Small-Dense LDL Cholesterol/Large-Buoyant LDL Cholesterol Ratio as an Excellent Marker for Indicating Lipodystrophy in HIV-Infected Patients

Pornpen Srisawasdi, PhD,1 Tanida Suwalak, MSc,1 Chonlaphat Sukasem, PhD,2 Anchalee Chittamma, PhD,1 Anothai Pocathikorn, PhD,3 Somlak Vanawan, MSc,1 Apichaya Puangpetch, PhD,2 Siwalee Santon, MSc,2 Wasun Chantratita, PhD,4 Sasisopin Kiertiburanakul, MD,5 and Martin H. Kroll, MD6

From the 1Division of Clinical Chemistry, 2Division of Pharmacogenomics and Personalized Medicine, and 4Division of Virology, Department of Pathology, Faculty of Medicine, Ramathibodi Hospital, Mahidol University, Bangkok, Thailand; 3Department of Pathology, Faculty of Medicine, Prince of Songkla University, Songkla, Thailand; 1Division of Infectious Diseases, Department of Medicine, Faculty of Medicine, Ramathibodi Hospital, Mahidol University, Bangkok, Thailand; and 6Quest Diagnostics, Madison, NJ.

Key Words: HIV infection; Antiretroviral therapy; Coronary heart disease; Lipodystrophy; LDL subclass; Small-dense low-density lipoprotein; Large-buoyant low-density lipoprotein

ABSTRACT

Objectives: To examine whether the lipid parameters are predicting factors for human immunodeficiency virus (HIV)–associated lipodystrophy.

Methods: Whole-body fat compositions of HIV-positive patients receiving stavudine-containing antiretroviral regimens (n = 79) were determined. Lipodystrophy was defined as a ratio of trunk fat mass/lower limb fat mass greater than 2.28. Blood samples were analyzed for total cholesterol (TC), triglycerides, low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), small-dense LDL-C (sdLDL-C), apoAI, apoB, lipoprotein(a), and CD4 cell counts. Large-buoyant LDL-C (lbLDL-C) was calculated (LDL-C minus sdLDL-C).

Results: Twenty-six patients were classified as having lipodystrophy. The mean values of triglycerides, HDL-C, sdLDL-C, apoB, TC/HDL-C, apolipoprotein (apo) B/apoAI, and sdLDL-C/lbLDL-C showed significant differences between patients with and without lipodystrophy (P < .02). Using logistic regression analysis, sdLDL-C/lbLDL-C was identified as a significant predictor of lipodystrophy (P < .001). At a ratio of 0.554, the odds ratio was 17.8 with a likelihood ratio of 5.5.

Conclusions: The sdLDL-C/lbLDL-C ratio is an excellent marker for indicating lipodystrophy in HIV-infected patients.

Advancement of human immunodeficiency virus (HIV) treatment with the development and refinement of highly active antiretroviral therapy (HAART), a combination of antiretroviral agents, has led to long-term disease protection and reduction of an otherwise fatal course.1,3 The most common antiretroviral drugs include nucleoside reverse transcriptase inhibitors (NRTIs), nonnucleoside reverse transcriptase inhibitors (NNRTIs), and protease inhibitors (PIs), which attack HIV in different ways to inhibit the replication process. Although the use of HAART in HIV-infected patients dramatically reduces AIDS-related morbidity and mortality, non-AIDS conditions, particularly cardiovascular disease (CVD), seem to increase mortality in these patients.4,6 A
large number of studies have demonstrated that HIV-infected patients under long-term therapy receiving HAART display HIV-associated lipodystrophy.7-12 The prevalence of lipodystrophy in this population has been reported to vary from 2% to 83%.7 The prevalence of the lipid abnormality in HIV-infected adults, as well as children and adolescents, has been reported as high as 50%.8,9

