The plasma lipoproteins are defined by their density or electrophoretic properties, but within each class there exists a heterogeneous array of subclasses with varying lipid and apolipoprotein compositions. The plasma apolipoproteins may play important roles in contributing to this lipoprotein heterogeneity. Distinct high density lipoprotein (HDL) subpopulations exist which contain apo A-I with or without a fixed complement of apo A-II, and the relative proportions of these populations can change with metabolic perturbations. Very low density lipoproteins (VLDL) are also heterogeneous regarding the presence of large or small forms of apo B. Using agarose column chromatography, we and others have described the existence of discrete subclasses of lipoprotein particles enriched in apo E. The apo E-enriched fractions in VLDL appear to be metabolically related to those of intermediate and high density lipoprotein (HDL) size. Using different techniques, others have also described HDL subfractions containing apo E. By a variety of criteria, these apo E-containing HDL are larger and less dense than the bulk of HDL and in vitro are related to the activity of lecithin cholesterol acyl transferase (LCAT). Studies with cholesterol-fed animals have shown the formation of a cholesterol ester-enriched HDL particle containing apo E as its sole apolipoprotein. In vitro a similar apo E-rich particle has been induced by incubation of HDL with exogenous cholesterol. The existence in human plasma of lipoprotein subclasses containing only apo E has not been demonstrated; characterization of apo E-enriched subclasses has been hindered by
The difficulty of isolating them from the quantitatively dominant lipoproteins containing apo B and apo A-I. The present study has focused on these apo E-containing subfractions and has taken advantage of three clinical syndromes in which one or more of the quantitatively major apolipoproteins are absent or deficient. The first syndrome, abetalipoproteinemia, is characterized by an absence of apo B-containing chylomicrons, VLDL, and low density lipoproteins (LDL). These patients, however, are able to regulate cholesterol synthesis by virtue of apo E-containing lipoproteins in the HDL density range. The second syndrome, Tangier disease, is characterized by very low levels of traditional HDL and by the major apoproteins, apo A-I and apo A-II. The HDL, which can be isolated between 1.063–1.21 g/ml, appears to be relatively enriched in apo A-II at the expense of the apo C peptides and apo A-I. In this syndrome, the lipoprotein distribution of apo E has not been reported. In a third syndrome, there is a near absence of HDL and apo A-I or apo C-III. Despite the extremely low levels of HDL cholesterol in these patients, gradient gel electrophoresis has recently been used to identify subclasses of HDL enriched in apo E and apo A-II. The present report shows that in all three syndromes, there is an apo E-enriched subpopulation of lipoproteins the size of LDL despite the absence of the major apolipoproteins B or A-I. This subpopulation of HDL is approximately the same size as in normal subjects.

**Methods**

**Subjects**

Subject 1 (24 years old) and Subject 2 (30 years old) were patients being treated for abetalipoproteinemia. As shown in Table 1, both had the low plasma cholesterol and triglyceride levels characteristic of abetalipoproteinemia. Both had apo B plasma levels that were at the limits of detection of our assay, confirming the diagnosis of abetalipoproteinemia. Subjects 3 and 4 were two patients diagnosed as homozygous for Tangier disease. Subject 4, whose samples were obtained with the assistance of Dr. Ernst Schaefer, has been extensively studied. Subject 5 (age 32) and Subject 6 (age 34) are sisters. The clinical features of their disease have been detailed elsewhere. Briefly, both patients had evidence of premature atherosclerosis with xanthomata and corneal clouding. Lipid-laden histiocytes were present in a perivascular location. The most obvious lipid abnormality was a severe HDL deficiency. The earlier detailed study attributed this lipid abnormality to an absence of plasma apo A-I and apo C-III. All six subjects, as well as the normolipoproteinemic control subject, were fasting when studied.

