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Abstract

In an attempt to evaluate high density lipoprotein (HDL) subfraction levels in liver diseases, HDL was separated by a precipitation method with dextran sulfate-Mg2+ from sera of 289 healthy adults and 50 patients with liver diseases. The HDL was subdivided into HDL2e and HDL3e by Utermann’s polyacrylamide gel electrophoresis with lauric acid. Ultracentrifugally separated HDL2 and HDL3 roughly corresponded to HDL2e and HDL3e, respectively. Male and female groups had different distributions of HDL2e/HDL3e ratios. Among healthy males, 121 cases had ratios less than 1.0 (mean +/- SD = 0.72 +/- 0.39, n = 150), while among healthy females, the ratios were generally larger than those of males and varied widely from 0.2 to 6.6 (mean +/- SD = 1.77 +/- 1.05, n = 139). Low levels of HDL-cholesterol were found in patients with liver diseases, except those with mild alcoholic liver injury and intrahepatic cholestasis. Apparent decreases in HDL3e, but not in HDL2e, were found in all cases with liver diseases investigated, even in those who did not show decreases in the total HDL level, when male and female patients were analyzed separately. The analysis of HDL subfractions by the present method is simple and useful for the study on altered lipid metabolism in liver diseases.

KEYWORDS: HDL2, HDL3, HDL-cholesterol, electrophoresis, liver disease

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Distribution of Electrophoretically Separated Serum High Density Lipoprotein Subfraction Levels among Healthy Students and Its Alteration in Patients with Liver Diseases

Satoru Ikeda

First Department of Internal Medicine, Okayama University Medical School, Okayama 700, Japan

In an attempt to evaluate high density lipoprotein (HDL) subfraction levels in liver diseases, HDL was separated by a precipitation method with dextran sulfate-Mg\(^{2+}\) from sera of 289 healthy adults and 50 patients with liver diseases. The HDL was subdivided into HDL\(_2^*\) and HDL\(_3^*\) by Utermann's polyacrylamide gel electrophoresis with lauric acid. Ultracentrifugally separated HDL\(_2^*\) and HDL\(_3^*\) roughly corresponded to HDL\(_2^\alpha\) and HDL\(_3^\alpha\), respectively. Male and female groups had different distributions of HDL\(_2^\alpha\)/HDL\(_3^\alpha\) ratios. Among healthy males, 121 cases had ratios less than 1.0 (mean ± SD = 0.72 ± 0.39, n = 150), while among healthy females, the ratios were generally larger than those of males and varied widely from 0.2 to 6.6 (mean ± SD = 1.77 ± 1.05, n = 139). Low levels of HDL-cholesterol were found in patients with liver diseases, except those with mild alcoholic liver injury and intrahepatic cholestasis. Apparent decreases in HDL\(_2^\alpha\), but not in HDL\(_3^\alpha\), were found in all cases with liver diseases investigated, even in those who did not show decreases in the total HDL level, when male and female patients were analyzed separately. The analysis of HDL subfractions by the present method is simple and useful for the study on altered lipid metabolism in liver diseases.

Key words: HDL\(_2^*\), HDL\(_3^*\), HDL-cholesterol, electrophoresis, liver disease

The importance of high density lipoprotein (HDL) as a negative risk factor of coronary heart disease has been shown by a number of reports since Miller and Miller (1) re-evaluated HDL-cholesterol (HDL-Chol) levels. HDL has been further subdivided by ultracentrifugation into HDL\(_2^*\) and HDL\(_3^*\) (2). HDL\(_2^*\) is more important than HDL\(_3^*\) in the prevention of atherogenesis (3, 4). The serum level of HDL, as measured by HDL-Chol (5), decreases in most liver diseases (6–9). It is, therefore, of some interest to see which HDL subfraction is responsible for the decrease in the total HDL level and of what liver diseases the HDL decrease is characteristic. Before solving these problems, the genetic background of the distribution of HDL subfractions should be elucidated since Utermann reported three major patterns of HDL subfractions in normal subjects (10).

In order to carry out a genetic study on samples of a large population and to apply it to clinical materials, less time-consuming and less costly techniques for separation of HDL subfractions are required. The electrophoretic system of Utermann (10) for subfractionation of HDL appears to be suitable for this purpose. The author re-evaluated Utermann's method in the present study,
and analyzed by his method the distribution of electrophoretically separated HDL$_2$ and HDL$_3$ among 289 healthy controls. On the basis of this analysis, the alterations in the HDL$_2$ and HDL$_3$ levels in patients with liver diseases were evaluated. The results of this work were reported previously in abstract form (11).