Lipodystrophy is a metabolic disorder characterized by abnormal body fat distribution, with either loss of subcutaneous adipose tissue (lipoatrophy) or accumulation of visceral adipose tissue (lipohypertrophy).13 In addition, metabolic abnormalities such as dyslipidemia and insulin resistance, which are commonly observed in patients with lipodystrophy, contribute to an increased risk of CVD.5 Several clinical studies have demonstrated the link between antiretroviral therapy, the HAART cocktail, and the development of metabolic abnormalities.14-16 NRTIs such as stavudine and zidovudine have induced lipoatrophy and dyslipidemia.16 Use of first-generation PIs, such as indinavir and saquinavir, has altered lipid and glucose parameters, including increased triglycerides (TG), decreased high-density lipoprotein cholesterol (HDL-C), and increased insulin resistance.14,15 In contrast to PIs and NRTIs, lipodystrophy and other metabolic complications have been rarely reported with NNRTIs such as nevirapine or efavirenz.17 The adverse effects of these drugs frequently occur when used in combination with other agents, especially lopinavir/ritonavir.18

Lipid tests, including TG, total cholesterol (TC), HDL-C, and low-density lipoprotein cholesterol (LDL-C), are widely used for the assessment of CVD in HIV-infected patients. Plasma lipoproteins consist of heterogeneous subclasses of particles with varying density, size, electrophoretic mobility, relative lipid-protein proportions, and binding affinity.19 LDL particles fractionate according to size and density into large, buoyant LDL (lbLDL; diameter ≥25.5 nm) and small, dense LDL (sdLDL; diameter <25.5 nm). The sdLDL particles are believed to be more atherogenic compared with lbLDL particles because their particles more readily penetrate the arterial wall and show a higher affinity to the intimal proteoglycans, as well as have a more prolonged plasma half-life, a lower binding affinity for the LDL receptor, and a lower resistance to oxidative stress than lbLDL.20 More evidence has indicated that an increased sdLDL-cholesterol (sdLDL-C) level resulting from changes in abnormal lipoprotein metabolism is closely associated with an increased risk of CVD and cerebrovascular disease.21-23 In addition, atherogenic dyslipidemia, as demonstrated by sdLDL-C, is closely associated with the metabolic syndrome and insulin resistance.24,25 Thus, sdLDL-C may be associated with lipodystrophy, which plays an important role for characterizing and monitoring lipodystrophy syndrome and its progression.

Patients with severe forms of subcutaneous lipoatrophy, particularly in the face, limbs, and buttocks, have a poor quality of life. Early diagnosis of lipodystrophy syndrome is important because it allows alteration of therapy early in the disease. To our knowledge, no previous study has evaluated whether sdLDL-C concentration and its fraction ratio might predict lipodystrophy. We hypothesize that these lipid parameters are associated with abnormal body fat distribution in HIV-infected patients receiving HAART.

Materials and Methods

Characteristics of the Study Population

A cross-sectional observational study of HIV-infected patients was conducted in the infectious diseases clinic at Ramathibodi Hospital (Bangkok, Thailand). Inclusion criteria for this study were that the patients had to be 15 years or older, infected with HIV, and treated with stavudine and maintained on a stavudine-containing antiretroviral regimen. The patients were invited to participate for evaluation and physical examination for lipodystrophy. The study was approved by the institutional review board of the Faculty of Medicine, Ramathibodi Hospital, Mahidol University. Informed consent was obtained from all study patients.

For all patients, baseline data included demographics, time since first positive HIV antibody test, a previous AIDS-defining illness, CD4 cell count, HIV RNA, past and current antiretroviral therapy, duration of antiretroviral therapy (ART), and duration of treatment with stavudine-containing regimens.

Body Composition Measures

Height, body weight, and waist and hip circumference were measured according to the World Health Organization recommendations.26 Body mass indices (BMIs) and waist-hip ratios were calculated.

Bioelectrical impedance analysis (BIA) was performed with a multifrequency impedance analyzer (Biospace InBody 720 body composition analyzer, GE Healthcare, CA) as described in the manufacturer’s manual. This analyzer was used to determine the body fat mass and the visceral fat area.

Whole-body dual-energy x-ray absoriometry (DXA) scans (Hologic whole-body DXA systems, Hologic, Bedford, MA) were conducted by a single operator. Scans were performed to quantify regional and total body fat mass, fat-free mass, and bone mass, as described in the Lipodystrophy Case Definition Study.27

Because the clinical diagnosis of lipodystrophy often is subject to self-report by patients and it is often difficult to distinguish lipodystrophy from lipoatrophy or from malnutrition, we used an objective case definition of lipodystrophy for HIV-infected patients developed by Asha et al.28

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Lipodystrophy was defined as a ratio of trunk fat mass to lower limb fat mass greater than 2.28.

