

BMJ Open Effects of polymorphisms in APOA5 on the plasma levels of triglycerides and risk of coronary heart disease in Jilin, northeast China: a case-control study

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ABSTRACT

Objective The goal of this study is to investigate the associations of apolipoprotein A5 (*APOA5*) polymorphisms with coronary artery disease (CAD) in a Chinese population.

Method This case-control study included 710 subjects (355 patients with CAD and 355 controls) who were recruited from a cross-sectional study. Three single nucleotide polymorphisms (SNPs) rs662799 (-1131T>C), rs651821 (-3A>G) and rs2075291 (G185C) in *APOA5* were selected and genotyped using the matrix-assisted laser desorption/ionisation time of flight mass spectrometry technology. The χ^2 test and haplotype analysis were performed to analyse the associations between *APOA5* SNPs and CAD using the SPSS V.22.0 software package and the online SNPStats program.

Results *APOA5* SNPs rs662799 and rs651821 exhibited significant differences in genotype and allele distributions between patients with CAD and control subjects. The SNP rs662799 was significantly correlated with an increased risk of CAD when a dominant model was considered. The SNP rs651821 was significantly correlated with an increased risk of CAD when a codominant model was considered. Moreover, the variant C alleles of rs662799 and the variant G alleles of the rs651821 polymorphism were significantly correlated with increased plasma triglyceride (TG) levels in the CAD group (all $p < 0.05$). Additionally, a mediating effect of TG on the associations between the *APOA5* rs662799 and rs651821 polymorphisms and CAD was observed.

Conclusion Based on these data, variants of the *APOA5* gene are associated with CAD susceptibility and may modulate plasma TG levels among a Chinese population.

INTRODUCTION

Apolipoprotein A5 (*APOA5*) is a newly discovered member of the APOA4/APOC3/APOA1 apolipoprotein cluster that is located on chromosome 11q23 and has emerged as a significant modulator of serum triglyceride (TG) concentration.¹ Functional studies with transgenic mice have revealed that *APOA5*-overexpressing mice display lower TG levels than controls; conversely, *APOA5*-knockout mice

Strengths and limitations of this study

- This study is significant because it is the first to use a mediation model to examine whether triglyceride levels act as a mediator of the associations between apolipoprotein A5 polymorphisms and coronary artery disease.
- Each patient included in our study is from a household in the selected communities, indicating a good representation.
- The population size is limited; future studies are needed to validate our results.

exhibit higher TG concentrations, indicating that *APOA5* plays an important role in plasma TG metabolism.¹

According to epidemiological studies, increased TG concentrations are related to an increased risk of coronary artery disease (CAD), the most common cause of death among both women and men over the age of 50 years.² High TG levels in combination with low high-density lipoprotein cholesterol (HDL-C) levels account for twice as many cases of CAD as low HDL-C alone.³

In humans, previous studies have consistently reported associations between common variants of *APOA5* and differences in plasma TG levels.⁴⁻⁶ Consistent with this finding, the *APOA5* -1131T>C, c.-3A>G and c.56C>G variants exhibit very strong associations with elevated TG levels in different racial groups, including Chinese,⁷ African Americans, Caucasians⁸ and Japanese schoolchildren,⁶ as well as a higher relative risk of developing dyslipidaemia. The strong associations between common genetic variations in the *APOA5* gene and hypertriglyceridaemia raised the possibility that they may be related to CAD. As shown in a recent epidemiological study, the *APOA5* -1131T>C, c.-3A>G and c.56C>G variants are associated with an

