Published in final edited form as:

Clin Chim Acta. 2009 April ; 402(0): 83–87. doi:10.1016/j.cca.2008.12.024.

The apolipoprotein A5 –1131T>C promoter polymorphism in Koreans: Association with plasma APOA5 and serum triglyceride concentrations, LDL particle size and coronary artery disease

Yangsoo Jang^{a,b,c,1}, Jean Kyung Paik^{d,e,1}, Yae Jung Hyun^{c,d}, Jey Sook Chae^{c,d}, Ji Young Kim^{c,d}, Ju Ree Choi^d, Sang Hak Lee^a, Dong-Jik Shin^{b,c}, Jose M. Ordovas^f, and Jong Ho Lee^{c,d,e,*}

^aCardiology Division, Cardiovascular Center, Yonsei University College of Medicine, Seoul, Republic of Korea

^bCardiovascular Genome Center, Yonsei University College of Medicine, Seoul, Republic of Korea

^cYonsei University Research Institute of Science for Aging, Yonsei University, Seoul, Republic of Korea

^dNational Research Laboratory of Clinical Nutrigenetics/Nutrigenomics, Department of Food and Nutrition, College of Human Ecology, Yonsei University, Seoul, Republic of Korea

^eDepartment of Food and Nutrition, Brain Korea 21 Project, College of Human Ecology, Yonsei University, Seoul, Republic of Korea

^fNutrition and Genomics Laboratory, JM-USDA-HNRCA, Tufts University, Boston, MA, United States

Abstract

Background—The association between –1131T>C single nucleotide polymorphism (SNP) of the apolipoprotein A5 gene (*APOA5*) and hypertriglyceridemia raised the possibility that this SNP could be related to coronary artery disease (CAD) risk. Therefore, we investigated the association of this *APOA5* –1131T>C SNP with circulating concentrations of APOA5, triglyceride and CAD in Koreans.

Methods—CAD patients ($n = 741$) and age-, sex-matched healthy controls ($n = 741$) were genotyped for the *APOA5* –1131T>C SNP. The main outcome measures were the odds ratio (OR) on CAD risk and lipid variables, APOA5 concentration and LDL particle size.

Results—The presence of the minor allele at the –1131T>C SNP was associated with an increased risk of CAD [OR 1.34 (95% CI, 1.09–1.65), $P = 0.007$] after adjusting for BMI, alcohol

consumption, systolic blood pressure and diastolic blood pressure. There was an association between the APOA5 concentration and the -1131T>C genotype in controls (T/T: 245 ± 7 ng/ml, T/C: 220 ± 6 , C/C: 195 ± 12 ; $P = 0.001$) and CAD patients (T/T: 218 ± 8 ng/ml, T/C: 185 ± 7 , C/C: 169 ± 12 ; $P < 0.001$). Subjects with T/C or C/C in control and CAD patient groups showed higher triglyceride than those with T/T genotype. Also, the -1131T>C polymorphism was associated with LDL particle size ($P = 0.003$), with the T/C or C/C controls having smaller size than the T/T controls.

Conclusions—The APOA5 -1131C allele is associated with reduced APOA5 concentration and with increased CAD risk. This is consistent with the observed association between the -1131C SNP, increased triglycerides as well as small LDL particle size.

Keywords

APOA5 -1131T>C polymorphism; Coronary artery disease; Triglycerides; Apolipoprotein A5 concentration; LDL particle size

1. Introduction

Apolipoprotein A5 (APOA5) has emerged as a significant modulator of serum triglyceride (TG) concentration. Overexpression of plasma APOA5 in mice resulted in decrease in plasma triglyceride concentrations, whereas in the APOA5 knockout mice TG concentrations were increased [1–3], supporting that APOA5 has an important role in triglyceride metabolism. In humans, the apoA5 gene (APOA5) variation has also been consistently associated with serum TG concentrations across ethnic groups [1,4–7].

Epidemiological and clinical studies have shown that increased TG concentrations are related to an increased risk of coronary artery disease (CAD) [8,9]. Consistent with this notion, it has been shown that the APOA5 -1131T>C SNP, probably through its association with hypertriglyceridemia, was associated with an increased risk of CAD in multiple ethnic population [10–13]. However, there is an also discrepant report of no association between APOA5 -1131C SNP and CAD risk [14]. Moreover, Talmud et al. [15] have recently reported that APOA5 concentrations were not associated with -1131T>C and APOA5 genotypes do not appear to have an impact on the development of type 2 diabetes. Therefore, the relation between APOA5 genotypes and concentration, hypertriglyceridemia and CAD remains poorly defined and understood. Consequently, our goal in the current study was to investigate whether subjects carrying this genetic variant would have decreased concentration of APOA5, hypertriglyceridemia and higher risk of CAD. We also examined its association with LDL particle size and HDL-cholesterol.

