A Hepatic Lipase Gene Promoter Polymorphism Attenuates the Increase in Hepatic Lipase Activity With Increasing Intra-abdominal Fat in Women

Molly C. Carr, John E. Hokanson, Samir S. Deeb, Jonathan Q. Purnell, Ellen S. Mitchell, John D. Brunzell

Abstract—High hepatic lipase (HL) activity is associated with an atherogenic lipoprotein profile of small, dense LDL particles and lower HDL-C-C. Intra-abdominal fat (IAF) is positively associated with HL activity. A hepatic lipase gene (LIPC) promoter variant (G→A^256) is associated with lower HL activity, higher HDL-C, and less dense LDL particles. To determine whether the LIPC promoter polymorphism acts independently of IAF to regulate HL, 57 healthy, premenopausal women were studied. The LIPC promoter A allele was associated with significantly lower HL activity (GA/AA = 104±34 versus GG = 145±57 nmoles · mL⁻¹ · min⁻¹, P = 0.009). IAF was positively correlated with HL activity (r = 0.431, P < 0.001). Multivariate analysis revealed a strong relationship between both the LIPC promoter genotype (P = 0.001) and IAF (P < 0.001) with HL activity. The relationship between IAF and HL activity for carriers and noncarriers of the A allele was curvilinear with the carriers having a lower apparent maximum level of plasma HL activity compared with noncarriers (138 versus 218 nmoles · mL⁻¹ · min⁻¹, P < 0.001). In addition, the LIPC A allele was associated with a significantly higher HDL-C-C (GA/AA = 16±7 versus GG = 11±5 mg/dL, P = 0.003). We conclude that the LIPC promoter A allele attenuates the increase in HL activity due to IAF in premenopausal women.

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Key Words: cholesterol ■ lipoprotein ■ visceral obesity ■ LIPC ■ triglyceride

Hepatic lipase (HL) is a 476 amino acid glycoprotein lipolytic enzyme that is a serine hydrolase. It appears to act not only in hydrolysis of triglyceride (TG) and phospholipid in HDL and LDL, but also as a ligand directing lipoproteins to hepatic cell surface receptors.1–5 HL is secreted by hepatocytes and anchored to the liver sinusoidal surface by heparin sulfate proteoglycans.6 The HL gene (LIPC) spans 35 kb of DNA, maps to chromosome 15q21, and is composed of nine exons and eight introns.7,8 HL plays a role in the metabolic processing of both HDL and LDL. HL acts to convert large, buoyant HDL to small, dense HDL by modulating the phospholipid content of the particle.9,10 By functioning as a ligand between the lipoprotein and cell surface receptors, HL has been shown to play a role in increased clearance of HDL particles11,12 and remnant lipoproteins.13 HL also catalyzes the hydrolysis of triglyceride and phospholipid in large, buoyant LDL forming small, dense more atherogenic LDL particles.14

Increased risk of premature coronary artery disease (CAD) has been shown to be associated with the presence of small, dense LDL particles.15–19 Similarly, low HDL-C is another major risk factor for CAD.20,21 It appears that gender differences in HDL-C levels may account, in part, for the temporal separation in CAD risk between men and women.20 HDL-C metabolism is modulated by both environmental and genetic factors that combine to account for the significant interindividual variation in HDL-C levels and hence CAD risk. Sedentary lifestyle,22 tobacco usage,23 and obesity24 are all associated with lower HDL-C levels. Forty to sixty percent of the variation in HDL levels appear to be genetic,25 and Cohen et al have demonstrated genetic linkage between the hepatic lipase promoter (LIPC) locus and HDL-C levels that account for approximately 25% of the interindividual variation in plasma HDL-C levels.26 HL activity appears to be influenced by several different factors including intra-abdominal fat (IAF),27 ethnic background,28,29 sex-steroid hormones,30–32 and LIPC genotype.28,29 Four polymorphisms have now been identified in the 5’ flanking region of the LIPC gene; a G to A substitution at position −250, C to T at −514, T to C at −710, and A to G at −763, which appear to be in complete linkage disequilibrium in white populations.33 The allele frequency of the substitution ranges from 0.153 to 0.2129 in white populations, 0.4520 to 0.5328 in African-Americans, and 0.47 in Japanese-Americans.29 This variation in allele frequency between ethnic groups may partially explain the observed higher
HDL-C levels seen in African-American and Japanese-American men. The less common haplotype of the LIPC gene promoter appears to be associated with decreased HL activity, increased HDL-C,29,33 and increased LDL buoyancy.29