increased risk of CAD, through their associations with hypertriglyceridaemia.^{4 9 10} However, there was report of a lack of association between *APOA5* -1131T>C and c.56C>G variants and CAD risk conflicts with these data.¹¹ This discrepancy could be explained by the observation that the distribution of the polymorphism of interest appears to be quite different in distinct ethnic populations, and these genetic differences may contribute to the varying prevalence rates of CAD among ethnic groups. Moreover, no other studies have confirmed these findings, and no similar studies have examined a northeastern Chinese population. Jilin is a special province because of specific weather, geographical location and eating habits of the population; thus, the prevalence of many chronic diseases, including CAD and metabolic syndrome, is high. Since different populations have contrasting profiles of CAD susceptibility and lipid risk factors, explorations of potential variations in the association between *APOA5* and CAD between different ethnicities are necessary. More importantly, the study has examined the association between the *APOA5* rs2075291 SNP and CAD. Therefore, in the present study, we chose two common SNPs, rs662799 and rs651821, and SNP rs2075291, which has not been described in a northeastern Chinese population and performed a case-control study to evaluate (1) whether *APOA5* polymorphisms rs662799, rs651821, rs2075291 and their haplotypes are associated with CAD in a northeastern Chinese population and (2) whether TG acted as a mediator of the associations of the *APOA5* polymorphisms with CAD using a mediation model.

MATERIALS AND METHODS

Patient and public involvement

A multistage stratified cluster sampling process was used to select the participants from nine areas of Jilin Province in northeast China in a community-based survey conducted in 2012. More detailed information about the sampling process is presented in previous publications.¹²⁻¹⁴ All participants were voluntarily involved in our research, including finishing a self-designed questionnaire that was previously developed to record information through person-to-person interviews. All trained staffs were involved in the recruitment to and conduct of the study. After the survey, we will send the results of all the participants to their home.

Sampling

In our study, 355 Han Chinese individuals with CAD from this survey were designated as cases, and 355 subjects without CAD were randomly selected from this survey as healthy controls. The inclusion criteria for the case group were: (a) angiographically confirmed coronary artery disease with $\geq 50\%$ occlusion of ≥ 1 major coronary arteries; (b) a confirmed myocardial infarction (MI) according to the WHO criteria for symptoms, elevated enzyme levels, or electrocardiographic changes; (c) no diagnosis of liver,

thyroid, pituitary or renal disease; and (d) no history of acute or chronic inflammatory diseases.

The inclusion criteria for the healthy subjects were no history of MI and no evidence of MI on an echocardiogram. Individuals with any history or diagnoses of vascular disease, cancer, liver disease, renal disease, thyroid disease or acute or chronic inflammatory disease were excluded.

All subjects included in the present study signed a written informed consent form. Our study was approved by the Ethics Committee of the School of Public Health of Jilin University.

Biochemical measurements

Information about the demographic parameters age, gender, height, weight, alcohol consumption and smoking status was obtained from a questionnaire and the data for the body mass index (BMI), waist circumference, hip circumference, systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate were measured in a physical examination. Alcohol consumption was categorised into three levels according self-reported drinking habits in number of times per week: normal drinker, frequent drinkers and never drinker. Subjects who drank more than three times per week were defined as frequent drinkers, subjects who drank more than one time but less than three times per week were defined as normal drinkers, and subjects who drank less than one time per week during their lifetimes were defined as never drinkers. The smoking status was categorised into current smoker, former smoker and never smoked, according to the self-reported number of cigarettes smoked and smoking days in the participants' lifetimes. Individuals who smoked at least 100 cigarettes in their lifetime and continued to smoke during the time of the survey were defined as current smokers, individuals who smoked at least 100 cigarettes but gave up smoking before the time of the survey were defined as former smokers and individuals who smoked <100 cigarettes during their lifetime were defined as never smokers. Systolic and diastolic blood pressure values were recorded by collecting an average of two measurements from each subjects while in a seated position after a 10 min rest. All subjects were asked to provide 5 mL of blood for biochemical analyses. The laboratory examinations included measurements of the levels of triglycerides (TG), total cholesterol (TC), low-density lipoprotein (LDL) and high-density lipoprotein (HDL). Blood samples were collected in the morning after an overnight fast, transported to the same laboratory under refrigeration and then stored at -20°C .