2. Materials and methods

2.1. Study population

In this study, we included 741 consecutive CAD patients and age-, sex-matched 741 healthy controls from an unrelated and ethnically homogenous Korean population. CAD patients were recruited from the Cardiovascular Genome Center, Yonsei University Severance Hospital, Seoul, Korea and the Division of Cardiology, National Health Insurance

Corporation Ilsan Hospital, Goyang, Korea. The inclusion criteria were as follows: (a) angiographically confirmed coronary artery disease with 50% occlusion of 1 major coronary arteries, (b) myocardial infarction (MI) confirmed according to the World Health Organization (WHO) criteria for symptoms, enzyme elevation, or electrocardiographic changes, (c) absence of nonatherogenic occlusion, such as osteal stenosis and spasm, (d) no orthopedic limitations or any diagnosis of diabetes mellitus, liver disease, renal disease, thyroid or pituitary disease, (e) no acute or chronic inflammatory disease.

Healthy control subjects were recruited from the Health Service Center during routine check-up visits or through advertisements briefly describing the study design. The inclusion criteria were no history of MI and no evidence of MI by echocardiogram. Individuals with any history or diagnosis of vascular disease, diabetes mellitus, cancer (clinically or by anamnesis), renal disease, liver disease, thyroid disease, or acute or chronic inflammatory disease were excluded. None of the healthy subjects were taking antidiabetic medications.

Before participation, the purpose of the study was carefully explained to all participants and their informed consent was obtained. The study protocol complied with the Guidelines for Genome/Genetic Research issued by the Korean government and was approved by the Institute of Review Board of Yonsei University, Seoul, Korea. The study was carried out in accordance with the Helsinki Declaration.

2.2. Anthropometric parameters, blood pressure measurements, and blood collection

Body weight and height were measured unclothed and without shoes in the morning. Blood pressure was obtained from the left arm of seated patients with an automatic blood pressure monitor (TM-2654, A&D, Tokyo, Japan) after 20 min of rest. Venous blood specimens were collected in EDTA-treated and plain tubes after an overnight fast and were stored at -70°C until analysis.

2.3. APOA5 -1131T>C genotyping

Genomic DNA was extracted from 5 ml of whole blood using a commercially available DNA isolation kit (WIZARD^R Genomic DNA purification kit, Promega Corp., Madison, WI) according to the manufacturer's protocol. -1131T>C genotyping was performed by SNP-IT^{TM} assays using single primer extension technology (SNPstream 25K^{TM} System, Orchid Biosystems, NJ). The results of yellow and/or blue color developments were analyzed with ELISA reader and the final genotype calls were made with QCRReview^{TM} program.

2.4. Serum lipid profiles

Blood fasting serum concentrations of total cholesterol and triglycerides were measured using commercially available kits on a Hitachi 7150 Autoanalyzer (Hitachi Ltd. Tokyo, Japan). After using dextran sulfate magnesium to precipitate serum chylomicron, low-density lipoprotein (LDL), and high-density lipoprotein (HDL) cholesterol from the supernatant were measured by an enzymatic method. LDL cholesterol was indirectly estimated in subjects with serum triglyceride concentrations $<4.52\text{ mol/l}$ (400 mg/ml) using

the Friedewald formula. In subjects with serum triglyceride concentrations ≥ 4.52 mol/l, LDL cholesterol was measured by an enzymatic method on a Hitachi 7150 Analyzer directly. Serum apolipoprotein A-I and B were determined by turbidometry at 340 nm using a specific anti-serum (Roche, Switzerland).

2.5. Plasma apolipoprotein A5

Plasma apolipoprotein A5 (APOA5) concentration was measured using an enzyme immunoassay (Human Apolipoprotein A ELISA Kit, Millipore, MO). The resultant color reaction was read at 450 nm using a Victor² (Perkin Elmer life sciences, Turku, Finland). The intra- and inter-assay CVs were 2.46% and 7.45%, respectively.

2.6. Plasma LDL particle size

Particle size distribution of LDL (1.019–1.063 g/ml) isolated by sequential flotation ultracentrifugation was examined by a pore-gradient lipoprotein system (CBS Scientific, CA) using commercially available non-denaturing polyacrylamide slab gels containing a linear gradient of 2–16% acrylamide (Alamo Gels Inc., San Antonio, TX). Standards of latex beads (34 nm), thyroglobulin (17 nm), apoferritin (12.2 nm) and catalase (10.4 nm) were used to estimate the relative migration (Rf) rates of each band. The gels were scanned by GS-800 Calibrated Imaging Densitometer (Bio-Rad Laboratories, Graz, Austria). LDL particle size was calculated with reference to the Rf value of the standards.

