A systematic review and meta-analysis of the relationship between lipoprotein lipase Asn291Ser variant and diseases

Yaomin Hu, Wei Liu,[1] Rong Huang, and Xiaoying Zhang

Division of Endocrinology, Department of Internal Medicine, Renji Hospital, Shanghai Jiaotong University, Shanghai 200127, China

Abstract **This systematic review attempted to summarize the associations between the Asn291Ser variant in the lipoprotein lipase (LPL) gene and dyslipidemia, the risk of type 2 diabetes mellitus (T2DM), and coronary heart disease (CHD). In addition, the relationships between the Asn291Ser variant and other metabolic diseases such as obesity and high blood pressure were also investigated in this systematic review. We systematically reviewed the literature by means of a meta-analysis. Twenty-one articles, including 19,246 white subjects, were selected for this meta-analysis. The summary standardized mean difference (SMD) of plasma triglyceride (TG) for carriers compared with noncarriers of the Asn291Ser variant was 3.23 ($P < 0.00001$). The summary SMD of plasma HDL-cholsterol (HDL-C) for carriers compared with noncarriers of the Asn291Ser variant was −3.42 ($P < 0.0001$). The summary SMD of the association of the Asn291Ser variant with plasma TG increased with increasing age and weight gain. Significant interactions between the LPL Asn291Ser variant and fasting glucose, T2DM, and CHD were seen ($P = 0.02$, 0.04, and 0.01, respectively). No significant interactions were seen between the LPL Asn291Ser variant and body mass index, waist–hip ratio, and blood pressure ($P > 0.05$). This meta-analysis indicates that the Asn291Ser variant in the LPL gene is a risk factor for dyslipidemia, characterized by hypertriglyceridemia and low HDL-C levels. And the Asn291Ser variant in the LPL gene predisposes to more severe dyslipidemia with increasing age and weight gain. Also, this meta-analysis shows that the LPL Asn291Ser variant is associated with CHD and T2DM.—** Hu, Y., W. Liu, R. Huang, and X. Zhang. **A systematic review and meta-analysis of the relationship between lipoprotein lipase Asn291Ser variant and diseases. *J. Lipid Res.* 2006.** 47: **1908–1914.**

Supplementary key words dyslipidemia • type 2 diabetes mellitus • coronary heart disease • obesity • high blood pressure

Lipoprotein lipase (LPL, E.C.3.1.1.34) is a key enzyme that hydrolyzes triglyceride (TG) contained in the core of both chylomicrons and VLDL particles (1–4). Because of this central role, the function of LPL could be associated with dyslipidemia and characterized by hypertriglyceridemia and low HDL-cholesterol (HDL-C) levels. The Asn291Ser variant in the LPL gene was first identified in 1994 (5). Since then, many studies have examined the association between the Asn291Ser variant in the LPL gene and dyslipidemia (6–26). It has been observed that the Asn291Ser variant in the LPL gene occurs more frequently in dyslipidemia subjects than in control subjects. Based on 13 studies published up to 1997 (27), Asn291Ser heterozygous carriers had an average increase in plasma TG of 31% and an average decrease in HDL-C of 0.12 mM ($P < 0.001$). However, since then, a number of other studies have been published in which no association was observed (10, 18, 20, 21, 24). In addition to study power and other methodological issues, interactions with important dyslipidemic risk factors such as increasing age and weight gain could have resulted in heterogeneous findings. Indeed, the dyslipidemic phenotype is more severe in older subjects or overweight/obese subjects. It has been suggested that old age and obesity positively interact with the Asn291Ser variant, thereby increasing dyslipidemic risk.

Also, given the large number of studies implicating the LPL Asn291Ser variant in type 2 diabetes mellitus (T2DM) and atherosclerosis, interactions between the LPL Asn291Ser variant and the risk of T2DM and coronary heart disease (CHD) were investigated in this meta-analysis.

The aim of our study was, therefore, to review the literature systematically by means of a meta-analysis, and to provide a quantitative summary estimate on the associations between the Asn291Ser variant in the LPL gene and dyslipidemia, the risk of T2DM, and CHD. In addition, the relationships between the Asn291Ser variant and other metabolic diseases such as obesity and high blood pressure (HBP) were also investigated in this meta-analysis.

Manuscript received 6 March 2006 and in revised form 25 April 2006.

Published, JLR Papers in Press, June 1, 2006.
DOI 10.1194/jlr.M600108-JLR200

Abbreviations: BMI, body mass index; CHD, coronary heart disease; HBP, high blood pressure; HDL-C, HDL-cholesterol; LPL, lipoprotein lipase; PL, pancreatic lipase; SMD, standardized mean difference; T2DM, type 2 diabetes mellitus; TG, triglyceride; WHR, waist–hip ratio.

[1] To whom correspondence should be addressed.
e-mail: Amin99@163.com

SUBJECTS AND METHODS

Search strategy

Studies were identified through a computerized Medline, Cochrane Collaboration Library database, and Science Citation Index search on studies published until January 2006 using the free test words *lipoprotein lipase*, *Asn291Ser variant*, *dyslipidemia*, *type 2 diabetes mellitus*, *coronary heart disease*, *obesity*, and *high blood pressure*. References in relevant publications were also examined. There were no language restrictions. The selection criteria of publications for inclusion in this meta-analysis were as follows: *1*) the Asn291Ser variant in LPL was studied; *2*) family, cross-sectional, case-control, or case-referent studies were included that identified both Asn291Ser variant carriers and noncarriers; *3*) data were reported on the relationship between at least one of the variables TG, HDL-C, LDL-C, body mass index (BMI), waist–hip ratio (WHR), systolic blood pressure, diastolic blood pressure, fasting plasma glucose, the risk of T2DM or CHD and the Asn291Ser variant in LPL.