SNP selection

As described in previous studies, three SNPs rs2075291, rs662799 and rs651821 were selected by the Haploview program (<http://hapmap.ncbi.nlm.nih.gov/>) to identify the association between *APOA5* gene polymorphism and CAD. The minor allele frequencies of these three SNPs were >0.05 in the Chinese Han population.

DNA extraction and genotyping rate

Genomic DNA was extracted from peripheral blood lymphocytes using a commercially available DNA isolation kit (Hangzhou, China). SNP genotyping was performed by matrix-assisted laser desorption ionisation time of flight mass spectrometry (MALDI-TOF-MS) (Sequenom, San, Diego, California, USA) using the MassARRAY system. The detection rates for rs662799, rs651821 and rs2075291 were 99.4% (706/710), 99.7% (708/710) and 99.0% (703/710), respectively.

Statistical analysis

The analyses were performed using the SPSS software (V.22.0). The Hardy-Weinberg equilibrium (HWE) was examined with the online SNPstats program (<http://bioinfo.iconcologia.net/SNPstats>). Differences in the baseline characteristics between patients with CAD and controls were examined using Student's t-tests or χ^2 tests. Differences in genotype between the CAD and control groups were examined using χ^2 tests. The associations of CAD with the SNPs were examined using multivariate logistic regression analyses after adjusting for age, gender, BMI, smoking status, drinking, SBP and DBP. Pearson's χ^2 tests were applied to compare the allele frequency of each polymorphism between the cases and controls. Plasma lipid levels in the CAD group were compared between the different genotypes by analysis of covariance. We selected the best models of inheritance for the SNPs based on the Akaike information criterion, and the haplotypes and associations were estimated using the SNPstats program.¹⁵

The mediation analysis was based on the model proposed by Baron *et al.*¹⁶ The following three multivariate linear regression models were examined, all of which were adjusted for age and gender: (1) $Y=cX+p_1age+q_1gender+e_1$; (2) $M=axe+p_2age+q_2gender+e_2$ and (3) $Y=c_rX+bM+p_3age+q_3gender+e_3$. The statistical test of the mediation effect included the following steps: (3) the association between the independent variable and the dependent variable (coefficient c) was assessed; (2) the association between the independent variable and the potential mediator (the coefficient a) was assessed; (3) both the independent variable and the potential mediator were simultaneously entered as predictors of the dependent variable, and the coefficient b was determined to establish the significance of the mediation effect; and (4) if the mediation effect was significant, the type of mediation effect was determined by calculating the coefficient c_r , which indicated either a full mediation effect (c_r not significant) or a partial mediation effect (c_r remained significant).

All p values were two-tailed and p values <0.05 were considered statistically significant.

RESULTS

Characteristics of the CAD patients and controls

The general characteristics of the 355 patients with CAD and the 355 controls are presented in table 1. The mean ages were 55.96 \pm 8.51 years in the case group and 53.90 \pm 7.51 years in the control group. The frequencies

Table 1 Clinical and biochemical characteristics of controls and patients with CAD

Parameters	Case(n=355)	Control(n=355)
Age (years)	55.96 \pm 8.51	53.90 \pm 7.51
Male, n (%)**	118 (33.20)	181 (51.00)
BP		
SBP (mm Hg)*	137.82 \pm 23.62	118.96 \pm 13.62
DBP (mm Hg)*	82.62 \pm 13.05	73.93 \pm 8.56
Smoking, n (%)**		
Current	89 (25.10)	114 (32.10)
Former	37 (10.40)	23 (6.50)
Never	229 (64.50)	218 (61.40)
Drinking, n (%)*	68 (19.20)	106 (29.86)
BMI (kg/m ²)*	25.23 \pm 4.02	21.91 \pm 2.74
TG (mmol/L)*	2.23 \pm 1.70	1.05 \pm 0.49
TC (mmol/L)*	5.24 \pm 1.09	4.68 \pm 0.93
LDL-C (mmol/L)*	3.18 \pm 0.92	2.77 \pm 0.78
HDL-C (mmol/L)*	1.37 \pm 0.42	1.52 \pm 0.24

Continuous variables are presented as mean \pm SD, and n (%) for frequency variables.