2.7. Statistical analyses

Statistical analyses were performed using SPSS ver. 12.0 for Windows (Statistical Package for the Social Science, SPSS Inc., Chicago, IL). Hardy–Weinberg Equilibrium (HWE) was examined by Haploview ver. 3.32 (<http://www.broad.mit.edu/mpg/haploview/>). Differences in clinical and metabolic variables between controls and CAD patients were tested by independent *t*-test and a general linear model to adjust for covariates. A χ^2 test was used to determine whether there were differences in cigarette smoking, alcohol consumption, and medication therapy among the genotype and between controls and CAD patients. Genotype distributions and allele frequencies were compared between controls and CAD patients by χ^2 test. The association between CAD and genotype was calculated using the odds ratio (OR) [95% confidence intervals (CIs)] of a χ^2 test and a logistic regression analysis with adjustment for sex, age, BMI, smoking, alcohol consumption, and diastolic blood pressure. One-way analysis of variance (ANOVA) followed by Bonferroni method was used to compare the differences among genotype groups in control subjects and CAD patients. We initially determined whether each variable presented a normal distribution before statistical testing, and then performed logarithmic transformation on the skewed variables. For descriptive purposes, mean values are presented using untransformed values. Results are expressed as mean $\pm$ S.E and a 2-tailed value of $P < 0.05$ was considered statistically significant.

3. Results

3.1. Characteristics of controls and CAD patients

General characteristics of the 741 CAD patients and the 741 age- and sex-matched controls are shown in Table 1. Drinking alcohol was more common in controls than in cases. Lipid-lowering drugs (LLD) (66% vs. 0%, patients and controls, respectively), antihypertensive drugs (80% vs. 6%), and antiplatelet drugs (81% vs. 2%) were used more frequently in patients. Likewise, systolic blood pressure and diastolic blood pressure were lower in CAD patients than controls. CAD patients had lower concentrations of total cholesterol, HDL cholesterol, LDL cholesterol, APOA5, apoA1, apoB, and glucose and higher concentrations of triglyceride, insulin, and HOMA-IR and smaller LDL particle size than controls both before and after adjustment for BMI, alcohol consumption, systolic blood pressure and diastolic blood pressure.

When we subdivided CAD patients into 2 groups: those not treated with lipid-lowering drugs (LLD) ($n = 249$) and those treated with LLD ($n = 492$), CAD patients not treated with LLD showed higher concentrations of total cholesterol (191 ± 3 vs. 162 ± 2 mg/dl; $P < 0.001$), LDL-cholesterol (115 ± 2 vs. 89 ± 2 mg/dl; $P < 0.001$), and apoB (87 ± 2 vs. 72 ± 1 mg/dl; $P < 0.001$) than those treated with LLD. There were no significant differences in APOA5 (201 ± 10 vs. 196 ± 6 ng/ml), HDL-cholesterol (45 ± 1 vs. 44 ± 1 mg/dl), apoA1 (132 ± 1 vs. 130 ± 1 mg/dl), triglyceride (158 ± 7 vs. 148 ± 4 mg/dl) and LDL particle size (25.1 ± 0.09 vs. 25.1 ± 0.06 nm) between CAD patients not treated with LLD and those treated with LLD.

3.2. Distribution of APOA5 –1131T>C polymorphism

The genotype distribution was in Hardy–Weinberg equilibrium in the entire population as well as in cases and controls separately. In healthy controls, the minor allele frequency of the –1131T>C was 0.29, which was consistent with a previous observation in a Korean population [16]. The genotypic distribution of the –1131T>C polymorphism was significantly different between controls and CAD patients ($P = 0.005$) (Table 2). The minor allele frequency was significantly higher in CAD patients (0.34) as compared with controls (0.29; $P = 0.003$). Based on these results, carriers of minor C allele were compared with T/T homozygotes and these subjects showed a significantly higher risk of CAD [OR 1.40 (95% CI, 1.14–1.72), $P = 0.001$] (Table 3). The significance of the association remained after adjusting for BMI, alcohol consumption, systolic blood pressure and diastolic blood pressure. [OR 1.34 (95% CI, 1.09–1.65), $P = 0.007$].