Biochemical Analyses

A blood sample was collected from each patient after a 10- to 12-hour overnight fast. All sera samples kept at –80°C were analyzed for TC, TG, LDL-C, and HDL-C on the Siemens Dimension RxL Max by using the Siemens enzymatic methods (Siemens Medical Solution Diagnostics, Tarrytown, NY). Apolipoprotein B (apoB), apolipoprotein AI (apoAI), and lipoprotein(a) were measured on the Siemens BN Prospeck by using the Siemens immunoturbidimetric assay (Siemens Medical Solution Diagnostics).

For sdLDL-C, a novel homogeneous enzymatic assay (Randox Laboratories, Antrim, UK) was used. This assay employs 2 liquid ready-to-use reagents containing a polyoxyethylene benzylphenyl ether selectively decomposing chylomicron, very low-density lipoprotein (VLDL), and HDL; a polyoxyethylene distyrenelphenyl ether selectively binding to sdLDL to protect it from the action of enzymes; and sphingomyelinase, which possesses higher affinity to larger LDL than sdLDL. We developed the user-defined sdLDL-C method on the Siemens Dimension RxL Max and performed it according to the manufacturer’s specifications. The coefficients of variation for sdLDL-C concentrations at 14.7, 32.4, and 65.0 mg/dL, respectively, were 3.34%, 1.67%, and 0.92% (mean, 1.98%) for within-run imprecision and 4.49%, 4.78%, and 3.76% (mean, 4.34%) for between-day imprecision.

We estimated the lbLDL-C by subtracting the sdLDL-C concentration from the LDL-C concentration. Non–HDL-C (TC concentration minus HDL-C concentration) and the ratio of TC/HDL-C, apoB/apoAI, sdLDL-C/lbLDL-C, TG/HDL-C, TG/sdLDL-C, and TG/lbLDL-C were calculated.

Statistical Analyses

Continuous data with a normal distribution are shown as mean (SD) values, and those with a nonnormal distribution are shown as median (interquartile range) values. Categorical data are shown as frequency and percentage. Continuous data were compared between the HIV groups with and without lipodystrophy using an independent Student’s t test or the Mann-Whitney test, depending on the distribution. The χ² test or Fisher exact test was used for categorical variables where appropriate. Correlation between body composition and lipid parameters was performed by Pearson correlation analysis. A backward stepwise logistic regression model was used to determine the associations between lipodystrophy and lipid markers. To determine factors associated with lipodystrophy, only lipid markers with P values less than .1 in the univariable analyses were selected as covariates in the regression models. The Wald χ² test and likelihood ratio test were used to assess the significance of variables in the model. The results are expressed as an odds ratio (OR) with the 95% confidence interval (95% CI). To evaluate the predictive accuracy, we calculated the area under the receiver operating characteristic (ROC) curve. The diagnostic value for lipodystrophy was performed from the plot of sensitivity against 1 – specificity. Sensitivity, specificity, likelihood ratio, and OR with 95% CI values were calculated to determine the best plausible markers. All analyses were performed using SPSS version 14.0 (SPSS, Chicago, IL). Outcomes were considered statistically significant when the P value was less than .05.

Results

Characteristics of the Study Samples

Of the total 79 patients with HIV infection, the median age was 41.0 years (interquartile range, 37-48 years), and the mean (SE) duration of antiretroviral therapy was 81.1 (2.2) months. Lipodystrophy in HIV-infected patients was objectively defined as the ratio between the trunk fat mass to the lower limb fat mass greater than 2.28.28 The clinical characteristics of all patients (34 men and 45 women), classified into 2 groups according to the presence (n = 26) and absence (n = 53) of lipodystrophy, are summarized in Table 1. Median CD4 cell count was significantly higher in patients without lipodystrophy (132 cell/mm³) than those with lipodystrophy (36 cell/mm³). Patients with lipodystrophy were more likely to have a lower percent body fat mass obtained either from BIA and DXA. Notably, age, weight, BMI, waist and hip circumferences, duration of stavudine treatment, CD4 cell count at baseline, total body fat mass (kg), and visceral fat area were not significantly different between patients with and without lipodystrophy (all P > .08). In contrast, waist/hip ratio, duration of ART, the percentage of total body fat mass and total lean mass, the trunk fat mass (kg)/lower limb fat mass (kg) ratio, and the percentage of trunk fat mass/lower limb fat mass ratio differed significantly between groups (all P < .04).