P values were analysed using Student's t-test and χ^2 test.

* $p<0.05$; ** $p<0.001$.

BP, blood pressure; BMI, body mass index; DBP, diastolic blood pressure; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; TC, total cholesterol; TG, triglyceride.

of smoking and drinking were imbalanced between the different groups. As expected, compared with controls, patients with CAD exhibited a higher incidence of conventional cardiovascular risk factors, including higher SBP, DBP, BMI, plasma total cholesterol, TG and LDL-C levels, but lower plasma HDL-C levels.

Distribution of the allele frequencies of the SNPs in the control and CAD patient groups

The distributions of three SNPs (rs662799, rs651821 and rs2075291) were in HWE in the entire population as well as separately in the cases and controls. The comparisons of the genotype distributions and allele frequencies of the polymorphisms in the *APOA5* gene between the subjects with and without CAD are presented in table 2. Two SNPs rs662799 and rs651821 exhibited significant differences in genotype distributions between the controls and patients with CAD ($p<0.05$ for all comparisons). Moreover, significant differences in allele distributions were observed between the controls and patients with CAD. However, differences in the genotype and allele frequency of SNP rs2075291 were not observed between the CAD and control groups.

Genotype distribution comparison

Table 3 presents the results of the multivariate logistic regression analysis employing the best selected model after adjusting for the confounding factors of age, gender, BMI, smoking status, drinking, systolic blood pressure and diastolic blood pressure. CAD was significantly associated with SNPs rs662799 and rs651821. In the dominant

Table 2 Comparison of genotype distributions and allele frequencies of polymorphisms in the *APOA5* gene between subjects with and without CAD

SNPs	Genotype/Allele		Case(%)	Control(%)	χ^2	P values
rs2075291	Genotype	GG	167 (47.00)	198 (55.80)	5.131	0.077
		GC	100 (28.20)	81 (22.80)		
		CC	83 (23.40)	74 (20.80)		
	Allele	G	434 (61.13)	477 (67.56)	2.344	0.872
		C	266 (38.87)	229 (32.44)		
rs662799	Genotype	TT	123 (34.70)	179 (50.90)	21.598	<0.001
		TC	149 (42.10)	97 (27.60)		
		CC	82 (23.20)	76 (21.60)		
	Allele	T	395 (55.79)	455 (64.63)	11.512	<0.001
		C	313 (44.21)	249 (35.37)		
rs651821	Genotype	AA	137 (38.70)	188 (53.10)	17.450	<0.001
		AG	130 (36.70)	113 (31.90)		
		GG	87 (24.60)	53 (15.00)		
	Allele	A	404 (57.06)	489 (69.07)	21.905	<0.001
		G	304 (42.94)	219 (30.933)		

CAD, coronary artery disease.

model, we observed a significant association between CAD risk and the SNP rs662799. In the codominant model, we also observed an association between CAD risk and SNP rs2075291.

APOA5 haplotype analysis

We performed a haplotype analysis to determine whether the three SNPs in the *APOA5* gene were associated with CAD. Three haplotypes were identified in the *APOA5* gene in our population. As shown in table 4, two haplotypes were associated with an increased risk of CAD (ORs=1.64 and 2.19).

APOA5 SNPs rs2075291, rs662799 and rs651821 and the plasma lipid profile

Table 5 presents the mean plasma lipid levels among the genotypes of *APOA5* SNPs rs2075291, rs662799 and rs651821 in the CAD groups. As expected, the *APOA5* rs662799 C alleles and rs651821 G allele significantly

correlated with increased plasma TG levels in the CAD group (all $p < 0.05$). However, no other significant differences were detected between the *APOA5* polymorphism in SNP rs2075291 and plasma TG levels.