3.3. Clinical characteristics and APOA5 concentration associated with the –1131T>C genotype

There were no significant genotype-related differences among control subjects having the –1131T>C genotype with respect to age, BMI, smoking or alcohol consumption (data not shown). Similarly, in CAD patients differences among genotype groups were not found with regard to age, BMI, smoking, alcohol consumption or pharmacological interventions (data not shown). Fig. 1 shows the influence of the –1131T>C genotype on APOA5 concentration in controls and CAD patients. There was a significant association between the APOA5

concentration and the -1131T>C genotype in controls (T/T: 245 ± 7 ng/ml, T/C: 220 ± 6 , C/C: 195 ± 12 ; $P = 0.001$) and CAD patients (T/T: 218 ± 8 ng/ml, T/C: 185 ± 7 , C/C: 169 ± 12 ; $P < 0.001$). Subjects with the C/C genotype showed lower APOA5 concentration than those with the T/T genotype.

3.4. Lipid profiles, LDL particle size, and glucose associated with the -1131T>C genotype

There was a significant association between the triglyceride concentration and the -1131T>C genotype in controls (T/T: 129 ± 3 mg/dl, T/C: 151 ± 5 , C/C: 172 ± 14 ; $P < 0.001$) and CAD patients (T/T: 135 ± 4 mg/dl, T/C: 156 ± 5 , C/C: 198 ± 17 ; $P < 0.001$) (Fig. 1). Subjects with the T/C or C/C genotype in control and CAD patient groups showed significantly higher triglyceride concentration than those with the T/T genotype. In control subjects, the APOA5 -1131T>C promoter polymorphism was associated with LDL particle size (T/T: 25.7 ± 0.06 nm, T/C: 25.6 ± 0.07 , C/C: 25.1 ± 0.16 ; $P = 0.003$) (Fig. 1), with the C/C controls having smaller size than the T/T or T/C controls ($P = 0.003$). In CAD patients, there was a tendency of association of the -1131T>C genotype with LDL particle size (T/T: 25.2 ± 0.07 nm, T/C: 25.1 ± 0.07 , C/C: 24.8 ± 0.17 ; $P = 0.055$) (Fig. 1). However, there were no genotype-related differences for total cholesterol, HDL-cholesterol, LDL-cholesterol, apoA1, apoB, glucose, insulin, and HOMA-IR in controls and CAD patients (Table 4).

4. Discussion

Our study supports a significant role of the APOA5 locus as a genetic factor for CAD risk. Our age- and sex-matched case-control design in Korean subjects reveals that the minor allele frequency of the -1131T>C SNP in the promoter of the APOA5 gene was significantly increased in patients with CAD as compared with controls, after adjusting for BMI, alcohol consumption, systolic blood pressure and diastolic blood pressure. Moreover, plasma APOA5 concentrations were lower in CC subjects as compared with TT subjects in both controls and CAD patients. Thus, these lower APOA5 concentrations may mediate the increased in CAD risk observed in association with the APOA5 -1131C allele through mechanisms involving the regulation of serum triglyceride concentrations [17,18], which are known to be correlated with the risk of atherosclerosis in a variety of studies [19,20].

The association observed in our study between the APOA5 -1131C allele and increased risk of CAD is consistent with previous reports in Hungarian subjects [10] and Chinese [11]. However, other studies did not find such association [21,22]. This discrepancy could be explained by the different genotype distribution of the APOA5 -1131T>C polymorphism in distinct ethnic populations. Indeed, the C allele is found at much higher frequency in Koreans (0.28) [16] as compared with Caucasians (<0.10) [1,4,10,12,23]. In addition, Korean men aged 30–69 years have maintained relatively low mean concentrations of total cholesterol (187 mg/dl) and LDL cholesterol (114 mg/dl) over the past 10 years [24], but mean serum concentration of triglyceride have increased approximately from 141 mg/dl in 1998 to 168 mg/dl in 2005 [24]. Furthermore, CAD mortality rates in Korea have dramatically increased from 13.8 per 100,000 in 1998 to 27.5 per 100,000 in 2005 [25].

Therefore, under these circumstances, the presence of the *APOA5* –1131C allele may have more impact on CAD risk in Koreans.

Consistent with previous reports [1,4–6,11,16,26,27], homozygosity for the –1131C allele was associated with 33% and 47% higher triglyceride as compared with TT subjects in both controls and CAD patients. Conversely, the association with *APOA5* concentrations was in the opposite direction, with CC subjects having significantly lower concentrations than TT subjects in both controls and cases. Overexpression of the *APOA5* gene in mice led to decreased plasma triglyceride concentrations, whereas its disruption resulted in hypertriglyceridemia [1,2], which parallels the associations observed in humans. More mechanistic information has been provided by Grosskopf et al. [18] with *APOA5*-deficient mice, reporting that *APOA5* significantly impacts triglyceride-rich lipoprotein catabolism without having much effect on their synthesis. On the other hand, Schaap et al. [17] concludes that *APOA5* influences TG secretion without affecting apoB secretion. Moreover, these investigators reported a relation between *APOA5* and lipoprotein lipase (LPL) activity. This is consistent with our data showing that *APOA5* genotype was not predictor of apoB concentrations.