Materials and Methods

Blood samples were obtained from 289 healthy students of Kagawa University (150 males and 139 females, their ages ranging from 18 to 22) 1 to 3 h after lunch and from 12-14 h fasted patients with liver diseases. There were 3 cases of acute hepatitis (AH), 13 of chronic hepatitis (CH), 10 of liver cirrhosis (LC), 9 of alcoholic liver injury (ALC), 5 of intrahepatic cholestasis (IHC), 4 of biliary obstruction (BO) and 6 of hepatocellular carcinoma (HCC). Sera were separated by centrifugation and kept at 4°C. All the analyses were completed within 3 weeks with reproducible results. Plasma samples were obtained with EDTA (disodium salt, Sigma Chemical Co., St. Louis, 1 mg/ml blood) from healthy adults to separate lipoprotein classes by preparative ultracentrifugation, which was carried out at 6°C in a Hitachi ultracentrifuge, Model 65 P. Lipoprotein subfractions, HDL$_2$ and HDL$_3$, were obtained by three successive runs of ultracentrifugation at d = 1.063, d = 1.125 and d = 1.21 (12, 13).

HDL$_2$ fractions were also separated by differential precipitation with dextran sulfate (molecular weight, 500,000; Pharmacia Fine Chemicals AB, Uppsala) and Mg$^{2+}$ as described by Kostner (14). Concentrations of HDL-Chol and total cholesterol (Chol) were determined by an enzymatic method with T-Choles (Enzymatic, International Reagent Corp., Kobe). The concentrations of Chol in lipoprotein fractions obtained by ultracentrifugation were determined by the method of Zak (15), those of triglycerides (TG) by the method of Van Handel and Zilversmit (16) and those of phospholipids by the method of Bartlett (17).

Polyacrylamide gel disc electrophoresis (PAGE) of lipoproteins was performed by the method of Utermann (10). The separating gel with a diameter of 8 mm contained 4.1 x 10$^{-3}$ M lauric acid and was photopolymerized. The concentration of the gel was varied from 4.5 to 10.0% with respect to acrylamide monomer. Lipoproteins in sera and lipoprotein fractions were prestained with Sudan Black B (18) and 40 $\mu$l of each sample was applied to a gel column. The ratio of HDL$_2$ to HDL$_3$ was estimated by densitometry of stained bands at a wave length of 570 nm and with a slit length and width of 5 mm and 0.5 mm, respectively.

Statistical significance was determined by Student's t test or $\chi^2$-test.

Results

When an HDL fraction separated by differential precipitation with dextran sulfate-Mg$^{2+}$ from the serum of a healthy adult was subjected to electrophoresis at various acrylamide concentrations without adding lauric acid, 3 distinct bands, the slowest one being the darkest, were observed at higher acrylamide concentrations, and 2 major bands, the faster one being darker, were observed at lower acrylamide concentrations (Fig. 1). HDL subbands with larger particle sizes appeared to migrate faster than those with smaller ones as the acrylamide concentration was lowered. In order to see the relationship between the electrophoretic mobility and the particle size, HDL and its subfractions separated by ultracentrifugation were subjected to PAGE at a 7.5% acrylamide concentration without adding lauric acid (Fig. 2). The original HDL was separated into 5 or 6 bands in this case. HDL$_2$ gave bands which corresponded to a group of slow migrating bands of HDL, and HDL$_3$ gave a broad band corresponding to a group of HDL bands with faster mobilities with considerable overlapping between HDL$_2$ and HDL$_3$ bands.

On the other hand, when PAGE was run with lauric acid at different acrylamide concentrations of 6 to 10%, the HDL fraction from the same control subject as in Fig. 2 gave two major lipoprotein bands at every
Fig. 1 PAGE without lauric acid of an HDL fraction separated from the serum of a healthy adult by the precipitation method. The amount of sample applied was 40 µl per column. Percent of acrylamide is shown on the left side of the figure.

Fig. 2 PAGE of ultracentrifugally separated HDL, HDL_{2}, and HDL_{3} fractions from a healthy adult at a 7.5% acrylamide concentration without lauric acid.
Fig. 3 PAGE with lauric acid of HDL fraction at different acrylamide concentrations. The sample used was the same one analyzed for Fig. 2.