Data collection

For each study, information was collected concerning the characteristics of the studies. These study characteristics were used to evaluate sources of variation in effect estimates. All articles were independently scored by two reviewers. Disagreements were solved in consensus meetings.

Study characteristics

Twenty-one family, cross-sectional, case-control, or case-referent articles were included for this meta-analysis (6–26). All studies included white subjects. We excluded one study because it examined the expression of the mutation instead of the prevalence (28). Nine studies were excluded because they did not provide clear data (29–37). Of the 21 included publications, it was impossible to separate a small number of homozygous from heterozygous individuals, and therefore these studies were just divided into two groups, carriers and noncarriers of the Asn291Ser variant.

Structural modeling for mutant LPL

LPL model structures were constructed using the molecular modeling system INSIGHT II program (Accelyers, Inc.; Burlington, MA) on a Silicon Graphics workstation and were based on the crystal structures of the human and porcine pancreatic lipases (PLs). The crystal structures of human and porcine PL (Protein Data Bank accession numbers 1lpa and 1eth) were obtained from the protein structure data base http://www.rcsb.org/pdb/).

Statistical analysis

Meta-analyses were performed on RevMan4.2.8 (the Cochrane Collaboration; http://www.cochrane.co.uk). To check for publication bias, a funnel plot was constructed. The results were reported using odds ratios and corresponding 95% confidence intervals (95% CIs) for dichotomous data and weighted mean difference and corresponding 95% CIs for continuous outcomes. If different measurement units were applied for the same continuous variable, standardized mean difference (SMD) and corresponding 95% CIs were used for this continuous outcome. Heterogeneity between trials was tested using a standard Chi squared test. If there was no heterogeneity, a fixed-effect model was used. If heterogeneity was found, sensitivity analysis was conducted. If the reasons that led to the heterogeneity could not be found by sensitivity analysis, the random-effect model was used.

RESULTS

Of the 21 included publications, nine provided separate associations in different populations (7, 8, 11, 14, 16, 18, 21, 24, 26). These associations were considered as separate studies. All of the studies, which included 19,246 white subjects, were selected for this meta-analysis. Funnel plots showed no asymmetry, either visually or in terms of statistical significance ($P > 0.05$) (**Fig. 1**), indicating no publication bias.

The SMD of plasma TG for carriers compared with noncarriers of the Asn291Ser variant ranged from −4.38 to 17.17 (**Fig. 2**). The summary SMD was 3.23 (95% CI, 2.49–3.97; $P < 0.00001$). Asn291Ser variant carriers had an average increase in plasma TG of 32.3%. The results indicate that the Asn291Ser variant is a risk factor for hypertriglyceridemia.

The SMD of plasma HDL-C for carriers compared with noncarriers of the Asn291Ser variant ranged from −12.0 to 0.73 (**Fig. 3**). The summary SMD was −3.42 (95% CI −5.07–−1.77; $P < 0.0001$). Asn291Ser variant carriers had an average decrease in plasma HDL-C of 34.2%. The results indicate that the Asn291Ser variant is a risk factor for low HDL-C.

Also, our results from the meta-analysis showed a significant effect of the Asn291Ser variant on plasma LDL-C concentrations (data not shown). The SMD of plasma LDL-C for carriers compared with noncarriers of the Asn291Ser variant ranged from −0.10 to 2.10. The summary SMD was 0.53 (95% CI, 0.02–1.05; $P = 0.04$).

We further examined whether the association of the Asn291Ser variant with plasma TGs depended on the age of the subject. Subjects aged 35 to 65 years old were included, and they were divided into three groups: 35–45 years old (four studies), 45–55 years old (five studies), and 55–65 years old (four studies). The summary SMD increased with the age of the subjects (**Fig. 4**). The SMD rose from 0.92 (95% CI, 0.20–1.64) for subjects aged 35 to 45 years to 3.25 (95% CI, 1.83–4.67) for subjects that were 55 to 65 years old. Also, similar effects of the Asn291Ser variant on HDL-C stratified by age were observed. The SMD was reduced from

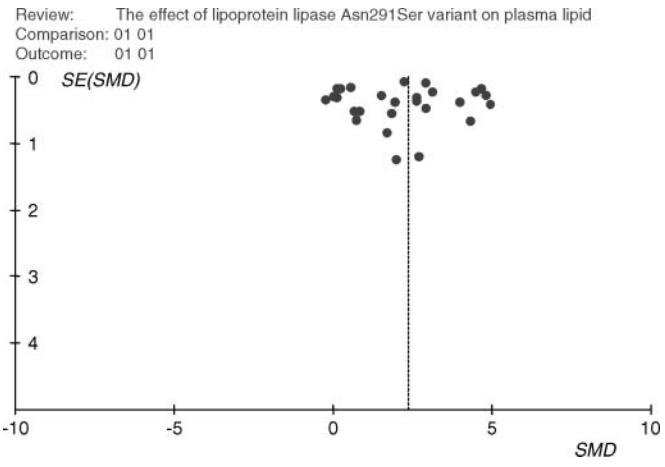

Fig. 1. Funnel plot for subjects with the Asn291Ser variant versus subjects without this variant, unadjusted. Dashed line indicates total summary odds. SE, standard error; SMD, standardized mean difference.