Table 2 shows the lipid parameters of HIV-infected patients stratified according to the presence and absence of lipodystrophy. Using univariable analysis, in the patients with lipodystrophy, mean concentrations of TG (both at ART initiation and baseline of study), sdLDL-C, and apoB were significantly higher, whereas mean concentrations of HDL-C were significantly lower than in those without lipodystrophy (all P < .02). The concentrations of TC (both at ART initiation and baseline of study), LDL-C, non–HDL-C, lbLDL-C, lipoprotein(a), and apoAI were not statistically different between the 2 groups. The lipid ratios of TC/HDL-C, apoB/apoAI, sdLDL-C/lbLDL-C, TG/HDL-C, TG/sdLDL-C, and TG/lbLDL-C were significantly higher in patients with lipodystrophy than in those without...
lipodystrophy (all $P < .005$). We observed that in comparison with the other lipid ratios, the ratio of sdLDL-C and lbLDL-C demonstrated the highest difference between the 2 groups ($F = 32.363$, $P < .001$). Moreover, the ratios of the standard deviation to the average obtained from sdLDL-C/lbLDL-C for the presence and absence of lipodystrophy were 0.28 and 0.31, respectively, which were lower than those for TG/HDL (0.68 and 0.76, respectively), TG/sdLDL-C (0.53 and 0.39, respectively), and TG/lbLDL-C (0.69 and 0.73, respectively). These ratios imply that the sdLDL-C/lbLDL-C ratio has the smallest spread of results.

**Correlation Between Body Composition and Lipid Parameters**

We assessed the correlation between lipid parameters and body composition involving waist and hip circumferences; total and lean body mass; the percentage fat mass of arm, leg, and trunk; and the trunk to limb ratio, each of which was considered to influence lipodystrophy in HIV-infected patients **Table 3**. Among the study subjects, none of the body compositions demonstrated a significant correlation with the concentrations of TC, LDL-C, non–HDL-C, and lipoprotein(a). The percentage fat mass of arm and lower limbs correlated negatively with TG level and sdLDL-C/lbLDL-C, TG/HDL-C, TG/sdLDL-C, and TG/lbLDL-C ratios and positively with HDL-C, while that of the trunk did not significantly correlate with any lipid parameters. Moreover, it was observed that the percent fat mass of the trunk to lower limb showed the best correlation with the sdLDL-C/lbLDL-C ratio (correlation coefficient, $r = 0.642$), followed by TG (r = 0.533), TG/HDL-C (r = 0.532), TG/lbLDL-C (r = 0.500), TC/HDL-C (r = 0.472), sdLDL-C (r = 0.460), HDL-C (r = 0.433), apoB/apoAI (r = 0.421), TG/sdLDL-C (r = 0.394), and apoB (r = 0.309).
Srisawasdi et al / Ratio of LDL Subclasses Predicts Lipodystrophy

In addition, the sdLDL-C/lbLDL-C ratio gave the highest correlation with the percentage of trunk fat mass/lower limb fat mass ratio ($R^2 = 0.412, P < .001$) and was negatively correlated with the percentage of lower limb fat mass ($R^2 = 0.185, P < .001$) but was not significantly associated with the percentage of trunk fat mass ($R^2 = 0.0006, P = .82$), as shown in Figure 1.

Association of Lipid Parameters With Lipodystrophy

A binary logistic regression model was applied to analyze the association between the lipid variables and lipodystrophy. TG, HDL-C, sdLDL-C, apoAI, apoB, and TC/HDL-C, apoB/apoAI, and sdLDL-C/lbLDL-C ratios were considered independent variables (method 1). Table 4