**Apolipoprotein Quantitation**

Apolipoproteins E, B, A-I, C-II, and C-III were measured by specific radioimmunoassays (RIA). Details of the RIA have been reported elsewhere for apo E, B, A-I, and C-III. The procedure for the apo C-II RIA is analogous to that for apo C-III in terms of the buffer components and mode of separation of bound from free antigen, but has not been previously detailed. In this procedure, a buffer of 0.1 M borate, 0.01% Triton X-100, 0.1% ovalbumin, and Trasylol (50 KIU/ml; pH 8.5) was used throughout. Bound antigen was separated from free by adding 10 μl of goat anti-rabbit serum 48 hours after the primary antibody incubation, followed by adding polyethylene glycol 6000 24 hours later to give a final concentration of 10%. The apo C-II used for tracer, standard, and immunization was purified from VLDL by delipidation, gel filtration, and ion-exchange chromatography as described for apo C-III. Subsequently, the apo C-III, which inevitably contaminated the apo C-II after ion-exchange chromatography, was removed by immunoaffinity chromatography on anti-apo C-III IgG Sepharose CL-4B.

This RIA has a working range of 0.10 to 10 ng apo C-II and has been validated by the same criteria previously applied to the radioimmunoassays for apo A-I, B, E, and C-III. Specifically, when five normoli-

**Table 1. Levels of Cholesterol, Triglyceride, and Apolipoprotein in Six Subjects**

<table>
<thead>
<tr>
<th>Subject</th>
<th>Sex</th>
<th>Cholesterol (mg/dl)</th>
<th>TG (mg/dl)</th>
<th>Apo B (μg/ml)</th>
<th>Apo Al (μg/ml)</th>
<th>Apo E (μg/ml)</th>
<th>Apo C-III (μg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abetalipoproteinemia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>F</td>
<td>28.0</td>
<td>ND</td>
<td>2.2</td>
<td>496</td>
<td>52.4</td>
<td>22.0</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>37.4</td>
<td>ND</td>
<td>4.8</td>
<td>823</td>
<td>59.2</td>
<td>18.0</td>
</tr>
<tr>
<td>Tangier</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>161</td>
<td>136</td>
<td>772.0</td>
<td>11.9</td>
<td>81.0</td>
<td>71.1</td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>50.0</td>
<td>262</td>
<td>829.0</td>
<td>10.6</td>
<td>29.7</td>
<td>52.5</td>
</tr>
<tr>
<td>AI/CIII-deficiency</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>F</td>
<td>111</td>
<td>31</td>
<td>768.0</td>
<td>0.008</td>
<td>48.3</td>
<td>0.005</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>108</td>
<td>38</td>
<td>913.0</td>
<td>0.009</td>
<td>39.9</td>
<td>0.005</td>
</tr>
<tr>
<td>Normal</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

ND = Not detectable.

*Mean ± SD of 108 male subjects.
CL-4B as described elsewhere. The specific goat anti-apo E IgG was coupled at an antigen density of 1.7 mg/ml gel. This coupling was performed at pH 8.3 after activation of the Sepharose with CNBr. For preparation of the anti-apo E immunoadsorbent, anti-apo E IgG was coupled at pH 6.5 to Sepharose CL-4B, as described elsewhere.

For isolation of apo E-containing lipoproteins, a sample was routinely applied at a slow rate (less than 8 ml/hour) to a quantity of gel sufficient to bind all of the apo E in the test sample. After a sample application, the gel was washed extensively with phosphate-buffered saline (PBS), and the nonspecifically bound material was eluted with 0.5 M sodium thiosulfate and immediately dialyzed against 100 volumes of PBS or 5 mM ammonium bicarbonate.

Results

Normal Subjects

When plasma is directly applied to 4% agarose columns, the plasma lipoproteins are well separated, as shown by the triglyceride and cholesterol profiles of the fasting normolipoproteinemic subject in Figure 1 A. As described previously, the plasma cholesterol describes two well-defined peaks, corresponding to LDL and HDL. In the fasting sample shown in Figure 1, the triglyceride eluted as a single broad peak representing VLDL. Apo B, the major apolipoprotein of plasma LDL, coeluted with the major cholesterol peak, confirming its identity with LDL. A small fraction of apo B also eluted in the VLDL region, but in normal fasting subjects this represented only about 0.1% of that in the LDL region, so that it is not seen on the scale of measurement in Figure 1. Apo A-I, the major apolipoprotein of HDL, eluted as a peak nearly coincident with, but slightly displaced from, the second cholesterol peak.