Estrogen levels appear to influence HL activity significantly. Estrogen replacement in postmenopausal women has been shown to reduce HL activity by 31% back to premenopausal levels.32 The higher HL activity and the resultant decrease in HDL-C with menopause may account for some of the increased risk for CAD in the postmenopausal state. Differences in body composition may explain the gender differences in HL activity. Despres et al have shown that IAF content is strongly correlated with HL activity and that this association is independent of the effect of total body adiposity.27 Premenopausal women have less IAF than men,34 and the modestly higher HL activity seen in postmenopausal women15 may be due to the increase in central adiposity that accompanies the menopausal transition.36,37

In the present study, we investigated whether the LIPC promoter polymorphism and intra-abdominal fat content contributed independently or interactively to the variation of HL activity and plasma lipids in premenopausal women.

Methods

Subjects

Fifty-seven premenopausal white women, aged 21 to 54 years, were studied (Table 1). Forty-seven women are participating in a longitudinal, community-based study and had been selected by phone contact as described previously.38 The women were identified as premenopausal as assessed by daily menstrual diaries. Some were oligomenorrheic as they neared the menopausal transition but all had menstrual flows in the previous 6 months. These are baseline data for subjects participating in a longitudinal study of lipid metabolism in the menopausal transition. Ten additional premenopausal women were recruited through advertisements at the University of Washington as normal controls for a study of weight loss.

None of the women were taking any lipid-altering medications, including β-blockers or estrogen. They had no lipid disorders or other medical conditions affecting lipid metabolism, including diabetes, liver disease, or pregnancy. Women were excluded from the study if they had a body mass index (BMI) of ≥40 kg/m2, or were characterized as having small, dense LDL (phenotype B or I) by gradient gel electrophoresis.15,40 The Human Subjects Review Committee of the University of Washington approved the study protocol. Informed consent was obtained from all participants.

The mean age of these 57 women was 44.4 ± 6.6 years (Table 1) with a mean weight of 70.9 ± 14.5 kg and a BMI of 26.0 ± 4.7 kg/m2. Their percent total body fat was 36.9 ± 7.7% as measured by total body DEXA scan (see below). There was a wide range in IAF area (10.5 to 183.5 cm²) at the umbilicus as measured by computed tomography (CT) scan, with a mean IAF area of 66.5 ± 44.5 cm². The mean abdominal subcutaneous fat area was 239.7 ± 134.5 cm².

Blood Collection

Blood was collected in 0.1% ethylenediamine-tetraacetic acid after a 12- to 16-hour overnight fast for DNA isolation, lipoprotein measurements, lipase activity, and density gradient ultracentrifugation (DGUC). Insulin and glucose were collected in sodium heparin in supine subjects at 15, 20, and 25 minutes. A heparin bolus of 60 U/kg was given, and blood was collected after 10 minutes in lithium heparin tubes (Becton Dickinson) for the measurement of lipase activity. Blood was immediately centrifuged at 4°C at 3000 rpm for 15 minutes. Lipid measurements were made on fresh plasma within 2 days. Lipase activities and DGUC were obtained on plasma that had been immediately frozen and stored at −70°C.

<table>
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<tr>
<th>TABLE 1. Subject Characterization (n=57)</th>
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<td>Age (y)</td>
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<td>IAF (cm²)</td>
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<td>SQF (cm²)</td>
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<td>WHR</td>
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<td>Total body fat (%)</td>
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<td>HL (nmol · mL⁻¹ · min⁻¹)</td>
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<td>LPL (nmol · mL⁻¹ · min⁻¹)</td>
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<tr>
<td>Estradiol (pg/mL)*</td>
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<td>FSH (mU/mL)*</td>
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<td>Insulin (µU/mL)</td>
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<td>Glucose (mg/dL)</td>
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<td>Total chol (mg/dL)</td>
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<td>HDL₃-C (mg/dL)</td>
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<td>Apo B (mg/dL)</td>
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<td>Lp(a) (mg/dL)*</td>
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<td>LDL-R²*</td>
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Values are mean ± standard deviation. *n=47.