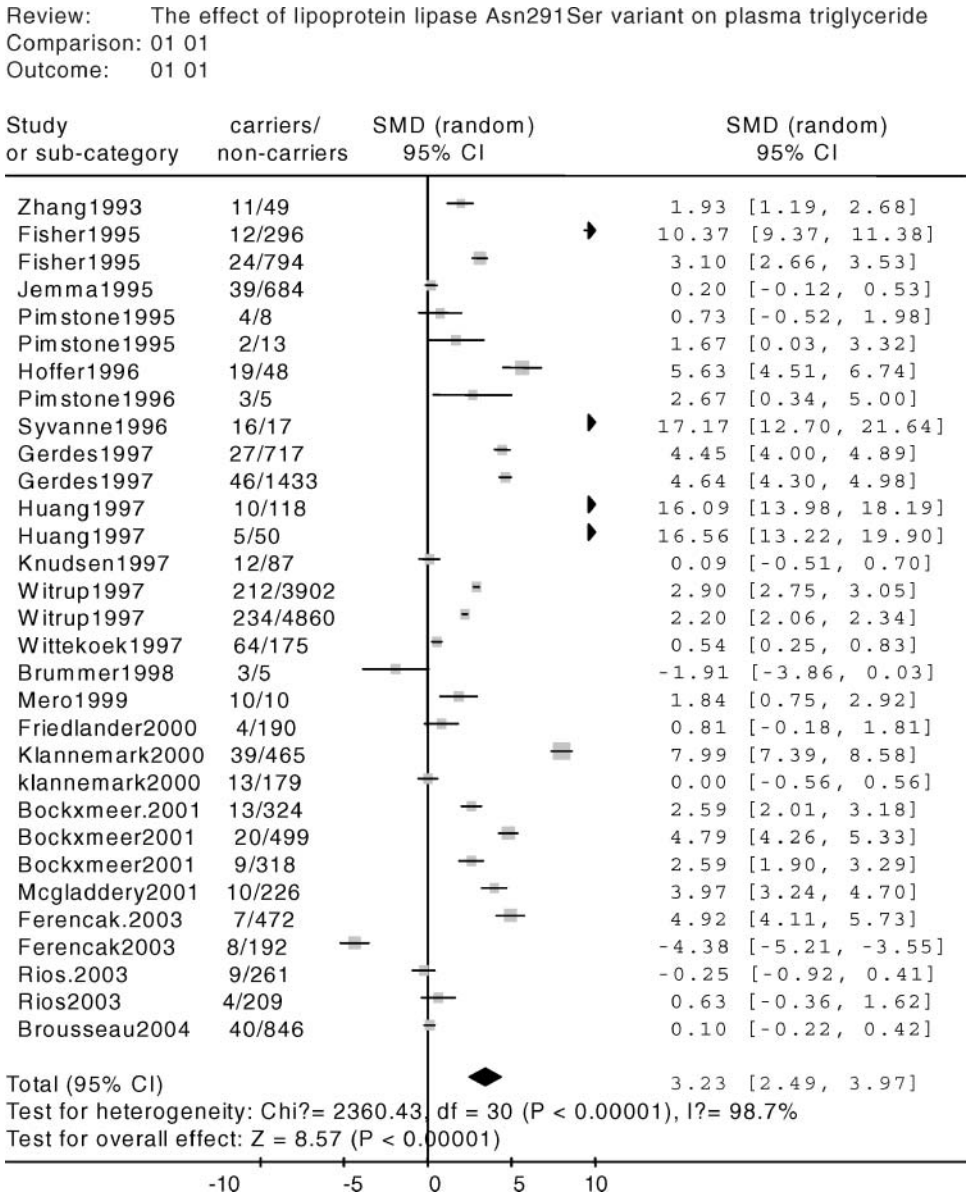

Fig. 2. Meta-analyses of all studies of the effect of the Asn291Ser variant on plasma triglyceride (TG). Size of squares indicating mean values is proportional to the weight that each study contributed to the aggregated result. Aggregated means with 95% confidence intervals are shown by diamond symbols. CI, confidence interval.

−1.12 (95% CI, −1.94–−0.31) for subjects aged from 35 to 45 years to −5.29 (95% CI, −6.80–−3.77) for subjects that were from 55 to 65 years old (data not shown).

Weight gain, another environmental factor, may influence lipid metabolism. We examined whether the association of the Asn291Ser variant with lipid metabolism depended on weight gain. Subjects were divided into three groups: normal weight (BMI <25 kg/m^2), overweight (BMI 25–30 kg/m^2), and obese (BMI >30 kg/m^2). Our meta-analyses also suggest that the Asn291Ser variant in the LPL gene predisposes to more severe dyslipidemia with weight gain (data not shown).

Given the large number of studies implicating the LPL Asn291Ser variant in subjects with obesity, HBP, T2DM, and atherosclerosis, interactions between the LPL Asn291Ser variant and the above metabolic disease parameters were investigated by meta-analysis (**Table 1**). Significant interactions between the LPL Asn291Ser variant and fasting glucose, T2DM, and CHD were seen (P = 0.02, 0.04, 0.01, respectively). No significant interactions were seen between the LPL Asn291Ser variant and BMI, WHR, or blood pressure ($P >$ 0.05). The results indicate that the Asn291Ser variant is a risk factor for T2DM and CHD but not for obesity and HBP.