Mediation analysis of the *APOA5* rs662799 and rs651821 polymorphisms, TG and CAD

We used a mediation model to examine whether TG levels acted as a mediator of the associations of the *APOA5* rs651821 and rs662799 polymorphisms with CAD as illustrated in figure 1A. Here, 'a', 'b', 'c' and 'c_r' are used to represent the coefficients in the mediation analysis model. The number of *APOA5* rs651821 G alleles positively correlated with CAD ($c = 0.9029$, $p < 0.001$). A positive correlation was observed between the number of the *APOA5* rs651821 G alleles and TG levels ($a = 0.4408$, $p < 0.05$). By testing 'b', we observed a significant mediation effect ($b = 1.6916$, $p < 0.05$), and the insignificance

Table 3 Genotype distribution comparisons and OR estimates of three SNPs between CAD subjects and controls

SNPs	Model	Genotype	Case (%)	Control (%)	OR (95% CI)*	P values
rs2075291	Recessive	G/G- G/C	267 (76.3)	279 (79.0)	1.00	0.300
		C/C	83 (23.7)	74 (21.0)	1.22 (0.84 to 1.76)	
rs662799	Dominant	T/T	123 (34.8)	179 (50.9)	1.00	<0.001
		T/C- C/C	231 (65.2)	173 (49.1)	1.61 (1.09 to 2.37)	
rs651821	Codominant	A/A	137 (38.7)	188 (53.1)	1.00	<0.001
		G/A	130 (36.7)	113 (31.9)	1.49 (1.04 to 2.16)	
		G/G	87 (24.6)	53 (15.0)	2.23 (1.43 to 3.47)	

*Adjusted for age, gender, BMI, smoking status, drinking, systolic blood pressure and diastolic blood pressure. BMI, body mass index; CAD, coronary artery disease; SNP, single nucleotide polymorphism.

Table 4 Association analysis of haplotypes derived from polymorphic sites using genotype data

Haplotype	SNP			Frequency			Adjusted OR (95% CI)*	P values
	rs651821	rs662799	rs2075291	Total	Cases	Controls		
1	A	C	G	0.7169	0.7505	0.6636	1.00	–
2	G	T	G	0.2171	0.1903	0.2594	1.64 (1.19–2.28)	0.003
3	G	T	T	0.0596	0.0509	0.0733	2.19 (1.22–3.95)	0.009
Rare†	*	*	*	0.0065	0.0023	0.0038	0.33 (0.05–2.40)	0.280

*OR was adjusted for age, gender, BMI, smoking status, drinking, systolic blood pressure and diastolic blood pressure.

†Haplotypes with frequencies <0.01.

BMI, body mass index; SNP, single nucleotide polymorphism.

of 'c_r' indicated a partial mediation effect ($c_r=0.1553$, $p<0.05$). Further calculations ($ab/c\times 100\%$) revealed a mediation effect of 82.58%. Moreover, similar results were obtained in the mediation analysis; TG levels exerted a partial mediation effect on the association between the APOA5 rs662799 polymorphism and CAD (figure 1B); the mediation effect was ($ab/c\times 100\%$) 46.60%.

DISCUSSION

Our case–control study including Chinese subjects revealed that SNPs rs662799 and rs651821 were significantly associated with an increased risk of CAD that was accompanied by elevated plasma TG levels after adjusting for age, gender, BMI, smoking status, drinking status, SBP and DBP. The C allele frequency of APOA5 rs662799 (30%) observed here was similar to frequencies observed in previously studies of Japanese¹⁷ and Chinese subjects^{7 18 19} but was much higher than the frequency reported for Caucasian populations (10%).