In contrast, both apo E and apo C-III eluted in complex profiles that were displaced from each other and from the distribution of the major apolipoproteins (Figure 1 B). Both apo E and apo C-III were found in the broad distribution of particles the size of VLDL. The second and third fractions, however, were not coincident with LDL and HDL. The apo E subfractions were more clearly separated in size from LDL and HDL than were the apo C-III fractions. One fraction of apo E, designated fraction II, has been shown to be of intermediate size and density relative to VLDL and LDL. The other fraction (fraction III) was the size of large HDL and had the density characteristics of HDL1 and HDL2. Although the distribution of apo E among these three fractions differs in normal subjects, the size of each fraction is constant relative to VLDL, LDL, and HDL. Particles containing apo C-III were also found in a size range slightly larger than LDL, but smaller than apo E-containing fraction II, and in another range that was intermediate in size between the peak of apo A-I-containing lipoproteins and apo E fraction III.
Figure 1. Lipid and apolipoprotein profiles of a normolipoproteinemic subject. A. The triglyceride and cholesterol profiles identify the VLDL, LDL, and HDL. B. The apo B and apo A-I peaks confirm the positions of LDL and HDL. Apo E is transported on VLDL-size lipoproteins and is also present as a component of two lipoprotein classes distinct from LDL and slightly larger HDL. Apo C-III is also a component of VLDL and of two lipoproteins distinct in size from LDL, HDL, and the apo E-containing lipoproteins.

Abetalipoproteinemia

Both subjects with abetalipoproteinemia had characteristically low total plasma cholesterol and undetectable plasma triglyceride levels (Table I). Separation of plasma lipoproteins by 4% agarose chromatography demonstrated an unusual pattern compared with normal subjects. There was, as expected, a complete absence of lipoproteins that were the size of chylomicrons, VLDL, and LDL. All of the plasma cholesterol chromatographed as a single peak in lipoproteins the size of large HDL based upon the normal elution volume of HDL cholesterol (Figure 2 A). As expected, the total plasma apo B levels approached the limit of detection of the radioimmunoassay. The values measured were so low that they may have reflected an artifact associated with the low serum dilutions assayed. No apo B could be detected in column chromatographic fractions. Apo A-I, on the other hand, was low and chromatographed just after the single cholesterol peak (Figure 2 B). Plasma apo E levels were normal in both subjects.

The apo E profile across the lipoprotein spectrum of Subject 1 (Figure 2 B) demonstrated a single fraction eluting with lipoproteins larger than those containing the major proportion of the plasma apo A-I and cholesterol. On the basis of elution volume, these apo E-containing lipoproteins were slightly
Larger than the fraction III defined in normolipoproteinemic subjects.

The profile of Subject 2 was identical. Plasma apo C-III was very low in both subjects. Virtually all of the plasma apo C-III was associated with lipoproteins that were intermediate in size between the apo E- and the apo A-I-containing HDL fractions.