DISCUSSION

One of the aims of the present meta-analysis was to investigate the relationship between the Asn291Ser variant in the LPL gene and dyslipidemia, and the results show

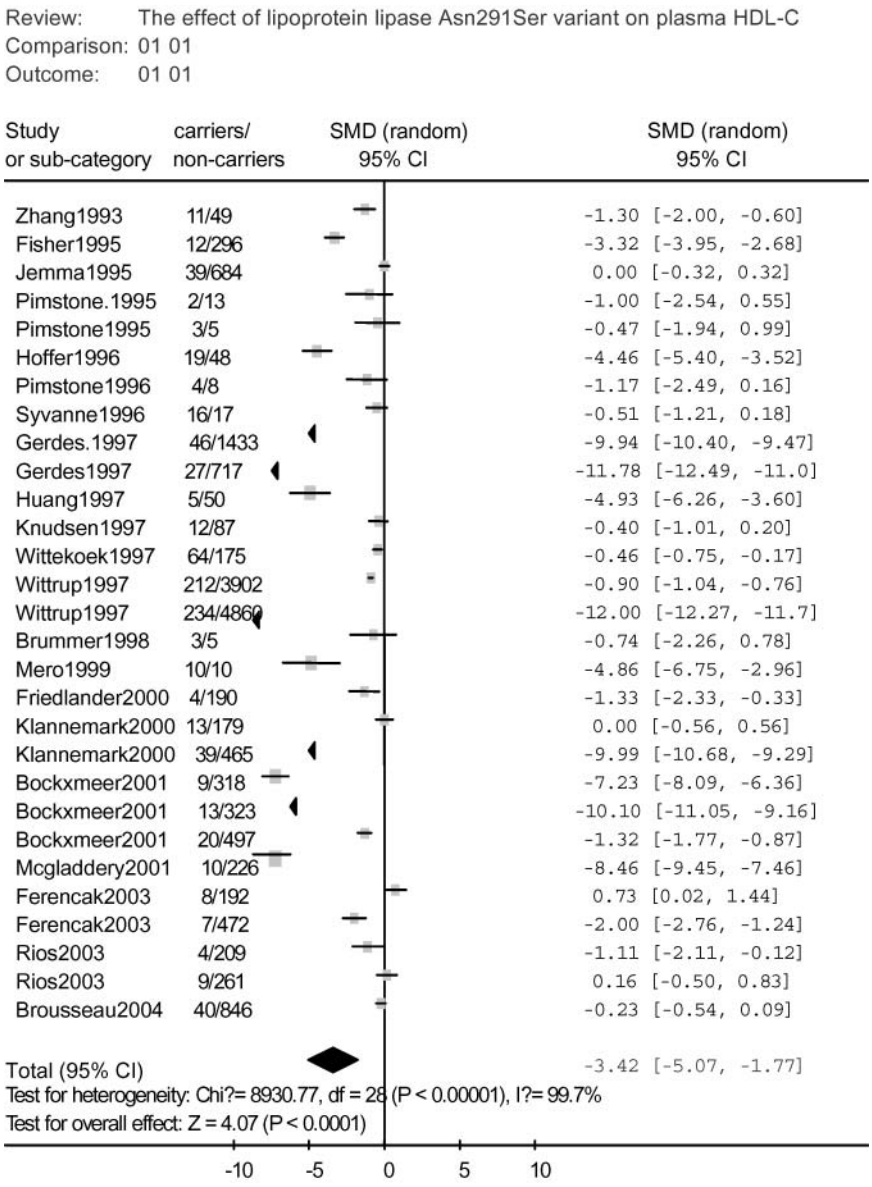

Fig. 3. Meta-analyses of all studies of the effect of the Asn291Ser variant on plasma HDL-cholesterol. Results are depicted as in Fig. 2.

that the Asn291Ser variant in the LPL gene is a risk factor for dyslipidemia and is characterized by hypertriglyceridemia, low HDL-C levels, and low LDL-C concentrations.

How could the Asn291Ser variant in the LPL gene influence the susceptibility to dyslipidemia?

LPL plays a central role in TG metabolism. The main activity of LPL is to hydrolyze TGs contained in the core of both chylomicrons and VLDL particles (1–4). LPL is a 55 kDa glycoprotein that is primarily synthesized in adipocytes, muscle cells, and macrophages. The enzyme is active as a noncovalent dimer and the dissociated monomer enzyme has no activity (38). Activated LPL is bound to the surface of endothelial cells and can be released into the blood by heparin. Genetic defects of LPL are responsible for the reduced TG-rich lipoprotein clearance, and mutations in the LPL gene have been shown to play a central role in the development of dyslipidemia in the general population (32, 38–40). The human gene encoding for LPL is located on chromosome 8p22 and consists of 10 exons that span approximately 35 kb (2).

To explore the molecular mechanism underlying the correlation between the LPL Asn291Ser variant and its activity, we constructed a three-dimensional (3-D) model of human LPL based on the known crystal structure of human PL (1LPA and 1LPB, http://www.rcsb.org/pdb). The 3-D model of LPL shows that Asn291 is located at the back of the molecule, in the region of heparin binding and homodimer formation and close to the activation center of the LPL protein. This important location of Asn291 (**Fig. 5**) could easily suggest its critical role in LPL activity. The Asn291Ser mutation may alter the heparin binding and consequently favor monomerization, as opposed to dimerization. This conformation change in Asn291 has been suggested to