Fig. 4 PAGE with lauric acid of ultracentrifugally separated HDL, HDL₂, and HDL₃ fractions. The samples used were the same ones analyzed for Fig. 2.
gel concentration (Fig. 3). The ratio of slow to fast migrating bands of HDL, as determined by densitometry, was relatively constant over the range of gel concentrations. When HDL and its subfractions used for Fig. 2 were subjected to PAGE with lauric acid at a 7.5% acrylamide concentration, 2 bands of HDL, each roughly corresponding to HDL₂ and HDL₃, were obtained (Fig. 4). Since the 2 electrophoretically separated HDL bands did not correspond exactly to HDL₂ and HDL₃ separated by ultracentrifugation, the 2 bands separated by PAGE with lauric acid were designated as HDL₂ʰ and HDL₃ʰ (19), corresponding to HDL₂ and HDL₃ respectively, with “ʰ” standing for electrophoresis. Although HDL₃ was considered to be heterogeneous from its broad appearance in PAGE with lauric acid, it did not reveal split peaks upon analytical ultracentrifugation (Fig. 5), indicating that the preparation was ultracentrifugally homogeneous. Therefore, the heterogeneity of HDL₃ in electrophoresis was not due to contamination of other HDL subfractions but was probably due to the charge difference.

The HDL₂ʰ/HDL₃ʰ ratios obtained by densitometry of normal controls were approximately equal to the HDL₂/HDL₃ ratios calculated from the cholesterol concentration of each HDL fraction obtained by ultracentrifugation (Table 1).

HDL fractions separated by differential precipitation with dextran sulfate-Mg²⁺ from the sera of the 289 healthy adults were analyzed by PAGE (7.5% acrylamide) with lauric acid. Although more than 2 subbands were obtained in some cases, they were grossly separable into 2 groups. The validity of this classification was tested by comparing the relative mobilities (Rm) of the major separated bands (2 or more). The
### Table 1: Comparisons of HDL\(_2^c\)/HDL\(_2^d\) ratios with HDL\(_2^d\)/HDL\(_2^c\) ratios\(^a\)

<table>
<thead>
<tr>
<th>Case</th>
<th>Components</th>
<th>Subfraction</th>
<th>Ratio</th>
<th>Subfraction</th>
<th>Ratio</th>
<th>Subfraction</th>
<th>Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>HDL(_2^d)</td>
<td>HDL(_2^c)</td>
<td>HDL(_2^d)/HDL(_2^c)</td>
<td>HDL(_2^c)</td>
<td>HDL(_2^d)</td>
<td>HDL(_2^d)/HDL(_2^c)</td>
</tr>
<tr>
<td>S.I.</td>
<td>Chol</td>
<td>24.0</td>
<td>42.2</td>
<td>0.57</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TG</td>
<td>18.3</td>
<td>27.3</td>
<td>0.67</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>PL</td>
<td>27.7</td>
<td>62.5</td>
<td>0.44</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>D.M.(^b)</td>
<td></td>
<td></td>
<td></td>
<td>6.7</td>
<td>11.9</td>
<td>0.56</td>
</tr>
<tr>
<td>C.K.</td>
<td>Chol</td>
<td>26.3</td>
<td>31.4</td>
<td>0.84</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TG</td>
<td>12.0</td>
<td>16.6</td>
<td>0.72</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>PL</td>
<td>29.6</td>
<td>71.1</td>
<td>0.42</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>D.M.(^b)</td>
<td></td>
<td></td>
<td></td>
<td>10.1</td>
<td>12.6</td>
<td>0.80</td>
</tr>
<tr>
<td>I.H.</td>
<td>Chol</td>
<td>14.2</td>
<td>34.1</td>
<td>0.42</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>TG</td>
<td>16.7</td>
<td>22.6</td>
<td>0.74</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>PL</td>
<td>16.1</td>
<td>34.3</td>
<td>0.47</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>D.M.(^b)</td>
<td></td>
<td></td>
<td></td>
<td>4.0</td>
<td>9.1</td>
<td>0.44</td>
</tr>
</tbody>
</table>

\(^a\): The determinations were made on ultracentrifugally separated fractions, and the units are in mg/dl.

\(^b\): D.M. is an arbitrary densitometric unit of electrophoretically separated HDL\(_2^c\) and HDL\(_2^d\).