**Tangier Disease**

Subject 3 was a normotriglyceridemic subject with Tangier disease diagnosed on the basis of extremely low HDL levels (2mg/dl) and the clinical signs associated with Tangier disease. The total plasma cholesterol level was 161 mg/dl (Table 1). In contrast to both normolipoproteinemic and abetalipoproteinemic subjects, agarose column separation showed all the plasma cholesterol and triglyceride to be localized in two fractions: a minor one in the void volume representing chylomicrons; and the other in LDL-size lipoproteins (Figure 3 A). Plasma apo B levels were low normal (Table 1). The major proportion of apo B coincided with the major cholesterol peak, with a minor proportion in a broad band representing lipoproteins the size of chylomicrons and VLDL (Figure 3 B). Plasma apo A-I levels were very low, less than 1% of normal (Table 1). Apo A-I was undetectable in most column fractions, but a small peak in a fraction the size of small HDL was seen by elution volume (data not shown). The plasma levels of apo E were in the high normal range, and RIA analysis of the column fractions detected two peaks: the first in the chylomicron region and the second in a fraction the size of apo E fraction III. This was not coincident with the small peak of detectable apo A-I and represented larger particles of HDL. The total plasma apo C-III was in the normal range and was distributed among three lipoprotein fractions. One was in the chylomicron region, one was slightly larger than LDL, and one was slightly smaller than the apo E-containing fraction III. Therefore, despite the near absence of apo A-I, both apo E and apo C-III maintained an apparently normal association with lipoprotein particles the size of large HDL. In this subject, apo C-III was also found in a fraction which appeared, on the basis of the elution volume, to be nonlipoprotein-bound and which coeluted with plasma glycerol.

The second Tangier patient, Subject 4, differed strikingly from Subject 3. This patient had been diagnosed on the basis of the clinical signs of the disease. In contrast to Subject 3, Subject 4 was hypertriglyceridemic. The total plasma apo B was normal and was found solely in lipoproteins that coeluted with the cholesterol peak, LDL (Figure 4). As with Subject 3, the apo A-I levels were very low (Table 1). The apo A-I detected in column fractions, however, was entirely within a broad peak representing lipoproteins extending from large to small HDL (data not shown). Apo E levels were in the low normal range and were distributed in a complex pattern across the lipoprotein spectrum. As with Subject 3, apo E was found in chylomicrons and in a broad peak of lipoproteins that were intermediate in size between the apo E- and the apo A-I-containing HDL fractions.

**Combined Apo A-I/C-III Deficiency**

Subjects 5 and 6 are sisters who have been characterized previously with respect to their clinical syndrome, plasma and lipoprotein lipid levels, and total plasma apolipoprotein concentrations. Like the
Figure 3. Lipid and apolipoprotein profiles of a subject with Tangier disease (Subject 3). A. Cholesterol and triglyceride profiles indicate the presence of chylomicrons and LDL with very low levels of HDL. B. The apo B profile coincides with the major cholesterol and triglyceride peak, confirming the presence of LDL. Apo A-I is barely detectable. A small proportion of apo E is present as a component of lipoproteins of chylomicron size, but the greatest proportion is transported on large HDL-size lipoproteins. Apo C-III is present on chylomicrons, LDL, large HDL, and in a lipoprotein-free form.

Figure 4. Lipid and apolipoprotein profiles of a subject with Tangier disease (Subject 4). A. This subject is slightly hypertriglyceridemic, and all lipoproteins are triglyceride-enriched. The cholesterol profile describes a major LDL peak with a minor fraction corresponding in size to HDL. B. Apo B is confined to a peak coincident with the major cholesterol peak, LDL. Apo E is distributed across a broad spectrum of lipoproteins the size of VLDL and intermediate-size lipoproteins and across a wide range of particles of HDL size. Apo C-III has a distribution similar to that of apo E, but is absent from HDL-size lipoproteins.
Tangier subjects, these patients had undetectable plasma HDL cholesterol levels and correspondingly low total cholesterol (Table 1). In contrast to Tangier patients, however, their triglyceride levels were also low. All of the plasma cholesterol and triglyceride chromatographed as lipoproteins having the size of LDL as shown for Subject 6 in Figure 5 A.