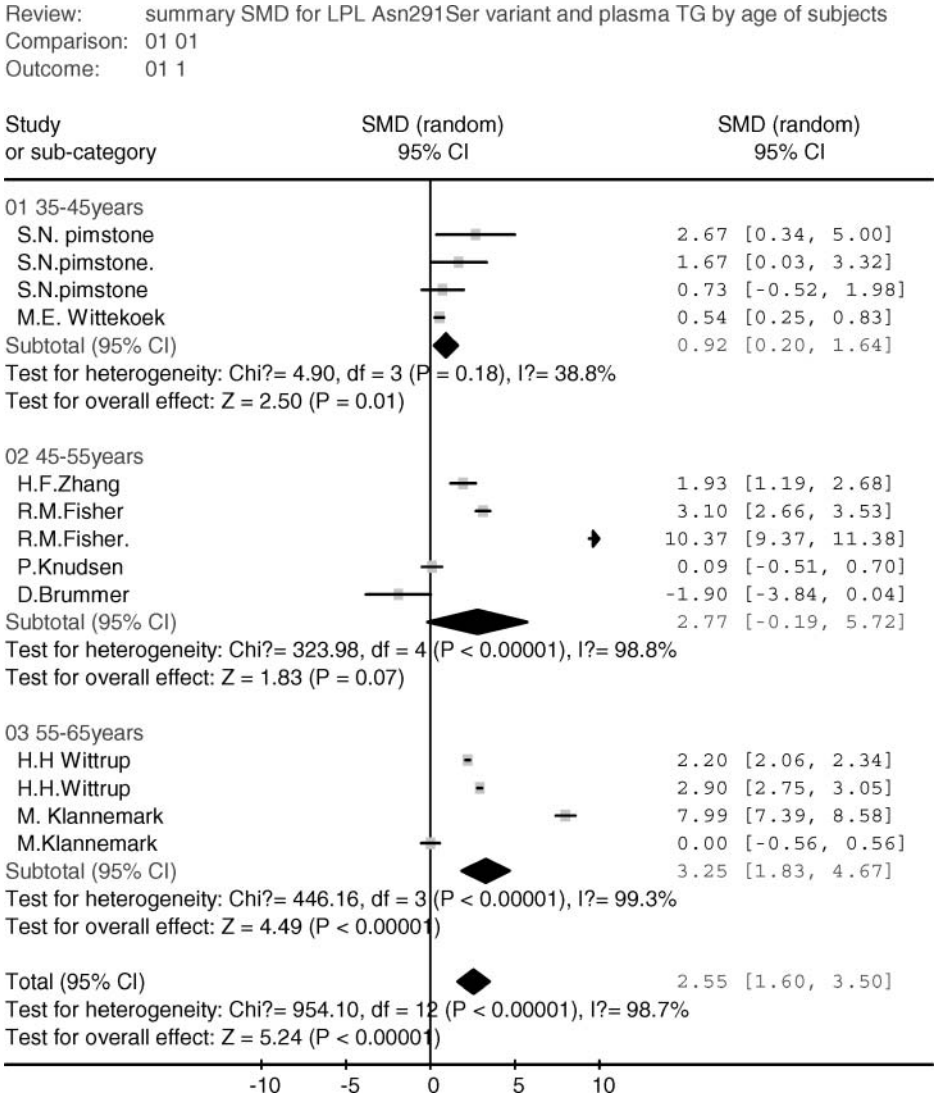

Fig. 4. Meta-analyses of all studies of the effect of the Asn291Ser variant on plasma TG by age of subjects. Results are depicted as in Fig. 2.

change the hydrogen bond pattern between Asn291 and its structural neighbors and to lead to a rapid dissociation from the active dimer to the inactive monomer (41).

In previous studies (23, 28), in vivo and in vitro experiments showed that mutant Asn291Ser LPL exhibited decreased catalytic activity. Also, in our unpublished observations, in vitro experiments showed that the mutant Asn291Ser LPL exhibited both decreased catalytic activity (about 60% of wild-type activity) and secretion ability (about half that of the wild type), supporting the idea that the Asn291Ser mutation is a pathogenic cause for decreased LPL activity and dyslipidemia.

TABLE 1. Meta-analyses of all studies of effect of the Asn291Ser variant on BMI, WHR, SBP, DBP, FPG, T2DM, and CHD

Outcome	N (Carriers/Noncarriers)	Test for Heterogeneity		Model	Overall Effect		z	P
		χ^2	P		WMD/OR	95% CI		
BMI	211/3,022	89.94	0.00001	Random	0.08	−0.42–0.58	0.32	0.75
WHR	95/1,378	242.7	0.00001	Random	0.02	0.00–0.05	1.67	0.09
SBP	132/836	20.03	0.0002	Random	7.18	−1.00–15.4	1.72	0.09
DBP	132/836	11.16	0.01	Random	3.26	−0.15–6.68	1.87	0.06
FPG	74/185	6.10	0.01	Random	0.42	0.06–0.79	2.29	0.02
T2DM	31/164	6.68	0.04	Random	2.26	1.02–4.99	2.02	0.04
CHD	1,203/6,192	13.84	0.003	Random	1.48	1.09–2.00	2.54	0.01

BMI, body mass index; WHR, waist–hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure; FPG, fasting plasma glucose; T2DM, type 2 diabetes mellitus; CHD, coronary heart disease; OR, odds ratio; SMD, standardized mean difference.

Yaomin Hu, Yan Ren, Robert Z. Luo, Xiang Mao, Xiaomin Chen, Xiujun Li, Xu Cao, Lu Guan, Xiang Chen, Jianwei Li, Yang Long, Xiangxun Zhang, Haoming Tian, unpublished data.

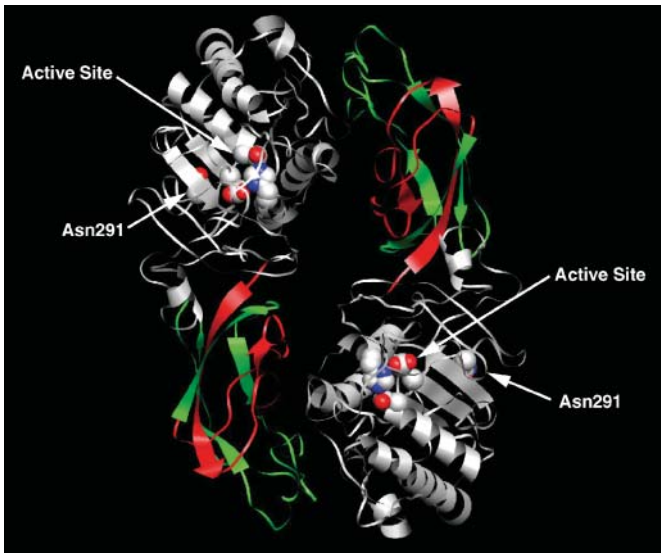

Fig. 5. The three-dimensional model structure of LPL proteins in a noncovalent homodimer with a head-to-tail configuration. The monomer LPL exhibited two domains: a large N-terminal domain (1–315 amino acid residues), and a small C-terminal domain (316–448 amino acid residues) that is essential for the formation and/or stability of the LPL homodimer. Arrows indicate the active center and the Asn291Ser mutation site.