### Table 2
Lipid Markers of HIV-Infected Patients Classified According to Lipodystrophy

<table>
<thead>
<tr>
<th>Lipid Markers</th>
<th>Lipodystrophy Positive (n = 26), Mean (SD)</th>
<th>Lipodystrophy Negative (n = 53), Mean (SD)</th>
<th>F Test</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lipid at ART initiation, mg/dL</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC</td>
<td>168.4 (34.0)</td>
<td>173.5 (36.7)</td>
<td>0.255</td>
<td>.622</td>
</tr>
<tr>
<td>TG</td>
<td>146.8 (70.5)</td>
<td>97.2 (41.2)</td>
<td>10.508</td>
<td>.002</td>
</tr>
<tr>
<td>Lipid at baseline, mg/dL</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC</td>
<td>211.8 (46.4)</td>
<td>212.9 (42.8)</td>
<td>0.010</td>
<td>.919</td>
</tr>
<tr>
<td>TG</td>
<td>266.4 (143.1)</td>
<td>133.9 (77.0)</td>
<td>28.701</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>LDL-C</td>
<td>134.0 (42.0)</td>
<td>131.2 (34.9)</td>
<td>0.103</td>
<td>.749</td>
</tr>
<tr>
<td>HDL-C</td>
<td>49.2 (10.8)</td>
<td>59.4 (14.9)</td>
<td>9.660</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Non–HDL-C</td>
<td>162.6 (45.5)</td>
<td>153.5 (37.1)</td>
<td>0.905</td>
<td>.345</td>
</tr>
<tr>
<td>sdLDL-C</td>
<td>53.0 (17.9)</td>
<td>40.4 (13.3)</td>
<td>12.546</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>lbLDL-C</td>
<td>88.9 (30.9)</td>
<td>97.8 (29.5)</td>
<td>1.534</td>
<td>.219</td>
</tr>
</tbody>
</table>

Apoptosis, apolipoprotein AI; ApoB, apolipoprotein B; ART, antiretroviral therapy; HDL-C, high-density lipoprotein cholesterol; HIV, human immunodeficiency virus; lbLDL-C, large-buoyant low-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; non–HDL-C, non-high-density lipoprotein cholesterol; sdLDL-C, small-dense low-density lipoprotein cholesterol; TC, total cholesterol; TG, triglycerides.

All biochemical measures are given in conventional units; conversions to SI units are as follows: cholesterol (mmol/L), multiply by 0.0259; triglycerides (mmol/L), multiply by 0.0113.

Non–HDL-C values are calculated by subtracting the HDL-C from the total cholesterol concentrations.

Large buoyant LDL values are calculated by subtracting the sdLDL-C from the LDL-C concentrations.
shows the regression coefficient, OR, and 95% CI of each lipid parameter for lipodystrophy after adjusting for sex, age, and BMI. Using backward stepwise logistic regression (method 2), only the sdLDL-C/lbLDL-C ratio (regression coefficient, 5.798; SE, 2.101) was identified to be significantly associated with lipodystrophy \((P = .01)\). We also performed a logistic regression that compared the ratios of sdLDL-C/lbLDL-C, TG/HDL-C, TG/sdLDL-C, and TG/lbLDL-C for detection of lipodystrophy (Table 4, method 3). The ratio of sdLDL-C/lbLDL-C provided the significant predictor of lipodystrophy and was the only ratio that was statistically significant.

To assess the predictive power of the sdLDL-C/lbLDL-C ratio for lipodystrophy, we performed the ROC analysis. Figure 2 shows the ROC curve (Figure 2A) and the coordinates of percent sensitivity and percent specificity curves at various levels of the sdLDL-C/lbLDL-C ratio (Figure 2B). The area under the ROC curve for the predicted probability of lipodystrophy was 0.817 (95% CI, 0.713-0.921; \(P < .001\)), indicating a good discrimination power.