Total plasma apo B levels were normal, and RIA analysis of column fractions localized all of the apo B to lipoproteins eluting with the sole cholesterol peak, LDL (Figure 5 B). Apo B in VLDL was below the limit of detection of the apo B RIA (less than 0.02 μg), consistent with the very low levels of triglyceride in this lipoprotein fraction. Plasma apo E levels were also normal in both subjects (Table 1). Apo E chromatographed as a small peak in the region of apo E fraction II, but the major portion eluted in association with lipoproteins that were the size of the large HDL fraction, fraction III, even though an HDL cholesterol peak was absent. Apo A-I levels were at the lower limits of the assay and apo C-III could not be detected in whole plasma. Plasma apo C-II levels were low, but easily measurable (6.6 μg/ml in Subject 5 and 6.5 μg/ml in Subject 6. The chromatographic profile of apo C-II showed three peaks: the first was larger than LDL; the second major peak coeluted with apo B in LDL; and the third coeluted with the apo E fraction III.

Apo A-II was also assayed on column chromatographic fractions of fasting plasma from these subjects, but these latter samples were obtained and chromatographed several months after those described above and shown in Figure 5. Therefore, they have not been included on this graph. In these samples, however, total plasma apolipoproteins were similar to those in Table 1 and the apo E chromatographic profile was identical to that shown in Figure 5. In both Subjects 5 and 6, apo A-II demonstrated a single peak of immunoreactivity that was displaced from apo E by an additional 20 ml to 25 ml elution volume, suggesting that most apo E and apo A-II were on different lipoprotein particles. This volume differential is approximately the same as that between the peak of the profile of apo E and that of apo A-I in normal plasma (Figure 1).

Immunoaffinity chromatography was also performed on the apo E peak from Subjects 5 and 6 to further characterize the physical association of apo E.

![Figure 5](http://atvb.ahajournals.org/)

**Figure 5.** Lipid and apolipoprotein profiles of a subject deficient in apolipoproteins C-III and A-I (Subject 6). A. All cholesterol and triglyceride are contained in a single lipoprotein peak the size of LDL. B. Apo B coelutes with the sole cholesterol peak. Apo E demonstrates a major peak in the region of HDL-size lipoproteins with minor peaks that are intermediate and LDL size. Apo C-II shows a major fraction coincident with LDL, but minor fractions are also present in VLDL and in the HDL region.
with apolipoprotein A-II in this population of lipoprotein particles. For this study, the fractions representing the apo E-containing lipoproteins of large HDL size were pooled and applied to an anti-apo E IgG affinity column as described under Methods. The apolipoprotein A-II content of the input, bound, and unbound fractions was then determined to estimate the proportion of apo A-II that was physically associated with apo E. In both subjects, all of the apo E bound, but only a small proportion of the apo A-II that applied to the anti-apo E immunoadsorbent was recovered in the bound fraction. For Subject 5, 6.4% of the applied apo A-II was bound; for Subject 6, 5.8% was bound. The presence of the E/A-II complex was not specifically examined. The total recoveries of apo A-II from these two columns were determined to be 63% and 86%, respectively. Apo C-II levels were also measured. Of the apo C-II applied, 56% in Subject 5 and 45% in Subject 6 bound to the anti-apo E matrix. In terms of the composition of this apo E-containing particle, the immunoaffinity data thus indicated that of the apolipoproteins directly measured (i.e., apo E, apo A-II, and apo C-II), apo E was not the sole apolipoprotein, but was more than 65% of the apolipoprotein mass.

Discussion

The present study has documented the existence of an apo E-containing lipoprotein that is the size of large HDL and that appears to be independent of the presence of the quantitatively major apolipoproteins, apo B and apo A-I. In normal and hyperlipidemic states, it is difficult to determine the relationship of the dynamic, but quantitatively minor, apolipoproteins to the major apolipoproteins which dominate in terms of mass. Apo E-enriched subpopulations have been reported in experimental studies and in usual metabolic states. Thus, cholesterol feeding causes the accumulation of cholesterol ester in a lipoprotein class with apo E as its sole protein moiety. In humans, LCAT-deficiency and alcoholic hepatitis have both been characterized as possessing a lipoprotein of HDL size and density which is unusually rich in apo E. In normal individuals, heparin-affinity chromatography has identified HDL subfractions containing apo E similar to as well as apo A-I, apo A-II, and C peptides. Analytical ultracentrifugation performed after the removal of apo B-containing lipoproteins has provided further evidence for the existence of discrete apo E containing large HDL which may be related to LCAT activity. The present study supports these studies and provides evidence that this lipoprotein population is independent from apo A-I-containing HDL.