Our meta-analysis also indicates that the Asn291Ser variant in the LPL gene predisposes to more severe dyslipidemia with increasing age and weight gain. It is thus conceivable that the functional role of LPL is of particular importance when the lipolytic system becomes challenged by environmental factors such as increasing age and weight gain.

In addition, this meta-analysis has indicated that the Asn291Ser variant is a risk factor for T2DM and CHD but not for obesity and HBP. Our study has demonstrated that LPL Asn291Ser variant carriers have a less favorable lipid profile. Thus, the observation that the LPL Asn291Ser variant, which is linked to a deleterious lipid profile, is associated with CHD is not surprising. Could the deleterious lipid profile result from the LPL Asn291Ser variant link to T2DM? Studies have shown that the primary increase in plasma TG and nonesterified fatty acid (NEFA) levels could lead to insulin resistance and impaired insulin secretion, which usually are required to manifest T2DM by multiple converging mechanisms. Through inhibition of both glucose oxidation and nonoxidative glucose metabolism (mostly glycogen synthesis), increased NEFA uptake is thought to be responsible, at least in part, for the skeletal muscle insulin resistance of type 2 diabetic patients (42). In the liver, increased NEFA oxidation may inhibit glucose oxidation and stimulate gluconeogenesis, thereby contributing to the inappropriate glucose production found in type 2 diabetic patients (43). There is evidence that chronic exposure of the β-cell to elevated FFA levels can cause damage to its function (44). Typically, this occurrence would result in the damage to β-cells characteristically seen in T2DM. The potential mechanisms underlying islet lipotoxicity might be involved in the fatty acid–mediated upregulation of inducible nitric oxide synthase and/or ceramide synthesis, either or both of which might lead to β-cell apoptosis (44).

In conclusion, this meta-analysis, including all available evidence to date, indicates that the Asn291Ser variant in the LPL gene is a risk factor for dyslipidemia, characterized by hypertriglyceridemia and low HDL-C levels. And the functional role of LPL is of particular importance when the lipolytic system becomes challenged by environmental factors such as increasing age and weight gain. Also, this meta-analysis shows that the LPL Asn291Ser variant, which is linked to a deleterious lipid profile, is associated with CHD and T2DM.

This study was supported by the National Scientific Foundation of China (No. 30570882)

REFERENCES

1. Goldberg, U., and M. Merkel. 2001. Lipoprotein lipase: physiology, biochemistry, and molecular biology. *Front. Biosci.* **6:** d388–d405.
2. Deeb, S., and R. Peng. 1989. Structure of the human lipoprotein lipase gene. *Biochemistry.* **28:** 4131–4135.
3. Hide, W. A., L. Chan, and W. H. Li. 1992. Structure and evolution of the lipase superfamily. *J. Lipid Res.* **33:** 167–178.
4. Olivecrona, T., O. Hernell, and T. Egelrud. 1975. Lipoprotein lipase. *Adv. Exp. Med. Biol.* **52:** 269–279.
5. Ma, Y., T. C. Ooi, M. S. Liu, H. Zhang, R. McPherson, A. L. Edwards, I. J. Forsythe, J. Frohlich, J. D. Brunzell, and M. R. Hayden. 1994. High frequency of mutations in the human lipoprotein lipase gene in pregnancy-induced chylomicronemia: possible association with apolipoprotein E2 isoform. *J. Lipid Res.* **35:** 1066–1075.
6. Zhang, H., P. W. Reymer, M. S. Liu, I. J. Forsythe, B. E. Groenemeyer, J. Frohlich, J. D. Brunzell, J. J. Kastelein, M. R. Hayden, and Y. Ma. 1995. Patients with apoe3 deficiency (E2/2, E3/2, and E4/2) who manifest with hyperlipidemia have increased frequency of an Asn 291→Ser mutation in the human LPL gene. *Arterioscler. Thromb. Vasc. Biol.* **15:** 1695–1703.
7. Fisher, R. M., F. Mailly, R. E. Peacock, A. Hamsten, M. Seed, J. S. Yudkin, U. Beisiegel, G. Feussner, G. Miller, S. E. Humphries, et al. 1995. Interaction of the lipoprotein lipase asparagine 291→serine mutation with body mass index determines elevated plasma triacylglycerol concentrations: a study in hyperlipidemic subjects, myocardial infarction survivors, and healthy adults. *J. Lipid Res.* **36:** 2104–2112.
8. Klannemark, M., L. Suurinkeroinen, M. Orho-Melander, L. Groop, and M. R. Taskinen. 2000. Interaction between the Asn291Ser variant of the LPL gene and insulin resistance on dyslipidaemia in high risk individuals for Type 2 diabetes mellitus. *Diabet. Med.* **17:** 599–605.
9. Friedlander, Y., E. Leitersdorf, R. Vecsler, H. Funke, and J. Kark. 2000. The contribution of candidate genes to the response of plasma lipids and lipoproteins to dietary challenge. *Atherosclerosis.* **152:** 239–248.
10. Brummer, D., D. Evans, D. Berg, H. Greten, U. Beisiegel, and W. A. Mann. 1998. Expression of type III hyperlipoproteinemia in patients homozygous for apolipoprotein E-2 is modulated by lipoprotein lipase and postprandial hyperinsulinemia. *J. Mol. Med.* **76:** 355–364.
11. Wittrup, H. H., A. Tybjaerg-Hansen, S. Abildgaard, R. Steffensen, P. Schnohr, and B. G. Nordestgaard. 1997. A common substitution (Asn291Ser) in lipoprotein lipase is associated with increased risk of ischemic heart disease. *J. Clin. Invest.* **99:** 1606–1613.
12. Jemaa, R., F. Fumeron, O. Poirier, L. Lecerf, A. Evans, D. Arveiler, G. Luc, J. P. Cambou, J. M. Bard, J. C. Fruchart, et al. 1995. Lipoprotein lipase gene polymorphisms: associations with myocardial infarction and lipoprotein levels, the ECTIM study. *J. Lipid Res.* **36:** 2141–2146.
13. Hoffer, M. J., S. J. Bredie, D. I. Boomsma, P. W. Reymer, J. J. Kastelein, P. de Knijff, P. N. Demacker, A. F. Stalenhoef, L. M. Havekes, and R. R. Frants. 1996. The lipoprotein lipase (Asn291→Ser) mutation is associated with elevated lipid levels in families with familial combined hyperlipidaemia. *Atherosclerosis.* **119:** 159–167.

