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Original Article

Association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and serum lipid levels

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Abstract: The objective of the present study was to detect the association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and serum lipid levels in the Mulao and Han populations. A total of 879 subjects of Mulao and 844 subjects of Han Chinese were included. The levels of serum total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C) and ApoA1 in Mulao, and triglyceride (TG), high-density lipoprotein cholesterol (HDL-C), ApoA1 and the ratio of ApoA1/ApoB in Han were different among the three genotypes of the rs1801690 SNP ($P < 0.05$-0.01). Subgroup analyses showed that the levels of TC, TG, LDL-C, and ApoA1 in Mulao males; ApoA1 in Mulao females; TC, TG, HDL-C and ApoB and the ApoA1/ApoB ratio in Han males; and HDL-C, ApoA1 and the ApoA1/ApoB ratio in Han females were associated with the genotypes of rs1801690 ($P < 0.05$-0.001). Serum lipid parameters were also associated with several environmental factors ($P < 0.05$-0.001). The Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene was associated with some serum lipid parameters in the two ethnic groups, but the trends of association suggest that the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene might have racial/ethnic-and/or gender-specificity.

Keywords: Lipids, apolipoprotein H, β2-glycoprotein-I, the Trp316Ser variant, environmental factor

Introduction

A number of case-control as well as prospective studies revealed an increased risk of cardiovascular disease (CVD) in patients with dyslipidemia [1]. Heritability studies based on twins suggested that approximately one third to one half of the variation in the serum lipid levels peak particle size can be attributed to genetic influences [2, 3]. Therefore, the understanding of the association of single nucleotide polymorphisms (SNP) and serum lipid levels has become crucial in the pursuit of reducing CVD [4].

Human apolipoprotein H (ApoH), also known as β2-glycoprotein I (β2GPI) (in this study, we will use *ApoH* to refer to the gene as used in human genome databases and β2GPI to refer to the protein as commonly used in the rheumatology literature) is a major autoantigen recognized by predominant antiphopholipid antibodies (APA) found in sera of many autoimmune diseases such as primary antiphospholipid syndrome (PAPS), systemic lupus erythematosus (SLE) [5, 6], plasma lipid levels [7, 8], Cerebrovascular disease [9]. *ApoH* spans 18 kilobases (kb) on chromosome 17q23-24 [10] and encodes for a mature protein of 326 amino acid (aa) residues. The β2GPI is a 50-kDa single chain plasma glycoprotein exhibiting internal homology comprised of four contiguous homologous regions of about 60 aa residues, and an additional variable fifth C-terminal domain. The variable configuration of the fifth domain is essential for the binding of β2GPI to anionic phospholipids [11-13]. Primer extensions determined alternate transcription start sites (TSSs) at 31 base pairs

(bp) and 21 bp upstream of the *ApoH* translation start codon [10]. TSS 31 bp upstream agreed completely with the consensus for an initiator element (Inr) known to sustain transcription initiation. Previously [14], an atypical TATA box and HNF-1α cis-elements have been identified to be critical for *ApoH* cell type-specific transcriptional regulation leading to differential expression of *ApoH* in humans.

The β2GPI is primarily expressed in the liver and sporadically in intestinal cell lines and tissues [15]. The plasma concentration of β2GPI is approximately 20 mg/dL of which a small portion is bound to lipoproteins and the rest exists in lipid free form [16-18]. There is a wide range of interindividual variation in β2GPI plasma levels, ranging from immunologically undetectable to as high as 35 mg/dL with a mean value of 20 mg/dL in Caucasians and 15 mg/dL in African Americans [19], which may have clinical relevance in β2GPI-related pathways. Family and heritability data have provided strong support for the genetic basis of β2GPI plasma variation but the exact molecular basis of this variation remains largely unknown. The β2GPI is suggested to regulate thrombin inactivation by heparin cofactor II [20] and thus variation in plasma β2GPI may affect prothrombic tendency in PAPs patients. Thus, it is important to determine the molecular basis of β2GPI plasma variation. Previously we have characterized complete DNA sequence variation in ApoH and identified~150 SNPs, including 13 SNPs and 1 deletion (-742delT) in the 5'-region [21]. Kamboh MI et al. reported that of the 21 QC-passed SNPs present in or near ApoH, six revealed nominal associations with anti-β2GPI, and the Trp316Ser variant (rs1801690) was the most significant SNP (P = 3.12E-03) [22].