IAP indicates intra-abdominal fat by CT scan; SQF, subcutaneous abdominal fat by CT scan; WHR, waist-to-hip ratio; Total body fat (%), measured by DEXA scan; HL, hepatic lipase; LPL, lipoprotein lipase; and FSH, follicle stimulating hormone.

Lipid and Hormone Determinations

Plasma total cholesterol, TG, HDL-C, LDL-C, HDL₂-C, Apo B, and Lp(a) were measured at the Northwest Lipid Research Laboratory as previously described.41,42 Glucose was measured by the glucose oxidase method using Trinder reagent (Sigma) with an inter- and intra-assay coefficient of variation (CV) of <2%. Insulin was measured by radioimmune assay, as described previously.43 Briefly, this assay uses antisera raised to intact insulin in rabbits and iodinated insulin as tracer and has an inter- and intra-assay CV of 8% and 6%, respectively. Glucose and insulin were measured 3 times serially (15 minutes after insertion of intravenous line), with blood drawn 5 minutes apart on each subject and the mean of the 3 samples is reported. Estradiol was measured by solid-phase chemoluminescent immunoassay and follicle-stimulating hormone was measured by solid-phase 2-site immunometric assay by the University of Washington Laboratory Medicine Department.

Post Heparin Lipase Activity

The total lipolytic activity was measured in plasma after heparin bolus as previously described.46 Glycerol tri-1(14C) oleate (Amer sham) and lecithin were incubated with postheparin plasma for 60 minutes at 37°C, and the liberated C14 labeled free fatty acids were then extracted and counted. Lipoprotein lipase activity was calculated as the lipolytic activity removed from the plasma by the incubation with a specific monoclonal antibody against lipoprotein lipase (LPL), and HL activity was determined as the activity remaining after incubation with the LPL antibody. Enzyme activity is expressed as nanomoles of free fatty acid released per minute per milliliter of plasma at 37°C. For each assay, a bovine milk LPL standard was used to correct for interassay variation, and a human postheparin plasma standard was included to monitor interassay variation. Inter-assay coefficient of variation of hepatic lipase is 6%, between assays is 14%.
A discontinuous salt density gradient was created in an ultracentrifuge tube using a modification of a previous method. Samples were centrifuged at 65,000 rpm for 70 minutes (total $\alpha_{v}=1.95 \times 10^{3}$) at 10°C in a Beckman VTi 65.1 vertical rotor. Thirty-eight peaks were then collected from the bottom of the centrifuge tube, and cholesterol was measured in each fraction. The coefficient of variation of the Rf value obtained by replicate analysis was 3.6%, as described previously. The relative flotation rate (Rf), which characterizes LDL peak buoyancy, was adjusted for the total number of fractions collected. The coefficient of relative flotation rate (Rf), which characterizes LDL peak buoyancy, was measured in each fraction. The 0.45-mL fractions were then collected from the bottom of the centrifuge tube using a modification of a previous method. A single blinded observer made all the CT measurements of intra-abdominal fat.

**Statistical Methods**

Statistical analysis was performed using Sigma Stat, version 2.0 (Jandel Scientific). Comparisons between genotypes were performed using unpaired t-tests. Linear regression analysis was used to determine the effects of body composition, age, insulin, estradiol, and promoter variants on HL activity and the relationship between HL activity and lipids. In Table 2, HL activity was adjusted linearly for the effect of IAF (cm²). Multiple linear regression was used to obtain correlation coefficients and probability value in Table 3. Multiple regression was also used to examine the relationship between IAF, total body fat (by DEXA), and HL activity after accounting for the variance due to the LIPC genotype, age, and estradiol.

**Relationship Between HL Activity and IAF**

There was a significant positive relationship between HL activity and all measures of body adiposity (Table 2). IAF (cm²), as measured by CT scan, was strongly correlated with HL activity ($r=0.431, P<0.001$). Other measures of central adiposity (trunk dexta, subcutaneous abdominal fat, waist-to-hip ratio) were also highly correlated with HL activity with correlation coefficients ranging from 0.393 to 0.452 (all $P<0.002$) (Table 2). Insulin was marginally related to HL activity ($r=0.227, P=0.089$); however, in multivariate analysis, this relationship was abolished ($P=0.93$) after accounting for IAF.