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**Fig. 6**: The distribution of Rm values of major bands separated by PAGE with lauric acid in males and females.
Ikeda: Distribution of electrophoretically separated serum high density lipoproteins

Fig. 7 The distribution of HDL_2^o/HDL_3^o ratios among males and females.

![Graph showing the distribution of HDL_2^o/HDL_3^o ratios among males and females.]

Fig. 8 The distribution of the logarithm of HDL_2^o/HDL_3^o ratios among males and females.

![Graph showing the distribution of the logarithm of HDL_2^o/HDL_3^o ratios among males and females.]

Rm values were distributed in 2 peaks for both males and females (Fig. 6). Since an Rm value of 0.43 best discriminated between the 2 peaks in the analysis of all the cases, HDL bands with Rm values equal to or greater than 0.43 were designated as HDL_2^o, and those with Rm values smaller than 0.43 as HDL_3^o. Distributions of the HDL_2^o/HDL_3^o ratios obtained in male and female groups are shown in Fig. 7. Male and female groups had different distributions of the ratio. Among males, 80.7% of the cases had HDL_2^o/HDL_3^o ratios less than 1.0, with a mean value of 0.72 and standard deviation of 0.39, while among females, the ratio varied widely from 0.2 to 6.6, with a mean value and a standard deviation of 1.77 and 1.05, respectively. Distribution of the logarithm of the HDL_2^o/HDL_3^o ratio approximated the normal distribution in males and females, each with a different peak corresponding to the original one, -0.2 to -0.4 for males and 0.2 to 0.4 for females (Fig. 8). However, the logarithmic distribution of the HDL_2^o/HDL_3^o ratio of all the subjects was not split into 2 peaks on a statistical basis.

Subjects were divided by the HDL_2^o/HDL_3^o ratio into 3 groups according to Utermann (10); those with ratios less than 1.0, those with ratios from 1.0 to 2.0 and those with ratios larger than 2.0. Cases belonging to the group with HDL_2^o/HDL_3^o ratios less than
Table 2  Total Chol and HDL-Chol concentrations, the HDL\(_2^c\)/HDL\(_3^c\) ratio and derived values (HDL\(_2^c\)-Chol, HDL\(_3^c\)-Chol) in healthy male and female adults\(^a\)

<table>
<thead>
<tr>
<th>Sex</th>
<th>No. examined</th>
<th>Total Chol (mg/dl)</th>
<th>HDL-Chol (mg/dl)</th>
<th>HDL(_2^c)-Chol(^b) (mg/dl)</th>
<th>HDL(_3^c)-Chol(^b) (mg/dl)</th>
<th>HDL(_2^c)/HDL(_3^c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>150</td>
<td>155 ± 22.6</td>
<td>46.3 ± 10.0</td>
<td>18.8 ± 8.3</td>
<td>27.5 ± 5.8</td>
<td>0.72 ± 0.39</td>
</tr>
<tr>
<td>Female</td>
<td>139</td>
<td>167 ± 23.5</td>
<td>55.0 ± 10.9</td>
<td>33.5 ± 12.4</td>
<td>21.5 ± 6.0</td>
<td>1.77 ± 1.05</td>
</tr>
<tr>
<td>Total</td>
<td>289</td>
<td>161 ± 23.6</td>
<td>50.5 ± 11.3</td>
<td>25.8 ± 12.8</td>
<td>24.6 ± 6.6</td>
<td>1.22 ± 0.94</td>
</tr>
</tbody>
</table>

\(a\): Values are given as the mean ± standard deviation.

\(b\): Calculated by the following equations:

\[
\text{HDL}_2^c\text{-Chol} = \text{HDL-Chol} \times \frac{\text{HDL}_2^c}{\text{HDL}_1^c + \text{HDL}_3^c}.
\]

\[
\text{HDL}_3^c\text{-Chol} = \text{HDL-Chol} \times \frac{\text{HDL}_3^c}{\text{HDL}_1^c + \text{HDL}_3^c}.
\]

Table 3  Correlation matrices for total Chol, HDL-Chol, HDL\(_2^c\)-Chol, HDL\(_3^c\)-Chol and the HDL\(_2^c\)/HDL\(_3^c\) ratio in healthy male and female adults