APOA5-deficient mice have shown decreased LPL activity and the accumulation of larger very low density lipoprotein (VLDL) particles [18], which are precursors of small, dense LDL. The present study showed that subjects with the TC or CC genotype in controls had smaller size of LDL particles than did control subjects with the TT genotype, despite the lack of significant differences in their respective LDL cholesterol concentrations. This is consistent with our previous findings [16] of a significant association between the *APOA5* –1131C allele and increased concentrations of small, dense LDL concentrations in healthy men. This difference may reflect that the CAD progression is related to small LDL particle size [28,29] regardless of the –1131T>C genotype.

In this study, subjects with C/C genotype showed slightly, but not significantly, lower HDL cholesterol concentration than those with the T/T genotype. There are discrepant results of an association between the *APOA5* –1131T/C and HDL cholesterol: Endo et al. [6] and Baum et al. [7] reported lower HDL-cholesterol concentrations associated with the T/C or C/C genotype however, other studies by Pennachio et al. [1] and Nabika et al. [30] did not find significant differences between genotypes. These inter-study differences could be related to sex differences in the expression of this association. In this regard, Evans et al. [31] have found that the HDL-lowering effect of the –1131T>C polymorphism is lesser in men than in women. Therefore, a high proportion of males (90%) in this study could explain a weak association between *APOA5* –1131T>C genotypes and HDL-cholesterol.

It has to be noted that our results share the limitations of cross-sectional, observational studies. We evaluated associations, rather than prospective prediction. Thus, cause–effect relationships cannot be easily established between *APOA5* –1131T>C genotypes, *APOA5* concentration and other biochemical factors. Despite these limitations, our data showed a confirmatory result of *APOA5* –1131T>C related to circulating triglyceride concentrations as shown in multiple independent populations [1,4–6,11,16,25,26] and an interesting

association between *APOA5* –1131C variant and increased risk for CAD as well as significantly reduced concentrations of APOA5 and small LDL particle size.

Acknowledgements

This work was supported by the Korea Science and Engineering Foundation (KOSEF) grant (M10642120002-06N4212-00210), Ministry of Science and Technology, Seoul, Korea, National Research Laboratory project #R0A-2005-000-10144-0, Ministry of Science and Technology, Seoul, Korea, Korea Health 21 R&D Projects, Ministry of Health & Welfare (A000385), Seoul, Korea, Brain Korea 21 Project, College of Human Ecology, Yonsei University, Seoul, Korea, Grant 58-1950-9-001 from the US Department of Agriculture Research Service, US and NIH grant HL54776, MSD Co. Ltd., and Seoul Science Fellowship.