To determine whether the measures of body adiposity related to HL activity were independent of IAF, HL activity was adjusted for IAF. As seen in Table 2, none of the

<table>
<thead>
<tr>
<th>TABLE 2. Linear Relationship Between HL or HL (Adjusted for IAF) With Potential Regulators of HL Activity</th>
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<tr>
<td><strong>HL Activity Versus</strong></td>
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<td>r</td>
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<td>BMI</td>
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<td>Total body fat (%)</td>
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<td>WHR</td>
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<tr>
<td>Trunk dexta (%)</td>
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<tr>
<td>IAF (cm²)</td>
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<td>SOF (cm²)</td>
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<tr>
<td>Age</td>
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<tr>
<td>Insulin</td>
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<tr>
<td>Estradiol</td>
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<tr>
<td>LIPC variant</td>
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</table>

*HL was adjusted for the linear relationship with IAF.

Abbreviations and units of measure are listed in legend of Table 1. Trunk dexta indicates truncal abdominal fat by DEXA.

DGUC

DNA was extracted from leukocytes of 10 mL freshly drawn blood by the method of Poncz et al. DNA was extracted from leukocytes of 10 mL freshly drawn blood by the method of Poncz et al.

**DNA Isolation and Analysis**

DNA was extracted from leukocytes of 10 mL freshly drawn blood by the method of Poncz et al. The polymorphism at nucleotide position −250 was determined by polymerase chain reaction amplification using a primer pair, as described previously.

**Body Composition**

Body composition was measured by CT scan (GE Highspeed Advantage) and DEXA scan. A single image was obtained by CT scan on inspiration at the level of the umbilicus. The CT image was analyzed for cross-sectional area of fat using a density contour scan on inspiration at the level of the umbilicus. The CT image was analyzed for cross-sectional area of fat using a density contour scan on inspiration at the level of the umbilicus. The laboratory has an interassay CV of 1.6%, 1.3%, and 1.3% for fat mass, lean mass, and % body fat, respectively.

**RESULTS**

**TABLE 3. Relationship Between HL and IAF With Lipoproteins (n=57)**

<table>
<thead>
<tr>
<th>Linear Regression</th>
<th>Multiple Linear Regression</th>
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<tbody>
<tr>
<td><strong>HL Activity Versus</strong></td>
<td><strong>IAF (cm²) Versus</strong></td>
</tr>
<tr>
<td>r</td>
<td>p</td>
</tr>
<tr>
<td>Total Chol</td>
<td>0.025</td>
</tr>
<tr>
<td>TG</td>
<td>0.162</td>
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<tr>
<td>VLDL-C</td>
<td>0.166</td>
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<tr>
<td>LDL-C</td>
<td>0.189</td>
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<tr>
<td>HDL-C</td>
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<tr>
<td>HDL-C</td>
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<tr>
<td>HDL-C</td>
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<tr>
<td>Lp(a)</td>
<td>0.014</td>
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<tr>
<td>Apo B</td>
<td>0.222</td>
</tr>
<tr>
<td>LDL-Rf</td>
<td>0.348</td>
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</table>

Units of measure are listed in legend of Table 1. A single blinded observer made all the CT measurements of intra-abdominal fat.

Total body fat (%) was measured using a dual x-ray DEXA scan (Hologic QDR 1500). The laboratory has an interassay CV of 1.6%, 1.3%, and 1.3% for fat mass, lean mass, and % body fat, respectively.

A discontinuous salt density gradient was created in an ultracentrifuge tube using a modification of a previous method. Samples were centrifuged at 65,000 rpm for 70 minutes (total $\alpha_{v}=1.95 \times 10^{3}$) at 10°C in a Beckman VTi 65.1 vertical rotor. Thirty-eight peaks were then collected from the bottom of the centrifuge tube, and cholesterol was measured in each fraction. The relative flotation rate (Rf), which characterizes LDL peak buoyancy, was obtained by dividing the fraction number containing the LDL-C peak by the total number of fractions collected. The coefficient of variation of the Rf value obtained by replicate analysis was 3.6%, as described previously.