14. Gerdes, C., R. M. Fisher, V. Nicaud, J. Boer, S. E. Humphries, P. J. Talmud, and O. Faergeman. 1997. Lipoprotein lipase variants D9N and N291S are associated with increased plasma triglyceride and lower high-density lipoprotein cholesterol concentrations: studies in the fasting and postprandial states: the European Atherosclerosis Research Studies. *Circulation.* **96:** 733–740.
15. Pimstone, S. N., S. M. Clee, S. E. Gagne, L. Miao, H. Zhang, E. A. Stein, and M. R. Hayden. 1996. A frequently occurring mutation in the lipoprotein lipase gene (Asn291Ser) results in altered postprandial chylomicron triglyceride and retinyl palmitate response in normolipidemic carriers. *J. Lipid Res.* **37:** 1675–1684.
16. Huang, P., K. Kostulas, W. X. Huang, M. Crisby, V. Kostulas, and J. Hillert. 1997. Lipoprotein lipase gene polymorphisms in ischaemic stroke and carotid stenosis. *Eur. J. Clin. Invest.* **27:** 740–742.
17. Knudsen, P., S. Murtomaki, M. Antikainen, S. Ehnholm, S. Lahdenpera, C. Ehnholm, and M. R. Taskinen. 1997. The Asn-291→Ser and Ser-477→Stop mutations of the lipoprotein lipase gene and their significance for lipid metabolism in patients with hypertriglyceridaemia. *Eur. J. Clin. Invest.* **27:** 928–935.
18. Rios, D. L., A. F. Vargas, G. M. Ewald, M. R. Torres, A. J. Zago, S. M. Callegari-Jacques, and M. H. Hutz. 2003. Common variants in the lipoprotein lipase gene in Brazil: association with lipids and angiographically assessed coronary atherosclerosis. *Clin. Chem. Lab. Med.* **41:** 1351–1356.
19. Rios, D. L., A. F. Vargas, G. M. Ewald, M. R. Torres, A. J. Zago, S. M. Callegari-Jacques, and M. H. Hutz. 1998. A common mutation in the lipoprotein lipase gene (N291S) alters the lipoprotein phenotype and risk for cardiovascular disease in patients with familial hypercholesterolemia. *Circulation.* **97:** 729–735.
20. Brousseau, M. E., A. L. Goldkamp, D. Collins, S. Demissie, A. C. Connolly, L. A. Cupples, J. M. Ordovas, H. E. Bloomfield, S. J. Robins, and E. J. Schaefer. 2004. Polymorphisms in the gene encoding lipoprotein lipase in men with low HDL-C and coronary heart disease: the Veterans Affairs HDL Intervention Trial. *J. Lipid Res.* **45:** 1885–1891.
21. van Bockxmeer, F. M., Q. Liu, C. Mamotte, V. Burke, and R. Taylor. 2001. Lipoprotein lipase D9N, N291S and S447X polymorphisms: their influence on premature coronary heart disease and plasma lipids. *Atherosclerosis.* **157:** 123–129.
22. McGladdery, S. H., and J. J. Frohlich. 2001. Lipoprotein lipase and apoE polymorphisms: relationship to hypertriglyceridemia during pregnancy. *J. Lipid Res.* **42:** 1905–1912.
23. Syvanne, M., M. Antikainen, S. Ehnholm, H. Tenkanen, S. Lahdenpera, C. Ehnholm, and M. R. Taskinen. 1996. Heterozygosity for Asn291→Ser mutation in the lipoprotein lipase gene in two Finnish pedigrees: effect of hyperinsulinemia on the expression of hypertriglyceridemia. *J. Lipid Res.* **37:** 727–738.
24. Ferencak, G., D. Pasalic, B. Grskovic, S. Cheng, B. Fijal, M. Sesto, J. Skodlar, and A. S. Rukavina. 2003. Lipoprotein lipase gene polymorphisms in Croatian patients with coronary artery disease. *Clin. Chem. Lab. Med.* **41:** 541–546.
25. Mero, N., L. Suurinkeroinen, M. Syvanne, P. Knudsen, H. Yki-Jarvinen, and M. R. Taskinen. 1999. Delayed clearance of postprandial large TG-rich particles in normolipidemic carriers of LPL Asn291Ser gene variant. *J. Lipid Res.* **40:** 1663–1670.
26. Pimstone, S. N., S. E. Gagne, C. Gagne, P. J. Lupien, D. Gaudet, R. R. Williams, M. Kotze, P. W. Reymer, J. C. Defesche, J. J. Kastelein, et al. 1995. Mutations in the gene for lipoprotein lipase: a cause for low HDL cholesterol levels in individuals heterozygous for familial hypercholesterolemia. *Arterioscler. Thromb. Vasc. Biol.* **15:** 1704–1712.
27. Wittrup, H. H., A. Tybjaerg-Hansen, and B. G. Nordestgaard. 1999. Lipoprotein lipase mutations, plasma lipids and lipoproteins, and risk of ischemic heart disease. A meta-analysis. *Circulation.* **99:** 2901–2907.