The association observed in our study between the APOA5 rs662799 variant and the increased risk of CAD

is consistent with previous reports examining Korean²⁰ and Czech populations.²¹ A recent study also reported similar results²²; Szalai *et al* reported a significantly higher frequency of the APOA5 C allele among patients with CAD than among controls (OR=1.98; 95% CI 1.14 to 3.48; $p<0.001$). However, other studies have not identified similar associations.^{23 24} In the present study, the APOA5 rs651821 polymorphism was a risk factor for CAD among a Chinese population. Our results are not consistent with the results of previous studies that have supported the hypothesis that rs651821 is not significantly associated with CAD risk in a Chinese population.^{25–27} This discrepancy might be explained by the fact that our study was a case–control study, and the cases were limited to survivors of CAD. This difference would certainly affect the full influence of the examined polymorphisms.

Some studies have reported that APOA5 polymorphisms are associated with lipid levels among diverse ethnic groups.^{1 5 28–30} We also successfully confirmed this association in the present study. For both APOA5 rs662799 and rs651821, patients with CAD presenting with the CC and

Table 5 Association between APOA5 genotype variants and clinical and biochemical parameters in CAD group

SNP	Genotype	Parameters			
		TG (mmol/L)	TC (mmol/L)	LDL-C (mmol/L)	HDL-C (mmol/L)
rs2075291	GG (n=135)	2.302±1.785	5.229±1.076	3.087±0.847	1.395±0.504
	GC (n=132)	2.640±1.958	5.204±1.167	3.230±1.077	1.181±0.295
	CC (n=83)	1.719±1.043	5.334±1.054	3.371±0.930	1.486±0.209
	P values	0.052	0.705	0.058	0.056
rs662799	TT (n=123)	1.900±1.339	5.228±1.012	3.226±0.904	1.439±0.416
	TC (n=100)	2.507±1.913	5.274±1.187	3.165±0.939	1.297±0.403
	CC (n=131)	2.811±2.121	5.125±1.149	3.009±0.982	1.266±0.440
	P values	0.010	0.764	0.428	0.403
rs651821	AA (n=137)	1.914±1.352	5.221±1.017	3.215±0.904	1.441±0.416
	AG (n=130)	2.496±1.913	5.295±1.181	3.182±0.936	1.303±0.412
	GG (n=87)	2.884±2.110	5.071±1.156	2.971±0.992	1.214±0.382
	P values	0.001	0.543	0.363	0.401

P values were calculated by ANCOVA.

ANCOVA, analysis of variance; CAD, coronary artery disease; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; TC, total cholesterol; TG, triglyceride.