<table>
<thead>
<tr>
<th>Sex</th>
<th>No. examined</th>
<th>Lipoproteins and their subfractions</th>
<th>HDL and its subfractions</th>
<th>HDL(_2^c)/HDL(_3^c)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Total Chol</td>
<td>HDL-Chol</td>
<td>HDL(_2^c)-Chol(^*)</td>
</tr>
<tr>
<td>Male</td>
<td>150</td>
<td>Total Chol</td>
<td>0.256</td>
<td>0.065</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL-Chol</td>
<td>0.815(^*)</td>
<td>0.557(^*)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL(_2^c)-Chol</td>
<td>-0.027</td>
<td>0.864(^*)</td>
</tr>
<tr>
<td>Female</td>
<td>139</td>
<td>Total Chol</td>
<td>0.386(^*)</td>
<td>0.290(^*)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL-Chol</td>
<td>0.857(^*)</td>
<td>0.049</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL(_2^c)-Chol</td>
<td>-0.473(^*)</td>
<td>0.897(^*)</td>
</tr>
<tr>
<td>Total</td>
<td>289</td>
<td>Total Chol</td>
<td>0.390(^*)</td>
<td>0.273(^*)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL-Chol</td>
<td>0.877(^*)</td>
<td>0.002</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HDL(_2^c)-Chol</td>
<td>-0.483(^*)</td>
<td>0.877(^*)</td>
</tr>
</tbody>
</table>

\(^*\): \(p < 0.001\).

1.0 composed 80.7% of the males and 23.0% of the females, those with ratios from 1.0 to 2.0 composed 18.7% of the males and 43.9% of the females, and those with ratios larger than 2.0 composed 0.6% of the males and 33.1% of the females. The percent distributions were approximately equal to those of Utermann’s study (10).

Mean concentrations of total Chol, HDL-Chol, the HDL\(_2^c\)/HDL\(_3^c\) ratio and derived values (HDL\(_2^c\)-Chol and HDL\(_3^c\)-Chol) of the 289 adults are given in Table 2. The differences between the male and the female groups were statistically significant (\(p < 0.001\)) in all the values given in Table 2. Correlation matrices for these values are given in Table 3. In males, the total Chol level correlated with HDL\(_2^c\), while HDL correlated with both HDL\(_2^c\) and HDL\(_3^c\). In females, the total Chol level correlated with both HDL and HDL\(_2^c\), while HDL correlated with HDL\(_2^c\), and HDL\(_3^c\) negatively correlated with HDL\(_2^c\). Thus, the high Chol level in females represents a high HDL level due to the increase in HDL\(_2^c\), whereas the high Chol level in males may indicate a high level of HDL\(_2^c\).

In patients with liver diseases, concentrations of total Chol, HDL-Chol and HDL subfractions were similarly analyzed, and
the results are given in Table 4. The total Chol level in IHC patients was significantly higher in males and that in LC patients was significantly lower in females than in the respective normal controls. Significant decreases in HDL-Chol level in liver disease patients, except for IHC and ALC patients, were frequently seen in females. The lower levels of HDL₃-Chol in liver disease patients, except for male patients with ALC and female patients with CH, were significant. The decreases in the HDL₃ level were apparent even in IHC and ALC patients, in whom HDL-Chol levels were rather high. Thus, the measurement of HDL₃ was more apparent than that of total HDL-Chol level to show the altered Chol metabolism in liver disease patients.