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**Statistical Methods**

Statistical analysis was performed using Sigma Stat, version 2.0 (Jandel Scientific). Comparisons between genotypes were performed using unpaired t-tests. Linear regression analysis was used to determine the effects of body composition, age, insulin, estradiol, and promoter variants on HL activity and the relationship between HL activity and lipids. In Table 2, HL activity was adjusted linearly for the effect of IAF (cm²). Multiple linear regression was used to obtain correlation coefficients and probability value in Table 3. Multiple regression was also used to examine the relationship between IAF, total body fat (by DEXA), and HL activity after accounting for the variance due to the LIPC genotype, age, and estradiol. This method computes a series of simultaneous normal linear equations that are soluble using inverse matrix algebra. Hardy-Weinberg equilibrium was tested using $x^2$ analysis. The significance level was set at $\alpha=0.05$.

An apparent $V_{max}$ for the HL activity was calculated using the Wolff linearization of the Michealis-Menton curve for enzyme kinetics. An apparent $V_{max}$ for the HL activity was calculated using the Wolff linearization of the Michealis-Menton curve for enzyme kinetics. An apparent $V_{max}$ for the HL activity was calculated using the Wolff linearization of the Michealis-Menton curve for enzyme kinetics.
measures of adiposity were related to HL activity after adjustment for IAF. Measures of body fat were highly correlated (eg, IAF and total body fat, \( r=0.735 \)), as IAF is included in the total body fat measurement. Therefore, in an attempt to determine which of these 2 measures, or both, relate to hepatic lipase activity, multivariate analysis was performed. In multiple linear analysis IAF \( (P=0.031) \) remained significantly related to HL activity, whereas total body fat (by DEXA) \( (P=0.48) \) was no longer significant. In addition, the relationship between insulin and HL activity was not independent of IAF.

**Relationship Between HL Activity and LIPC Genotype**

When the women are categorized based on the presence or absence of the rare LIPC (A) allele, there was a significant difference in HL activity \( (145\pm77 \text{ versus } 104\pm34 \text{ nmoles} \cdot \text{mL}^{-1} \cdot \text{min}^{-1}) \) in GG versus GA and AA combined, respectively (Figure 1). The GG genotype was associated with a 39% higher HL activity as compared with the GA and AA genotypes combined \( (P=0.009) \). The frequency of the A allele is 0.17, and the population sample studied is in Hardy-Weinberg equilibrium \( (x^2=0.04, P=0.98) \).

As previously seen in men,\(^{29}\) there appears to be an absence of women with higher levels of HL activity among carriers of the A allele compared with those having the GG genotype (Figure 1). The range of HL activity was much wider among the subjects with the GG genotype \( (47 \text{ to } 281 \text{ nmoles} \cdot \text{mL}^{-1} \cdot \text{min}^{-1}) \) as compared with carriers of the A allele \( (53 \text{ to } 166 \text{ nmoles} \cdot \text{mL}^{-1} \cdot \text{min}^{-1}) \). In the univariate analysis (Table 2), the polymorphism in the promoter region of the LIPC gene accounted for 12% \( (r^2=0.118) \) of the variance in the HL activity in these premenopausal women.

**Combined Effects of the LIPC Genotype and IAF on HL Activity**

Multivariate analysis revealed that both IAF \( (P=0.001) \) and the LIPC genotype \( (P=0.001) \) were significantly associated with HL activity \( (r=0.577) \), after accounting for each other. These 2 variables accounted for 33% of the variance in HL activity.

Within each genotype, there was a highly significant curvilinear relationship between HL activity and IAF \( (GG, r=0.779; GA/AA, r=0.873; P<0.001) \) (Figure 2). With increasing IAF, HL activity increases to a different apparent maximum depending on genotype. Using the Wolff linearization of Michaelis-Menten kinetics (see Methods), one can calculate a maximal level for hepatic lipase activity within each genotype. The maximal level for the GG genotype was significantly higher than that for the carriers of the A allele \( (218 \text{ versus } 138 \text{ nmoles} \cdot \text{mL}^{-1} \cdot \text{min}^{-1}, P<0.001) \). At high levels of IAF, there was a wide divergence of the HL activity between the 2 genotypes.