References

1. Pennacchio LA, Olivier M, Hubacek JA, et al. An apolipoprotein influencing triglycerides in humans and mice revealed by comparative sequencing. *Science*. 2001; 294:169–173. [PubMed: 11588264]
2. van der Vliet HN, Sammels MG, Leegwater AC, et al. Apolipoprotein A-V: a novel apolipoprotein associated with an early phase of liver regeneration. *J Biol Chem*. 2001; 276:44512–44520. [PubMed: 11577099]
3. van der Vliet HN, Schaap FG, Levels JH, et al. Adenoviral overexpression of apolipoprotein A-V reduces serum concentrations of triglycerides and cholesterol in mice. *Biochem Biophys Res Commun*. 2002; 295:1156–1159. [PubMed: 12135615]
4. Pennacchio LA, Olivier M, Hubacek JA, Krauss RM, Rubin EM, Cohen JC. Two independent apolipoprotein A5 haplotypes influence human plasma triglyceride concentrations. *Hum Mol Genet*. 2002; 11:3031–3038. [PubMed: 12417524]
5. Aouizerat BE, Kulkarni M, Heilbron D, et al. Genetic analysis of a polymorphism in the human apoA-V gene: effect on plasma lipids. *J Lipid Res*. 2003; 44:1167–1173. [PubMed: 12671030]
6. Endo K, Yanagi H, Araki J, Hirano C, Yamakawa-Kobayashi K, Tomura S. Association found between the promoter region polymorphism in the apolipoprotein A-V gene and the serum triglyceride concentration in Japanese schoolchildren. *Hum Genet*. 2002; 111:570–572. [PubMed: 12436249]
7. Baum L, Tomlinson B, Thomas GN. APOA5 –1131T>C polymorphism is associated with triglyceride concentrations in Chinese men. *Clin Genet*. 2003; 63:377–379. [PubMed: 12752569]
8. Cullen P. Evidence that triglycerides are an independent coronary heart disease risk factor. *Am J Cardiol*. 2001; 86:943–949. [PubMed: 11053704]
9. Talmud PJ, Hawe E, Miller GJ, Humphries SE. Nonfasting apolipoprotein B and triglyceride concentrations as a useful predictor of coronary heart disease risk in middle-aged UK men. *Arterioscler Thromb Vasc Biol*. 2002; 22:1918–1923. [PubMed: 12426225]
10. Szalai C, Keszei M, Duba J, et al. Polymorphism in the promoter region of the apolipoprotein A5 gene is associated with an increased susceptibility for coronary artery disease. *Atherosclerosis*. 2004; 173:109–114. [PubMed: 15177130]
11. Bi N, Yan SK, Li GP, Yin ZN, Chen BS. A single nucleotide polymorphism –1131T>C in the apolipoprotein A5 gene is associated with an increased risk of coronary artery disease and alters triglyceride metabolism in Chinese. *Mol Genet Metab*. 2004; 83:280–286. [PubMed: 15542401]
12. Lai CQ, Demissie S, Cupples LA, et al. Influence of the APOA5 locus on plasma triglyceride, lipoprotein subclasses, and CVD risk in the Framingham Heart Study. *J Lipid Res*. 2004; 45:2096–2105. [PubMed: 15342688]
13. Hsu LA, Ko YL, Chang CJ, et al. Genetic variations of apolipoprotein A5 gene is associated with the risk of coronary artery disease among Chinese in Taiwan. *Atherosclerosis*. 2006; 185:143–149. [PubMed: 16054149]
14. Lee KW, Ayyobi AF, Frohlich JJ, Hill JS. APOA5 gene polymorphism modulates concentrations of triglyceride, HDL cholesterol and FERHDL but is not a risk factor for coronary artery disease. *Atherosclerosis*. 2004; 176:165–172. [PubMed: 15306190]

15. Talmud PJ, Cooper JA, Hattori H, Miller IP, Miller GJ, Humphries SE. The apolipoprotein A-V genotype and plasma apolipoprotein A-V and triglyceride concentrations: prospective risk of type 2 diabetes. Results from the Northwick Park Heart Study II. *Diabetologia*. 2006; 49:2337–2340. [PubMed: 16917759]
16. Jang Y, Kim JY, Kim OY, et al. The –1131T>C polymorphism in the apolipoprotein A5 gene is associated with postprandial hypertriglycerolemia; increased small, dense LDL concentrations; and oxidative stress in nonobese Korean men. *Am J Clin Nutr*. 2004; 80:832–840. [PubMed: 15447887]
17. Schaap FG, Rensen PC, Voshol PJ, et al. ApoAV reduces plasma triglycerides by inhibiting very low density lipoprotein–triglyceride (VLDL–TG) production and stimulating lipoprotein lipase-mediated VLDL–TG hydrolysis. *J Biol Chem*. 2004; 279:27941–27947. [PubMed: 15090553]
18. Grosskopf I, Baroukh N, Lee SJ, et al. Apolipoprotein A-V deficiency results in marked hypertriglyceridemia attributable to decreased lipolysis of triglyceride-rich lipoproteins and removal of their remnants. *Arterioscler Thromb Vasc Biol*. 2005; 25:2573–2579. [PubMed: 16166565]
19. Hokanson JE, Austin MA. Plasma triglyceride concentration is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol concentration: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk*. 1996; 3:213–219. [PubMed: 8836866]
20. Jeppesen J, Hein HO, Suadicani P, Gyntelberg F. Triglyceride concentration and ischemic heart disease: an eight-year follow-up in the Copenhagen Male Study. *Circulation*. 1988; 97:1029–1036. [PubMed: 9531248]
21. Wong WM, Hawe E, Li LK, et al. Apolipoprotein AIV gene variant S347 is associated with increased risk of coronary heart disease and lower plasma apolipoprotein AIV concentrations. *Circ Res*. 2003; 92:969–975. [PubMed: 12676816]
22. Talmud PJ, Martin S, Taskinen MR, et al. APOA5 gene variants, lipoprotein particle distribution, and progression of coronary heart disease: results from the LOCAT study. *J Lipid Res*. 2004; 45:750–756. [PubMed: 14729863]
23. Horínek A, Vráblík M, Ceska R, Adámková V, Poledne R, Hubacek JA. T–1131–>C polymorphism within the apolipoprotein AV gene in hypertriglyceridemic individuals. *Atherosclerosis*. 2003; 167:369–370. [PubMed: 12818421]
24. Korean Ministry For Health, Welfare and Family Affairs. National Health and Nutrition Survey. 1998 2005.
25. Korean National Statistical Office. Annual Report on the Cause of Death Statistics (based on vital registration). 1998 2005.
26. Kao JT, Wen HC, Chien KL, Hsu HC, Lin SW. A novel genetic variant in the apolipoprotein A5 gene is associated with hypertriglyceridemia. *Hum Mol Genet*. 2003; 12:2533–2539. [PubMed: 12915450]
27. Pennacchio LA, Rubin EM. Apolipoprotein A5, a newly identified gene that affects plasma triglyceride concentrations in humans and mice. *Arterioscler Thromb Vasc Biol*. 2002; 23:529–534. [PubMed: 12615678]
28. Rosenson RS, Otvos JD, Freedman DS. Relations of lipoprotein subclass concentrations and low-density lipoprotein size to progression of coronary artery disease in the Pravastatin Limitation of Atherosclerosis in the Coronary Arteries (PLAC-I) trial. *Am J Cardiol*. 2002; 90:89–94. [PubMed: 12106834]
29. Vakkilainen J, Steiner G, Ansquer JC, et al. Relationships between low-density lipoprotein particle size, plasma lipoproteins, and progression of coronary artery disease: the Diabetes Atherosclerosis Intervention Study (DAIS). *Circulation*. 2003; 107:1733–1737. [PubMed: 12665498]
30. Nabika T, Nasreen S, Kobayashi S, Masuda J. The genetic effect of the apoprotein AV gene on the serum triglyceride concentration in Japanese. *Atherosclerosis*. 2002; 165:201–204. [PubMed: 12417270]
31. Evans D, Buchwald A, Beil FU. The single nucleotide polymorphism –1131T>C in the apolipoprotein A5 (APOA5) gene is associated with increased triglycerides in patients with hyperlipidemia. *J Mol Med*. 2003; 81:645–654. [PubMed: 12937897]