**Ratio of sdLDL-C to lbLDL-C in Indicating Lipodystrophy**

The appropriate cutoff value of the sdLDL-C/lbLDL-C ratio for the clinical diagnosis of lipodystrophy was investigated by considering the estimated risks for lipodystrophy at various cutoff points. To our knowledge, 3 objective case definitions of HIV-associated lipodystrophy use different DXA regional fat thresholds. Therefore, 3 risk estimate models were conducted to examine the optimal cutoff value of the sdLDL-C/lbLDL-C ratio for indicating lipodystrophy. Model 1 used the case definition criterion for the ratio of trunk fat mass to lower limb fat mass greater than 2.28 for all samples (26 with lipodystrophy and 53 without lipodystrophy). Model 2 used the criterion for the ratio of percent fat mass of the trunk to percent fat mass of the lower greater than or equal to 1.50 for all samples (29 with lipodystrophy and 50 without lipodystrophy). Model 3 used the criterion for the ratio of percent fat mass of the trunk to percent fat mass of the lower limb greater than or equal to 1.329 and 1.961 for women and men, respectively (23 with...
were tabulated in Table 5. Model 1, at the optimal cutoff for the sdLDL-C/lbLDL-C ratio, had a sensitivity of 73%, a specificity of 87%, an OR (95% CI) of 17.8 (5.5-57.8), and a likelihood ratio of 5.5 for predicting lipodystrophy in HIV-infected patients. Model 2 had a sensitivity of 69%, a specificity of 88%, an OR (95% CI) of 16.3 (5.1-52.0), and a likelihood ratio of 4.8 for predicting lipodystrophy in HIV-infected patients.

Figure 2 Receiver operating characteristic curve (A) and coordinate of percent sensitivity and percent specificity curves at various levels of the small-dense low-density lipoprotein cholesterol (sdLDL-C)/large-buoyant low-density lipoprotein cholesterol (lbLDL-C) ratio (B). The area under the receiver operating characteristic curve is 0.817.

Table 4 Logistic Regression Analysis for Lipodystrophy by Lipid Parameters

<table>
<thead>
<tr>
<th>Variables</th>
<th>Logistic Regression, ( \beta ) (SE)</th>
<th>Odds Ratio (95% Confidence Interval)</th>
<th>( P ) Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method 1c</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TG</td>
<td>0.001 (0.006)</td>
<td>1.002 (0.990-1.014)</td>
<td>.763</td>
</tr>
<tr>
<td>HDL-C</td>
<td>0.032 (0.074)</td>
<td>1.033 (0.893-1.195)</td>
<td>.662</td>
</tr>
<tr>
<td>sdLDL-C</td>
<td>-0.027 (0.072)</td>
<td>0.974 (0.846-1.121)</td>
<td>.710</td>
</tr>
<tr>
<td>ApoAI</td>
<td>-0.060 (0.051)</td>
<td>0.942 (0.852-1.041)</td>
<td>.238</td>
</tr>
<tr>
<td>ApoB</td>
<td>0.090 (0.071)</td>
<td>1.094 (0.952-1.257)</td>
<td>.203</td>
</tr>
<tr>
<td>TC/HDL-C ratio</td>
<td>0.386 (0.988)</td>
<td>1.471 (0.212-10.194)</td>
<td>.770</td>
</tr>
<tr>
<td>ApoB/apoAI ratio</td>
<td>-9.002 (8.004)</td>
<td>0.000 (0.000-800.9)</td>
<td>.261</td>
</tr>
<tr>
<td>sdLDL-C/lbLDL-C ratio</td>
<td>5.797 (6.350)</td>
<td>329.3 (0.001-8.373 \times 10^7)</td>
<td>.361</td>
</tr>
<tr>
<td>Constant</td>
<td>0.525 (0.279)</td>
<td>—</td>
<td>.933</td>
</tr>
<tr>
<td>Method 2d</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>sdLDL-C/lbLDL-C ratio</td>
<td>5.798 (2.101)</td>
<td>329.76 (5.37-20,248.5)</td>
<td>.006</td>
</tr>
<tr>
<td>Constant</td>
<td>-4.621 (1.094)</td>
<td>—</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Method 3e</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>sdLDL-C/lbLDL-C ratio</td>
<td>11.469 (4.679)</td>
<td>95,733 (9.967-9.195 \times 10^6)</td>
<td>.014</td>
</tr>
<tr>
<td>TG/HDL-C ratio</td>
<td>0.153 (0.212)</td>
<td>1.165 (0.769-1.764)</td>
<td>.470</td>
</tr>
<tr>
<td>TG/sdLDL-C ratio</td>
<td>0.831 (0.456)</td>
<td>2.295 (0.939-5.609)</td>
<td>.069</td>
</tr>
<tr>
<td>TG/lbLDL-C ratio</td>
<td>-1.245 (0.797)</td>
<td>0.288 (0.060-1.372)</td>
<td>.288</td>
</tr>
<tr>
<td>Constant</td>
<td>-7.824 (2.492)</td>
<td>—</td>
<td>.002</td>
</tr>
</tbody>
</table>

ApoAI, apolipoprotein AI; ApoB, apolipoprotein B; HDL-C, high-density lipoprotein cholesterol; lbLDL-C, large-buoyant low-density lipoprotein cholesterol; sdLDL-C, small-dense low-density lipoprotein cholesterol; SE, standard error; TC, total cholesterol; TG, triglycerides.