The studies of two subjects with abetalipoproteinemia demonstrated the complete absence of apo E-containing lipoproteins that have VLDL and intermediate size, suggesting that in vivo the presence of apo E-containing subclasses in these density inter-

vals requires the structural integrity of apo B-containing lipoproteins. In these subjects, apo E levels in plasma were normal, but all of the apo E was confined to a single lipoprotein class that was intermediate in size between LDL and HDL. Both HDLc and HDLr of abetalipoproteinemia have been well characterized previously as enlarged, less dense, and more apo E- and cholesterol ester-rich than normal HDL. This is probably the result of an absence of triglyceride-rich lipoproteins that are needed for normal lipolytic transfer and exchange reactions with HDL. Previous studies have documented a metabolic interrelationship between triglyceride-rich lipoprotein hydrolysis and apo E-containing lipoproteins in HDL. Thus, these data from abetalipoproteinemic subjects who lack the substrate for triglyceride hydrolysis and the recipient triglyceride-rich lipoproteins for exchange reactions support the theory of a metabolic origin of apo E-containing HDL that is independent of the apo B metabolic cascade in triglyceride-rich lipoproteins.

The data on the four HDL-deficient subjects indicated that apo E-containing HDL can also exist independently of apo A-I-containing lipoproteins. The two subjects with apo A-I and apo C-III deficiency provided the most striking evidence for this. Both subjects had nearly undetectable HDL cholesterol levels, as reported previously. These subjects, however, clearly possess lipoprotein particles of HDL size as shown in the present report and by a recent study by Forte et al. Our data have shown that nearly all (77.1% and 86.6%) of their total plasma apo E was present in a large HDL fraction that was of the elution volume shown to be characteristic of apo E-containing HDL in normal subjects. Forte et al. have shown in studies using electron microscopy and gradient gel electrophoresis of HDL separated between 1.063-1.21 g/liter that these lipoprotein particles are spherical. They also observed that apo E contributed a significant proportion of the protein in this HDL fraction, although apo A-II clearly dominated in their study and was complexed with apo E, particularly in the higher density range.

Our data confirm the presence of apo A-II eluting in the HDL region of the column at approximately a 3:1 (wt/wt) ratio with apo E (data not shown). The chromatographic profiles of apo E and apo A-II, however, were not coincident; the peak of apo E eluted earlier than that of apo A-II. Our data do not address the presence of the E/A-II complex, but the difference in their chromatographic profiles and our observation that most apo A-II was not bound to an anti-apo E IgG affinity column when the intact apo E-containing fraction was applied suggest that the two apolipoproteins may be mostly on separate lipoprotein particles, at least in the region of the chromatographic profile where apo E was dominant. The fact that Forte et al. concentrated on a density interval that excluded part of the apo E-containing lipoprotein that we analyzed may explain some of the differences in the two reports.
Thus, the data from both these reports suggest the existence of apo E-containing HDL that are not dependent upon apo A-I-containing HDL for their integrity. In normal subjects, it is clear that apo A-I also contributes significantly to the protein mass in apo E-containing HDL that is isolated by immunoaffinity chromatography, but the results from these two subjects suggest a flexibility in apolipoprotein composition. This potential for flexibility in surface apolipoprotein composition has been shown in vitro. Furthermore, even within these individuals, the composition may not be fixed. One of the apo A-I/C-III-deficient subjects was restudied in a protocol involving sampling before and 4 hours after a high-fat meal. When restudied while fasting, this subject had 42% of her apo C-II in the peak that paralleled that of apo E in HDL (more than previously, Figure 5 B). After feeding, however, virtually all of the apo C-II shifted to chylomicrons and VLDL-size lipoproteins, but only a small proportion (5.2%) of the apo E moved to these lipoproteins.