Fig. 1.

Association of the *APOA5* -1131T>C polymorphism with concentrations of APOA5, TG and LDL particle size in normal controls and CAD patients. Data are expressed as the mean $\pm$ S.E. $^{\dagger}P < 0.05$, $^{\ddagger}P < 0.01$, $^{\S}P < 0.001$ compared with wild-type and $^*P < 0.05$, $^{\#}P < 0.01$ compared with heterozygote in each group tested by one-way analysis of variance (ANOVA) with Bonferroni method.

Table 1

Anthropometric and biochemical parameters of controls and CAD patients

	Controls (<i>n</i> = 741)	CAD patients (<i>n</i> = 741)
Age (year)	55.2 ± 0.32	55.2 ± 0.32
Male/female, <i>n</i> (%)	665 (89.7)/76 (10.3)	665 (89.7)/76 (10.3)
Body Mass Index (kg/m ²)	24.8 ± 0.10	25.1 ± 0.10 [†]
Cigarette smoker, <i>n</i> (%)	194 (26.2)	165 (22.3)
Alcohol drinker, <i>n</i> (%)	517 (69.8)	430 (58.0) ^{†††}
Antidyslipidemic therapy, <i>n</i> (%)	–	492 (66.4) ^{†††}
Antihypertensive therapy, <i>n</i> (%)	46 (6.2)	594 (80.3) ^{†††}
Antiplatelet therapy, <i>n</i> (%)	15 (2.0)	599 (80.9) ^{†††}
Blood pressure (BP)		
Systolic BP (mmHg)	127.6 ± 0.63	125.2 ± 0.64 ^{††}
Diastolic BP (mmHg)	80.3 ± 0.41	78.0 ± 0.36 ^{†††}
Cholesterol (mg/dl)		
Total-C	196.6 ± 1.30	171.8 ± 1.50 ^{††††***}
HDL-C	49.9 ± 0.46	44.2 ± 0.41 ^{††††***}
LDL-C	118.7 ± 1.21	97.6 ± 1.40 ^{††††***}
Triglyceride (mg/dl) ^f	141.6 ± 2.97	151.2 ± 3.34 ^{†††}
Apolipoprotein A5 (ng/ml) ^f	230.8 ± 4.24	197.3 ± 5.18 ^{††††***}
Apolipoprotein A1 (mg/dl)	143.0 ± 1.02	130.8 ± 0.84 ^{††††***}
Apolipoprotein B (mg/dl)	87.7 ± 0.93	77.0 ± 0.84 ^{††††***}
LDL particle size (nm)	25.6 ± 0.05	25.1 ± 0.05 ^{††††***}
Glucose (mg/dl) ^f	90.9 ± 0.49	89.4 ± 0.44 ^{†*}
Insulin (μU/ml) ^f	8.21 ± 0.16	10.1 ± 0.18 ^{††††***}
HOMA-IR ^{I f}	1.86 ± 0.04	2.25 ± 0.04 ^{††††***}

Mean ± S.E. or percentage.