ApoAI, apolipoprotein AI; ApoB, apolipoprotein B; HDL-C, high-density lipoprotein cholesterol; lbLDL-C, large-buoyant low-density lipoprotein cholesterol; sdLDL-C, small-dense low-density lipoprotein cholesterol; SE, standard error; TC, total cholesterol; TG, triglycerides.

Appliances and statistical analyses were performed using the NCSS software and R software. The area under the receiver operating characteristic curve is 0.817.

Lipodystrophy and 56 without lipodystrophy. It was found that the optimal cutoff value of the sdLDL-C/lbLDL-C ratio obtained from the different experimental models of DXA-defined lipodystrophy was the same value, 0.554 (data not shown). The estimated risks of the sdLDL-C/lbLDL-C ratio of 0.554 for identifying lipodystrophy with different models
a likelihood ratio of 5.7. For model 3, the sdLDL-C/lbLDL-C ratio gave the lowest estimated risk with a sensitivity of 61%, a specificity of 80%, an OR (95% CI) of 5.7 (2.0-16.4), and a likelihood ratio of 2.8.

### Discussion

HAART suppresses HIV RNA to undetectable levels in HIV-positive patients, allowing immune recovery. The recovery can be measured by increases in CD4 cell counts. However, lipodystrophy frequently occurs in patients and is recognized to be an adverse effect of HAART. Lipodystrophy is considered the result of lipid and metabolic disturbances and has an increased risk of myocardial infarction and clinical cardiovascular disease events.

A large number of studies have presented the association between lipodystrophy syndromes and lipid and lipoprotein concentrations in HIV-infected patients. The pattern of dyslipidemia seen in lipodystrophy, consisting of marked hypertriglyceridemia and reduced HDL-C levels, is similar to that in obesity and the metabolic syndrome. The other potential atherogenic lipid parameters, TC, LDL-C, non–HDL-C, lipoprotein(a), apoB/apoAI, and sdLDL particles, have been increasing in this patient population.

Interestingly, our results indicated that the concentrations of TC, LDL-C, non–HDL-C, lbLDL-C, lipoprotein(a), and apoAI were not significantly different between patients with and without lipodystrophy. In addition, these lipid markers were not significantly associated with limb (both arm and leg) fat mass, trunk fat mass, and the ratio of trunk to limb fat mass (Table 3).

Treatment of HIV with HAART could have a direct impact on metabolic dysregulation. From the loss of limb fat observed in several prospective studies, elevated rates of glycerol and free fatty acid turnover have been reported, resulting from defective adipocyte triglyceride storage, a situation that can increase hepatic triglyceride synthesis. These processes can then induce an increasing rate of hepatic TG-enriched large VLDL secretion, causing high generation of sdLDL particles, which plays an important role in the development of atherosclerosis. The Swiss HIV Cohort Study reported that sdLDL-C and apoB levels were associated with an increased risk of coronary events in HIV-infected patients treated with antiretroviral therapy. The results of our study indicated that the mean apoB and sdLDL-C concentrations in patients with lipodystrophy were higher than in those without lipodystrophy. As we know, the level of apoB represents the total burden of atherogenic particles (ie, VLDL), intermediate-density lipoprotein, LDL, and lipoprotein(a) particles because each of these particles contains 1 apoB molecule. Thus, patients with lipodystrophy appear to have an enhanced atherogenic lipoprotein synthesis, which has a markedly increased risk for CVD.