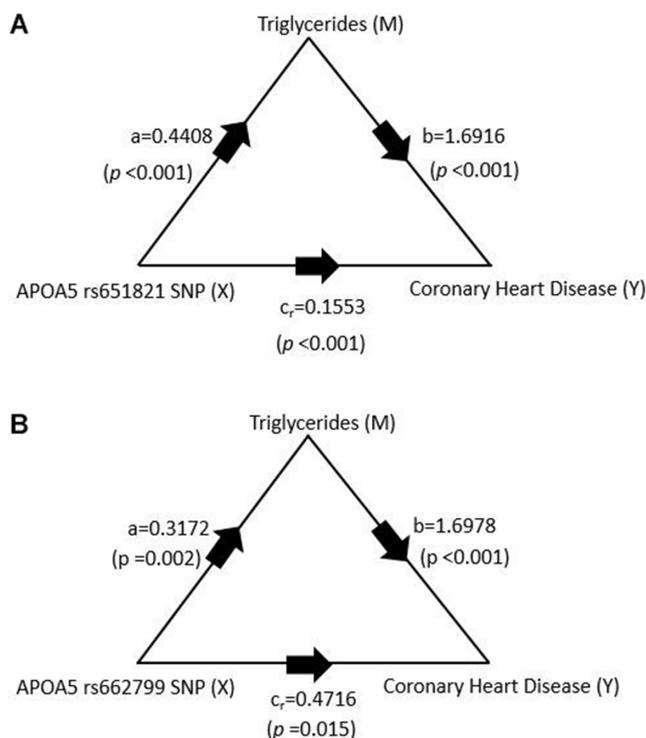


Figure 1 Mediation analysis APOA5 rs651821 and rs662799 SNP, TGs and CAD. In order to distinguish among the three linear regression models constructed in the mediation analysis, the coefficients in the model were represented by 'a,' b,' 'c' and 'c_r'. The first regression model used 'c' as the coefficient of SNP in association with CAD; the second model used 'a' as the coefficient of SNP in association with TGs; the third model put TGs and SNP as independent variables simultaneously, and used 'b' and 'c_r' as the coefficients in association with CAD. 'X', 'Y' and 'M' were used to represent the independent variable, the dependent variable and the mediator in the mediation analysis model. APOA5, apolipoprotein A5; CAD, coronary heart disease; SNP, single nucleotide polymorphism; TG, triglyceride.

GG genotypes exhibited higher TG levels than subjects with the TT and AA genotypes, respectively. However, we did not observe a significant association between the APOA5 rs2075291 SNP and plasma TG levels. Similar to previous reports, the TG levels were approximately 25% higher in carriers of the APOA5 rs662799 C allele than in non-carriers, indicating that this allele is also an important genetic determinant of triglyceride levels in our Chinese population.

Researchers have not clearly determined whether the increased risk of CAD is due to the function of APOA5 per se or mediated by changes in the atherogenic lipid profile. Hence, we used a mediation model to examine whether TG levels acted as a mediator of the associations of the APOA5 rs651821 and rs662799 polymorphisms with CAD. TG levels were a significant mediator in this model. The mediation model was used for the first time to analyse the associations between APOA5 polymorphisms, TG levels and CAD. Our study identified a novel mechanism underlying the established association between

APOA5 polymorphisms and CAD in large epidemiological studies. Based on accumulating evidence, high TG levels and genetic variations are associated with increased cardiovascular morbidity and mortality risks.³¹ However, many determinants have not yet been clarified because genetic variation explains only 5%–50% of the overall variations in TG levels and CAD risk.³² Environmental and lifestyle variations may affect the observed associations between the genetic variants and disease outcomes.

Our study has certain limitations. First, glycaemia is one of the most important CV risk factors; thus, diabetes should have been included in our study. However, because of limited funding, we were unable to test for diabetes and plasma APOA5 levels; we plan to perform a further study in the future when we have sufficient funding. Second, our study was a case–control study; thus, the cause–effect relationships between the APOA5 rs662799 and rs651821 genotypes and TG and HDL-C concentrations were not easy to establish. In addition, CAD is a complex disease and several SNPs have been reported to correlate with CAD and myocardial ischaemia in other populations.^{33–37} We did not study these SNPs because of limited funding. CAD is a complex disease with a complex pathophysiology caused by interactions between multiple endogenous and exogenous factors. Many diagnostic criteria for CAD have been reported. In our study, all healthy subjects had no history of MI and no evidence of MI on an echocardiogram, but this method may not exclude potential patients with CAD who are asymptomatic and did not experience an infarction. Therefore, in future studies, subjects without CAD should be chosen using stricter criteria. Despite these limitations, our study confirmed that common variants of the APOA5 gene are associated with CAD and contribute to the variation in human plasma TG levels. Therefore, APOA5 variants are significant predictors of a high TG risk. Variations in this gene may be associated with the development of CAD. As reported in the recent study by Caussy *et al.*³⁸ miRNAs may be involved in the association of APOA5 with CAD; therefore, we plan to perform additional studies to confirm our hypothesis of the functions of these SNPs and validate our findings.

CONCLUSION

In conclusion, APOA5 SNPs rs662799 and rs651821 were significantly associated with an increased risk of CAD and accompanied by elevated plasma TG levels. According to the results of the mediation analysis, the effects of rs662799 and rs651821 on CAD were mediated by TG levels.

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Competing interests None declared.

Patient consent Obtained.

Ethics approval All participants in this study provided written informed consent form and our study was approved by the ethics committee of the School of Public Health, Jilin University (reference number: 2012-R-011).

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