**Relationship of Lipids to HL Activity, LIPC Variants, and IAF**

There were also differences between genotypes in total cholesterol \( (176\pm29 \text{ versus } 200\pm22 \text{ mg/dL in GG versus GA/AA, respectively}) \) that were confined to differences in the HDL-C subfractions (Table 4). The significantly lower HL activity in the carriers of the A allele was accompanied by a statistically significant increase \( (23\%) \) in HDL-C in comparison with the noncarriers \( (53\pm12, P=0.003) \), as well as higher HDL-C and HDL-C levels. HDL-C and HDL-C levels were 45\% \( (16\pm7 \text{ versus } 11\pm5, P=0.003) \) and 17\% \( (49\pm10 \text{ versus } 42\pm8, P=0.009) \) higher, respectively, in the carriers of the A allele compared with the noncarriers. Univariate analysis showed that HDL-C was correlated with HL activity \( (r=0.323, P=0.01) \) along with HDL-C \( (r=0.449, P<0.001) \) but HDL-C \( (r=0.201, P=0.13) \) was not correlated (Table 3).

LDL-Rf, a measure of LDL particle peak density, was not statistically different between genotypes. This may be due to the fact that women were excluded from the study if they had small, dense LDL, therefore truncating the sample such that there was not enough statistical power to demonstrate the effects seen in males. HL activity was related to LDL-Rf \( (r=0.348, P=0.016) \) such that the higher HL activity was
associated with more dense LDL. HL activity was not related to total cholesterol, LDL-C, TG, Apo B, or Lp(a).

The relationship between IAF and lipid measures were all significant with the exception of Lp(a) (Table 3). In multivariate analysis, with both IAF and HL activity in the model, the relationship between IAF and the HDL subfractions disappeared. All other measures that were related to IAF in univariate analysis remained significant after accounting for HL activity. The relationship between HL and HDL_{2-C} remained significant in multivariate analysis, but all other lipids measures were no longer related to HL activity after accounting for IAF.

### Discussion

In the present study, the investigation of the relationship between the −250 LIPC polymorphism, central adiposity, and HL activity in premenopausal women leads to 3 conclusions. First, central adiposity and the LIPC gene promoter polymorphism are both associated with variation in HL activity in women. Second, the relationship between central obesity and HL activity is different depending on the LIPC promoter polymorphism, such that the presence of the A allele appears to attenuate the increase in HL activity with high intra-abdominal fat levels. Third, there is a strong relationship between IAF and Apo B containing particles (TG, VLDL-C, LDL-C), but the relationship between IAF and HDL_{2-C} appears to be mediated through their association with HL activity.

We have shown that central adiposity and the polymorphism at position −250 of the LIPC gene are both associated with plasma HL activity. In univariate analysis, central obesity is positively correlated with HL activity (Table 2), whereas the presence of the LIPC A allele is negatively associated with HL activity. These relationships remain significant in multivariate analysis revealing that both the LIPC genotype and IAF may act to regulate HL activity. Another demonstration of the relationship of the LIPC promoter variants with HL activity is seen (Table 2) after adjusting for the effects of IAF on HL and finding that the relationship between the LIPC variants and HL activity remains.

Although there was a significant linear relationship between HL activity and IAF in the GG genotype ($r = 0.537$) and the combined GA and AA genotypes ($r = 0.432$), we found that the curvilinear relationships fit the data better ($r = 0.779$ and 0.873, respectively) (Figure 2). A maximal level of HL activity is approached as IAF increases in the 2 groups, and the maximum HL activity is 58% lower in the presence of the A allele than in its absence. The lower maximal level of HL activity in the carriers of the A allele is compatible with the lower mean hepatic lipase activity seen in both men and women$^{28,29}$ and the truncated range of expression seen in normal men and men with CAD$^{29}$ who carry the A allele.

It has been previously shown that HL activity is regulated by increasing amounts of intra-abdominal fat.$^{27}$ Several groups have also shown that the LIPC promoter polymorphisms, all in linkage disequilibrium in whites, are associated with HL activity.$^{28,29,53,54}$ Our data show that the LIPC promoter polymorphism influences the relationship between IAF and HL activity. The difference in HL activity between the 2 genotypes is most evident when 2 curves diverge at relatively high levels of IAF (Figure 2). In fact, the maximal expression of HL activity between genotypes is highly significant ($P < 0.001$). Thus the potential benefit of the LIPC promoter polymorphism to limit HL activity is most evident in women with higher levels of intra-abdominal fat. These results suggest that the less common promoter variant is not as transcriptionally active as the common allele, which may place a limit on gene expression capacity at higher IAF values.