^flog-transformed.^IHOMA-IR = {fasting insulin (μU/ml) × fasting glucose (mg/dl)}/22.5.[†]*P* < 0.05,^{††}*P* < 0.01,^{†††}*P* < 0.001 by independent *t*-test or χ^2 -test and^{*}*P* < 0.05,^{**}*P* < .01, and^{***}*P* < 0.001 by general linear with adjustment for BMI, alcohol consumption, systolic blood pressure, and diastolic blood pressure when compared with controls.

Table 2
Genotype distribution of the *APOA5* –1131T>C SNP in control and CAD groups

<i>APOA5</i> –1131T>C	Controls		CAD patients		P-value
	(n = 741)		(n = 741)		
	n	%	n	%	
T/T	382	51.6	320	43.2	
T/C	295	39.8	343	46.3	0.005
C/C	64	8.6	78	10.5	
T/T + T/C	677	91.4	663	89.5	NS
T/C + C/C	359	48.4	421	56.8	0.001
C allele frequency	423	28.5	499	33.7	0.003

Table 3 Unadjusted and adjusted OR for CAD patients to the *APOA5* –1131T>C genotype

<i>APOA5</i> –1131T>C	Unadjusted OR (95% CI) [†]	<i>P</i>	Adjusted* OR (95%CI) [†]	<i>P</i>
T [‡] vs. C	1.27 (1.09–1.49)	0.003	1.23 (1.05–1.44)	0.012
T/T+T/C [‡] vs. C/C	1.24 (0.88–1.76)	0.217	1.20 (0.84–1.71)	NS
T/T [‡] vs. T/C+C/C	1.40 (1.14–1.72)	0.001	1.34 (1.09–1.65)	0.007

[†] Confidence interval.

* Adjusted for BMI, alcohol consumption, systolic blood pressure and diastolic blood pressure.

[‡] Reference.

Association of the *APOA5* –1131T>C genotype with lipid variables in healthy controls and CAD patients

Table 4

<i>APOA5</i> –1131T>C	Controls (<i>n</i> = 741)			CAD cases (<i>n</i> = 741)			<i>P</i> value
	TT (<i>n</i> = 382)	TC (<i>n</i> = 295)	CC (<i>n</i> = 64)	TT (<i>n</i> = 320)	TC (<i>n</i> = 343)	CC (<i>n</i> = 78)	
Cholesterol (mg/dl)							
Total-C	196 ± 1.87	198 ± 2.01	198 ± 4.20	172 ± 2.29	172 ± 2.18	172 ± 4.81	NS
HDL-C	50.5 ± 0.63	49.7 ± 0.74	46.9 ± 1.41	45.2 ± 0.61	43.5 ± 0.61	43.1 ± 1.25	NS
LDL-C	120 ± 1.72	117 ± 1.93	118 ± 3.57	99.3 ± 2.03	97.3 ± 2.11	91.9 ± 4.65	NS
ApoA1 (mg/dl)	143 ± 1.46	144 ± 1.57	138 ± 3.49	132 ± 1.34	130 ± 1.19	130 ± 2.50	NS
ApoB (mg/dl)	86.8 ± 1.33	88.7 ± 1.48	88.8 ± 2.57	76.5 ± 1.23	77.5 ± 1.26	76.2 ± 2.74	NS
Glucose (mg/dl) ^f	90.4 ± 0.66	90.9 ± 0.78	94.2 ± 1.73	88.8 ± 0.61	89.5 ± 0.66	91.3 ± 1.61	NS
Insulin (μU/ml) ^f	8.02 ± 0.23	8.45 ± 0.23	8.19 ± 0.53	10.1 ± 0.26	10.1 ± 0.27	9.86 ± 0.56	NS
HOMA-IR ^g	1.81 ± 0.06	1.92 ± 0.06	1.93 ± 0.15	2.24 ± 0.06	2.26 ± 0.07	2.22 ± 0.14	NS

Mean ± S.E.,

^f tested by log-transform.

Tested by one-way analysis of variance (ANOVA).

^g HOMA-IR = {fasting insulin (μU/ml) × fasting glucose (mg/dl)}/22.5.