28. Zhang, Q., E. Cavallero, M. M. Hoffmann, J. Cavanna, A. Kay, A. Charles, S. Braschi, W. Marz, L. Perlemuter, B. Jacotot, et al. 1997. Mutations at the lipoprotein lipase gene locus in subjects with diabetes mellitus, obesity and lipaemia. *Clin. Sci. (Lond.).* **93:** 335–341.
29. Souverein, O. W., J. W. Jukema, S. M. Boekholdt, A. H. Zwinderman, and M. W. Tanck. 2005. Polymorphisms in APOA1 and LPL genes are statistically independently associated with fasting TG in men with CAD. *Eur. J. Hum. Genet.* **13:** 445–451.
30. Taylor, K. D., M. T. Scheuner, H. Yang, Y. Wang, T. Haritunians, N. Fischel-Ghodsian, P. K. Shah, J. S. Forrester, G. Knatterud, and J. I. Rotter. 2004. Lipoprotein lipase locus and progression of atherosclerosis in coronary-artery bypass grafts. *Genet. Med.* **6:** 481–486.
31. Mailly, F., J. Palmen, D. P. Muller, T. Gibbs, J. Lloyd, J. Brunzell, P. Durrington, K. Mitropoulos, J. Betteridge, G. Watts, et al. 1997. Familial lipoprotein lipase (LPL) deficiency: a catalogue of LPL gene mutations identified in 20 patients from the UK, Sweden, and Italy. *Hum. Mutat.* **10:** 465–473.
32. Razzaghi, H., C. E. Aston, R. F. Hamman, and M. I. Kamboh. 2000. Genetic screening of the lipoprotein lipase gene for mutations associated with high triglyceride/low HDL-cholesterol levels. *Hum. Genet.* **107:** 257–267.
33. Busca, R., J. Peinado, E. Vilella, J. Auwerx, S. S. Deeb, S. Vilaro, and M. Reina. 1995. The mutant Asn291→Ser human lipoprotein lipase is associated with reduced catalytic activity and does not influence binding to heparin. *FEBS Lett.* **367:** 257–262.
34. Spence, J. D., M. R. Ban, and R. A. Hegele. 2003. Lipoprotein lipase (LPL) gene variation and progression of carotid artery plaque. *Stroke.* **34:** 1176–1180.
35. Yang, Y., E. Ruiz-Narvaez, T. Niu, X. Xu, and H. Campos. 2004. Genetic variants of the lipoprotein lipase gene and myocardial infarction in the Central Valley of Costa Rica. *J. Lipid Res.* **45:** 2106–2109.
36. Hall, S., P. J. Talmud, D. G. Cook, P. D. Wicks, M. J. Rothwell, P. Strazzullo, G. A. Sagnella, and F. P. Cappuccio. 2000. Frequency and allelic association of common variants in the lipoprotein lipase gene in different ethnic groups: the Wandsworth Heart and Stroke Study. *Genet. Epidemiol.* **18:** 203–216.
37. Feliciello, I., S. Capossela, M. Cantile, A. Barbato, F. P. Cappuccio, P. Strazzullo, and G. Chinali. 2004. Pair-wise detection of eight common DNA polymorphisms of the human lipoprotein lipase gene by PCR-RFLP. Findings of the Olivetti Heart Study. *Nutr. Metab. Cardiovasc. Dis.* **14:** 205–210.
38. Jap, T. S., S. F. Jenq, Y. C. Wu, C. Y. Chiu, and H. M. Cheng. 2003. Mutations in the lipoprotein lipase gene as a cause of hypertriglyceridemia and pancreatitis in Taiwan. *Pancreas.* **27:** 122–126.
39. Benlian, P., J. L. De Gennes, L. Foubert, H. Zhang, S. E. Gagne, and M. Hayden. 1996. Premature atherosclerosis in patients with familial chylomicronemia caused by mutations in the lipoprotein lipase gene. *N. Engl. J. Med.* **335:** 848–854.
40. Yang, T., C. P. Pang, M. W. Tsang, C. W. Lam, P. M. Poon, L. Y. Chan, X. Q. Wu, B. Tomlinson, and L. Baum. 2003. Pathogenic mutations of the lipoprotein lipase gene in Chinese patients with hypertriglyceridemic type 2 diabetes. *Hum. Mutat.* **21:** 453–463.
41. Razzaghi, H., B. W. Day, R. J. McClure, and M. I. Kamboh. 2001. Structure-function analysis of D9N and N291S mutations in human lipoprotein lipase using molecular modeling. *J. Mol. Graph. Model.* **19:** 487–490.
42. Haffner, S. M., and H. Miettinen. 1997. Insulin resistance implications for type II diabetes mellitus and coronary heart disease. *Am. J. Med.* **103:** 152–162.
43. Rebrin, K., G. M. Steil, L. Getty, and R. N. Bergman. 1995. Free fatty acid as a link in the regulation of hepatic glucose output by peripheral insulin. *Diabetes.* **44:** 1038–1045.
44. Mcgarry, J. D. 2002. Dysregulation of fatty acid metabolism in the etiology of type 2 diabetes. *Diabetes.* **51:** 7–18.