The presence of the A allele also confers benefit to the lipid profile, which is characterized by a 23% higher HDL-C, mainly due to a 45% higher HDL_{2-C}, the most antiatherogenic HDL particle.$^{55}$ There does not appear to be any significant effect of the LIPC promoter variants on the Apo B containing particles, including VLDL or LDL in these women.

High HL activity is associated with a more atherogenic lipid profile characterized by reduced HDL_{2-C} and increased amounts of small, dense LDL. Tahvanainen et al have shown that carriers of the A allele have significantly higher amounts of triglyceride in IDL, LDL, and HDL particles, making them more buoyant among subjects with lower HL activities.$^{54}$ In this study, we observed strong positive correlations between HL activity and HDL-C and HDL_{2-C} levels. This is accompanied by a strong negative correlation between HL activity and LDL-Rf. All of these differences suggest that high HL activity is potentially atherogenic.

There are also strong correlations between the presence of central adiposity and lipid measures. IAF is positively correlated with TG, VLDL-C, and Apo B and negatively related to HDL_{2-C} and LDL-Rf. To determine whether HL activity and IAF both effect these lipids, after accounting for the other, multiple regression was performed. IAF remained associated...
Figure 3. Hepatic lipase mediates the combined effects of IAF and LIPC promoter genotype on atherogenic lipoproteins. Elevated IAF is associated with higher hepatic lipase activity. The magnitude of this association is dependent on the LIPC genotype. Higher hepatic lipase activity promotes the conversion of HDL₂ to HDL₃ and the conversion of large, buoyant to small, dense LDL. In addition to the effect mediated by hepatic lipase, IAF is associated with a residual increase in small, dense LDL. This pathway is related to an increase in plasma triglyceride that stimulates cholesterol ester transfer protein mediated transfer of neutral lipids leading to small, dense LDL.

with measures of Apo B containing particles (TG, VLDL-C, LDL-C, Apo B), whereas HL remained associated with HDL₃-C. The effects of IAF on HDL₃-C particles were reduced, suggesting that HL activity may mediate the relationship between central obesity and HDL₃-C metabolism.

We propose a model that describes the relationship between central adiposity and hepatic lipase with a downstream effect on lipoproteins (Figure 3). Hepatic lipase increases as a function of IAF. The magnitude of this increase depends on the LIPC genotype. Higher HL activity increases the conversion of HDL₂ to HDL₃ and the conversion of large, buoyant LDL to small, dense LDL. The link between IAF and HDL₃-C metabolism appears to be mediated, at least in part, by hepatic lipase. There is an additional influence of IAF on LDL density beyond the effect of hepatic lipase, perhaps by altering the synthesis of TG-rich lipoproteins.

This cohort of healthy, premenopausal women has been rigorously characterized. They have been carefully selected as to their menopausal status, and they have a wide range of body fat content. The mean BMI of these women is 26.0 kg/m², which closely resembles that of the American female population of the same age (26.1 kg/m²). Serum estradiol and follicle-stimulating hormone levels were not associated with HL activity, perhaps due to the wide fluctuations in estradiol that occur in these menstruating women who were not studied on coordinated phases of their menstrual cycle. However, estradiol was correlated with HL activity in multivariate analysis as it contributed to the relationship between HL and IAF.

Others have shown a relationship between HL activity and insulin. As seen in Table 2, all measures of body adiposity were significantly related to HL activity. Nie et al have shown that HL activity is related to BMI, and we confirm this. In addition, we sought to investigate the relationship of different fat depots with HL activity. Measures of central adiposity, trunk DEXA, and waist-to-hip ratio were highly correlated with HL activity but measure both IAF and subcutaneous fat. Depres et al previously demonstrated that IAF is related to HL activity, independent of total body adiposity. Our data confirm that the relationship between HL activity and IAF is independent of total body adiposity. These data suggest that the IAF depot may be an important regulator of HL activity.

In conclusion, in these normal white women, the relationship between hepatic lipase and IAF is modified by the presence of promoter variants in the LIPC gene. The LIPC A allele attenuates the expression of hepatic lipase in subjects with high IAF. Thus, in subjects with large amounts of central fat deposition, the LIPC promoter variants affect hepatic lipase levels and the subsequent levels of atherogenic lipoproteins. This study indicates a potential differential genetic susceptibility to increased atherogenic risk factors in the presence of central obesity.

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