Compared with HIV-infected patients without lipodystrophy, patients with lipodystrophy have the highest concentrations of sdLDL-C but similar concentrations of total cholesterol and LDL-C (Table 2). Patients with lipodystrophy most likely overproduce sdLDL-C and probably have decreased production of lbLDL-C. Consequently, the ratio of sdLDL-C/lbLDL-C can be relatively high in HIV-associated lipodystrophy. In addition, our findings demonstrated that the sdLDL-C/lbLDL-C ratio has the highest correlation with body composition indices, including waist to hip ratio, total mass, total lean mass, and regional fat mass (ie, percentage of arm fat mass, leg fat mass, and trunk fat mass/lower limb fat mass ratio) compared with the other lipid markers. The ratio of sdLDL-C/lbLDL-C can play an important role in predicting an abnormal lipid distribution.

The ability to gain an early diagnosis of lipodystrophy improves the quality of life among people living with HIV. The diagnosis of HIV-associated lipodystrophy is based on physical examination and the fat mass ratio. The fat mass ratio is the most objective tool for diagnosis because it can detect slight losses of peripheral adipose tissue that may be not detected by physical examination. Moreover, subjective diagnoses made by physicians, patients, or both often contain bias, while the definition, which contains only objective indices, is unbiased. Although using objective data obtained from DXA scans seem to be a sensitive technique in the diagnosis of lipodystrophy, they are often not used in HIV routine clinical practice because of high costs and scarce availability. In our study, we demonstrated that the sdLDL-C/lbLDL-C ratio was strongly and significantly associated with...
lipodystrophy (P = .01). The sdLDL-C/lbLDL-C ratio can be an effective index for detecting lipodystrophy in the HIV-infected patient treated with HAART.

The optimal cutoff value was derived with 3 different models, each using varying criteria quantifying lipodystrophy. Using objective data obtained by DXA thresholds of the fat mass ratio, we found the same optimal cutoff value at 0.554, which had a moderate degree of accuracy with a sensitivity ranging from 61% to 73% and a specificity ranging from 80% to 88%. Asha et al suggested that the absolute fat mass was better than a percentage fat mass because regional fat percentages change with aging. Findings from this study indicated that the trunk to lower limb fat mass at a cutoff of greater than 2.28 tends to be better than other criteria for objectively identifying HIV-associated lipodystrophy. Our study showed high discrimination for lipodystrophy with an OR of 17.8 (95% CI, 5.5-57.8) and a likelihood ratio of 5.5.

The ratio of sdLDL-C/lbLDL-C should improve assessment of lipodystrophy prevalence, risk factors, and pathogenesis; provide prevention and treatment approaches; and assist in diagnosis. The ratio of sdLDL-C/lbLDL-C is particularly powerful in recognizing a dysregulation of lipid metabolism and predicting adverse remodeling of lipodystrophy. The high ratio of sdLDL-C/lbLDL-C in response to HAART, as seen with the alteration of lipid metabolism and developing lipodystrophy, may provide additional information about cardiovascular risk.

Limitations of this study include that it was cross-sectional in design and based on a small sample size of an Asian population. The study did not include other ethnicities, and it is possible that other ethnicities might have different body fat distributions. Because there was a disproportionate predominance by males in the disease group, our results may be more applicable to males. Further study may be able to assign correct proportions to females, bearing in mind that our results reflect the natural prevalence of disease in our population. The cross-sectional design cannot definitely elucidate a cause-and-effect relationship between these variables. Therefore, further study of the clinical significance of the ratio of sdLDL-C/lbLDL-C as an easily determinable value for the management of lipodystrophy syndrome and its progression in HIV-infected patients during therapeutic approaches is warranted.

Conclusion

Early detection of lipodystrophy in HIV patients receiving therapy would afford an opportunity to alter therapy, which might decrease cardiovascular risk. The ratio of sdLDL-C and lbLDL-C is an excellent marker for detecting lipodystrophy in an HIV-infected population. Because DXA scans are expensive, the routine use of the sdLDL-C/lbLDL-C ratio at a cutoff value of 0.554 may preselect patients who would most benefit from a DXA scan. Moreover, in regional areas that do not perform DXA scans, the assessment of lipodystrophy by using a ratio of sdLDL-C/lbLDL-C can provide an alternative approach and be useful in screening HIV patients for lipodystrophy.

Address reprint requests to Dr Sukasem: Division of Pharmacogenomics and Personalized Medicine, Dept of Pathology, Faculty of Medicine Ramathibodi Hospital, Rama VI Rd, Rajthevee, Bangkok 10400, Thailand; chonlaphat.suk@mahidol.ac.th.

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