circulation for hydrolysis by LPL and delivery of TAG to the periphery.

There are previous reports of TAG and total protein composition of VLDL subfractions,[48-51] but few report the complete apolipoprotein composition.[52,53] In agreement with the present results, Karpe et al[53] reported higher concentrations of apo CII in $VLDL_1$ compared with $VLDL_2$ in healthy subjects in the fasting state, similar concentrations of apo CI in the two fractions, and higher concentrations of apo E in $VLDL_2$ compared with $VLDL_1$. In that study, apo CIII concentrations were higher in $VLDL_1$ than in $VLDL_2$ in the post prandial period, but were not significantly different in the fasting state.[53] In an earlier report, Kane et al[52] isolated five VLDL subfractions from normolipidemic subjects and examined the relative proportions of apo R-serine (apo CI), R-glutamine (apo CII), and R-alanine (apo CIII) and the arginine-rich polypeptide (apo E) within each subfraction. Proportions of apo CI increased with increasing density, whereas those of apo CII decreased. There was no discernible trend in the percentages of apo CIII and apo E in the different VLDL subfractions. Carlson and Ballantyne[44] also examined the relative proportions of apo CII and apo CIII in VLDLs of S_f 20 to 60, S_f 60 to 100, and S_f 100 to 400. In healthy subjects, the proportion of apo CII decreased with particle size, as did the CII:CIII ratio—findings similar to those presented here.

The rationales for using arteriovenous differences to study the metabolic actions of a tissue are well recognized.[7,8,15,54] We used the arteriovenous-difference method across subcutaneous adipose tissue, a tissue rich in LPL, to study the action of LPL on $VLDL_1$ and $VLDL_2$ in vivo. This technique allows study of the action of LPL alone on its substrates without the confounding action of hepatic lipase or the uptake of lipoprotein particles by receptor-mediated mechanisms. The insulin concentrations observed were typical for subjects after an overnight fast. Plasma insulin concentrations regulate LPL,[55] and both plasma insulin concentrations and adipose tissue LPL action are lower postabsorptively than postprandially.[56] Changes in $VLDL_1$ composition across the LPL-rich capillary bed were marked and relatively easy to interpret in this study. However, the interpretation of changes in $VLDL_2$ composition is more difficult, since TAG clearance from larger $VLDL_1$ particles is expected to yield particles in the $VLDL_2$ density interval in venous samples. It is not known what proportion of arterial $VLDL_1$ particles is converted within the adipose tissue capillaries into venous $VLDL_2$ and what proportion is converted directly into venous lipoprotein particles of a greater density (IDL or LDL). Thus $VLDL_2$ arteriovenous differences are the net result of clearance of $VLDL_2$ from the arterial blood and the addition of $VLDL_1$ remnants.

Substantial proportions of TAG and apo B were removed from the $VLDL_1$ fraction by adipose tissue, whereas $VLDL_2$-TAG increased across the tissue (not significant). This can be explained by LPL hydrolysis of $VLDL_1$-TAG generating $VLDL_1$ remnant particles that were isolated in the $VLDL_2$ fraction of the venous plasma. The lack of significant net changes in $VLDL_2$-apo B and -TAG concentrations indicates that the rate of clearance of arterial $VLDL_2$ did not exceed the rate of $VLDL_2$ production as a result of LPL-mediated hydrolysis of $VLDL_1$. There was significant removal of apo CII and CIII from $VLDL_1$ across the adipose tissue. There are at least two possible explanations for this. Firstly, the loss of apo C from $VLDL_1$ across this tissue bed was of the same order of magnitude as the removal of TAG (as previously observed in vitro for apo CII and apo CIII[45]), which might suggest that particles were moving into the $VLDL_2$ fraction. Secondly, the apparent loss of apo CII and CIII might have been due to their transfer to HDL.[30,57]

In contrast to $VLDL_1$, the net effect of passage through adipose tissue was much smaller on the $VLDL_2$ subfraction. However, adipose tissue venous plasma apparently contained more apo E in $VLDL_2$ than arterial plasma. This finding was interesting given that there was no significant decrease in $VLDL_1$-apo E across the adipose tissue to accompany the change in $VLDL_2$. There are three possible explanations for this observation. Firstly, within the radioimmunoassay, there may have been an increased antibody access to apo E epitopes following delipidation of the VLDL particle by LPL, similar to that observed in hepatic lipase–treated chylomicron remnants.[58] Differences in either the primary structure or the lipid-protein composition of a lipoprotein can cause heterogeneous expression of apolipoprotein epitopes.[59] TAG-depleted products of $VLDL_1$ hydrolysis, which had increased apo E epitope expression, could have caused the observed increase in $VLDL_2$-apo E following passage through the fat depot. However, this explanation is unlikely, since the assay used for apo E estimation[17] was modified to include detergents that were expected to delipidate and expose all apo E binding sites. Secondly, apo E may have been transferred from HDL to the VLDL fraction, a phenomenon recognized in the postprandial state[53,57] and also during the action of LPL in vitro.[60] Thirdly, de novo synthesis of apo E occurs in most organs by a variety of cell types, including macrophages,[61,62] and thus apo E synthesized within adipose tissue (or its blood vessels) may have been added to VLDL particles during their passage through this tissue bed.

In conclusion, our studies in a group of healthy men demonstrated novel differences in $VLDL_1$ and $VLDL_2$ in terms of composition and local metabolism in vivo. Per particle, $VLDL_1$ contained more apo CI, CII, and CIII than $VLDL_2$, and the apo CII:CIII ratio was higher. Alterations in the $VLDL_1$ subfraction following a single pass through adipose tissue were substantially greater than those observed for $VLDL_2$.

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