The results from the Tangier patients showed more heterogeneity (as reported by others) and were consistent with the conclusion that the apo E-enriched HDL fraction can exist in the presence of very low levels of apo A-I. Previous studies on Tangier subjects have described HDL that contained apo A-II as the sole apolipoprotein and have failed to detect any apo C peptides in the HDL density range. These studies did not specifically measure apo E, however, and did use ultracentrifugation as a preparative technique, which could alter the apo E distribution.

One consistent difference between the profiles of subjects with Tangier disease and those with apo A-I/C-III deficiency was the presence of apo E in triglyceride-rich lipoproteins in the former but not latter syndrome. Both subjects with apo A-I/C-III deficiency had very low triglyceride levels, whereas Tangier patients frequently have hypertriglyceridemia. It is not known whether this reflects differences in the source of the apo A-I defect in the two disorders (catabolism vs synthesis), or whether the presence of apo C-III is a prerequisite for the maintenance of normal plasma levels of triglyceride-rich lipoproteins, the latter providing a reservoir for cholesterol ester and apo E transfer and exchange from HDL. Turnover studies of apo B in VLDL in the apo A-I/C-III deficiency patients indicate a very rapid catabolism of VLDL, an observation that is compatible with the latter hypothesis.

The persistent presence in these abnormal subjects of apo E-enriched lipoprotein particles that are the size of large HDL raises the issue of the existence and nature of this particle in normal individuals. From the size of these lipoproteins, it is possible that they represent discoidal particles as have been described for LCAT-deficient and alcoholic subjects. Other studies, however, have indicated that the HDL of the apo A-I/C-III-deficient subjects and of Tangier subjects are spherical and not discoidal. Similarly, we have shown in normolipidemic subjects that the HDL subfraction that contains apo E is spherical after isolation by immunoaffinity chromatography. It is therefore likely that this lipoprotein is a discrete entity existing in low concentrations in both normal and abnormal states. The fact that both Tangier and apo A-I/C-III-deficient subjects have nearly undetectable levels of cholesterol in their HDL implies that this lipoprotein particle only contains a small proportion of the plasma steady-state cholesterol mass. Our data using immunoaffinity chromatography to isolate the particle from normal, HDL-size particles have shown that only 2% to 4% of the cholesterol in lipoproteins of this size is retained on an anti-apo E immunoadsorbent; this supports this conclusion but this does not necessarily relate to metabolic significance. The apo E-containing subpopulation may be a class of dynamic lipoprotein particles with constantly changing apolipoprotein and lipid composition that are products of cholesterol ester and triglyceride exchange and of transfer and hydrolytic reactions.

Acknowledgments

The authors thank N. Ngai, L. Goldman, T. Han, and R. Veiss for their invaluable assistance in the laboratory. We also give special thanks to M. Fernandez for preparation of this manuscript.

References

1. Cheung MC, Albers JJ. Distribution of high density lipoprotein particles with different apoprotein composition: Particles with Al and All and particles with Al but no All. J Lipid Res 1982;23:747–753
17. Illingworth DR, Alam NA, Sundberg EE, Hagemann FC, Layman DL. Regulation of low density lipoprotein receptors by plasma lipoproteins from patients with abetalipoproteinaemia. Proc Natl Acad Sci 1983;80:3475–3479

Index Terms: apolipoprotein E • abetalipoproteinaemia • Tangier disease • apolipoprotein A-I/C-III deficiency • lipoproteins
Apo E-containing lipoproteins in low or high density lipoprotein deficiency.
J C Gibson, A Rubinstein, W V Brown, H N Ginsberg, H Greten, R Norum and H Kayden

doi: 10.1161/01.ATV.5.4.371

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://atvb.ahajournals.org/content/5/4/371

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Arteriosclerosis, Thrombosis, and Vascular Biology can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Arteriosclerosis, Thrombosis, and Vascular Biology is online at:
http://atvb.ahajournals.org//subscriptions/