### Discussion

Electrophoretic separation of HDL subfractions has been studied by several workers. Narayan et al. (20) fractionated HDL by PAGE into several bands, although none of the bands corresponded to respective HDL subfractions separated by ultracentrifugation. It seems reasonable to separate HDL₂ and HDL₃, which were separated originally by ultracentrifugation (2), by polyacrylamide gradient gel slab electrophoresis (21) since this method separates particles by the difference in size. However, the resolution of HDL₂ and HDL₃ shown by Anderson et al. (21) was not satisfactory, because each HDL subfraction contained subcomponents overlapping each other. It should be realized that the separation mechanism by ultracentrifugation and that by PAGE, even in gradient gels, are fundamentally different. The former is mainly dependent on the difference in density of the particles to be separated and the latter is due to the molecular sieving determined by the size and shape as well as the charge (not for gradient gels) of the particles. A compromise be-
between the two methods was achieved by Utermann's electrophoretic system, in which lauric acid is incorporated into the ordinary PAGE to minimize the charge effect (10). A better resolution of HDL₂ and HDL₃ by Utermann's method (10) than by Davis's (22) PAGE was also demonstrated in the present study. Although the validity of the separation of HDL into two major components was shown in this study by statistical analysis of the distribution of lipoprotein band mobilities, further heterogeneity of electrophoretically separated HDL subfractions, i.e., the presence of more than 2 components of HDL, cannot be neglected. In fact, the subdivision of HDL₂ and HDL₃ has been shown by isoelectric focusing (23, 24). HDL heterogeneity is also shown to be caused by different isolation procedures (25, 26). Anderson et al. (4) divided HDL₂ into HDL₂₈ and HDL₂₀, although no clear-cut correspondence of several minor components of HDL₂ obtained by Utermann's method to HDL₂₈ and HDL₂₀ was seen in our work (unpublished results). Based on these observations, the two major bands separated by Utermann's electrophoretic system were named HDL₂ₑ₈ and HDL₂ₑ₀ in order to make a clear distinction from HDL₂ and HDL₃, which were separated by ultracentrifugation.

Functional similarities of HDL₂ₑ₈ and HDL₂ₑ₀ to HDL₂ and HDL₃, respectively, were shown in our previous study (19), in which Nicomol, the derivative of nicotinic acid, increased mainly HDL₂ₑ₈ as nicotinic acid did HDL₂ (27). The predominance of HDL₂ₑ₈ and HDL₂ₑ₀ in women is another similarity. The mean HDL₂ₑ₈/HDL₂ₑ₀ ratio in females was 1.77 and that in males was 0.72. These values were close to the reported ratio measured by ultracentrifugation (4).

Utermann divided electrophoretic patterns of HDL subfractions into 3 types: predominance of the HDL₂ band, similar intensities of the HDL₂ and HDL₃ bands and predominance of the HDL₃ band (10). However, this classification is not based on statistical analysis of the distribution of HDL₂ and HDL₃ among the cases studied. The analysis of HDL₂ and HDL₃ distributions is important in the evaluation of the factors which determine the levels of HDL₂ and HDL₃. In the present study, the distribution of the HDL₂ₑ₈/HDL₂ₑ₀ ratio was shown to be continuous in contrast with Utermann's classification. It appears, therefore, that the levels of HDL₂ and HDL₃ are not controlled by a single gene. In considering the significant difference in the HDL₂ₑ₈/HDL₂ₑ₀ ratio between males and females, hormonal factors may play an important role in the control of the HDL₂ₑ₈/HDL₂ₑ₀ ratio.

The positive correlation existing between the total Chol and HDL (or HDL₂ₑ₈) in females even in the presence of a highly negative correlation between the HDL₂ₑ₈ and HDL₂ₑ₀ levels is important in the interpretation of high total Chol levels. The possibility of the longevity syndrome (29) must be considered first when young women were shown to have high levels of total Chol. In a diabetic group, a positive correlation between total Chol and HDL₃-Chol has been reported (30), indicating a difference in the mechanism causing increased total Chol levels.

In patients with liver diseases, the concentration of HDL-Chol was lower than that of normal subjects except for patients with IHC and ALC as reported earlier (6). The lower level of HDL-Chol was found in this study to be due to the decrease in the HDL₂ₑ₈ level. The lower HDL₂ₑ₈ level existed even in the presence of an increased HDL level, indicating the importance of a low HDL₂ₑ₈ level as a common underlying phenomenon in hepatic injury. Tanaka et al. (31) and Koga et al. (32) reported similar results obtained by different methods.

HDL₂ is considered to be formed from nascent HDL and further converted to HDL₃,
with a recycling or shuttle system between HDL₂ and HDL₃ (33). Assuming this hypothesis to be correct, a hepatic lipase (33), which catalyzes the conversion of HDL₂ to HDL₃, is an enzyme highly sensitive to liver injury and independent of cholestasis or the ethanol effect (7). Hormonal imbalance in liver diseases may be another important factor, because the ratios of HDL₃ to HDL₂ in male patients with liver disease are as high as those in female patients. Whatever the mechanism of the HDL₃ decrease may be, the present method is simple and clinically useful in the evaluation of abnormal HDL metabolism in liver diseases.

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References


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Reprint requests to:
Satoru Ikeda
Department of Internal Medicine
(Second Unit)
Shimane Medical University
89-1 Enya-cho
Izumo 693, Japan