It has been implicated in the development of atherosclerosis this glycoprotein can reduce cellular accumulation of cholesterol by decreasing cholesterol influx and increasing cholesterol efflux [23]. It activates lipoprotein lipase [24], and genetic variations in this gene have been associated with variation in high-density lipoprotein cholesterol (HDL-C) and triglyceride (TG) levels [25-27]. *ApoH* is a novel apolipoprotein mainly associated with low and very low-density lipoprotein (VLDL). Genome-wide association studies (GWASs) have identified many SNPs underlying variations in plasma lipid levels. We explore whether additional loci associated with plasma lipid phenotypes, such as HDL-C, low-density lipoprotein cholesterol (LDL-C), total cholesterol (TC), and TG, can be identified by a dense gene-centric approach. A previous study identified several lipid-related SNPs in previously unreported genes including ApoH for LDL-C [28]. However, little is known about the exact association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene with serum lipid levels.

Among a total of 56 ethnic groups in China, Han is the largest one. Mulao, on the other hand, is one of the minorities with a population of 207,352 according to the China's fifth national census in 2000. They live in the Luocheng Mulao Autonomous County, Guangxi Zhuang Autonomous Region. They call themselves "Ling" and a smaller group call themselves "Jin" or "Bendiren". Historical data trace the history of this ethnic minority back to the Jin Dynasty (AD 265-420). It is believed that the Mulao people are the descendants of the ancient "Baiyue tribe" in southern China and ethnically related to the neighboring ethnic groups. In a previous study, Xu *et al*. [29] showed that the genetic relationship between Mulao nationality and other minorities in Guangxi was much closer than that between Mulao and Han or Uighur nationalities. To our knowledge, no prior study has evaluated the association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and serum lipid levels between the Mulao and Han populations. This study was undertaken to assess the association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and several environmental factors with serum lipid profiles in the Mulao and Han populations.

Materials and methods

Subjects

The study populations including 879 unrelated participants (434 males, 49.37% and 445 females, 50.63%) of Mulao and 844 unrelated subjects (417 males, 49.41% and 427 females, 50.59%) of Han were randomly selected from our previous stratified randomized samples [30, 31]. All participants were agricultural workers from Luocheng Mulao Autonomous County, Guangxi Zhuang Autonomous region, People's Republic of China. The age of the participants

ranged from 15 to 80 years with the mean age of 52.59 ± 14.74 years in Mulao and 52.43 ± 14.01 years in Han; respectively. The age distribution and gender ratio were matched between the two groups. All participants were essentially healthy with no history of CVD such as coronary artery disease and stroke, diabetes, hyper- or hypo-thyroids, and chronic renal disease. They were free from medications known to affect serum lipid levels. Informed consent was taken from all participants. The study design was approved by the Ethics Committee of the First Affiliated Hospital, Guangxi Medical University.

Epidemiological survey

The epidemiological survey was carried out using internationally standardized methods, following a common protocol [30, 31]. Information on demographics, socioeconomic status, and lifestyle factors was collected with standardized questionnaires. Alcohol consumption was categorized into groups of grams of alcohol per day: < 25 and ≥ 25. Smoking status was categorized into groups of cigarettes per day: < 20 and ≥ 20. Several parameters such as blood pressure, height, weight, waist circumference, and body mass index (BMI) were measured. The methods of measuring above parameters were referred to previous studies [32-35].

Biochemical measurements

A fasting venous blood sample of 5 ml was drawn from the participants. The levels of TC, TG, HDL-C and LDL-C in the samples were determined by enzymatic methods with commercially available kits. Serum ApoA1 and ApoB levels were assessed by the immuneturbidimetric immunoassay [36, 37].

Genotyping

Genomic DNA was isolated from peripheral blood leukocytes using the phenol-chloroform method [32-35]. The Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene was genotyped by polymerase chain reaction and restriction fragment length polymorphism (PCR-RFLP). PCR amplification was performed using 5'-CAAAGAAATCTCTATCTCAC-3' as the forward and 5'-ACCAGCCTGGCCAACATGGT-3' as reversed primer pair. Each amplification reaction was performed in a total volume of 25 μl, 12.5 μl of 2 × *Taq* PCR MasterMix (constituent: 0.1 U *Taq* polymerase/μl, 500 μM dNTP each and PCR buffer) and nuclease-free water 8.5 μl, 20 pmol/L of each primer and 100 ng of genomic DNA, processing started with 5 min of pre-denaturing at 95°C and followed by 45 s of denaturing at 94°C, 30 s of annealing at 60°C and 1 min of elongation at 72°C for 35 cycles. The amplification was completed by a final extension at 72°C for 10 min. Then each restriction enzyme reaction was performed with 10 μl of amplified DNA, 8 μl of nuclease-free water, 1 μl of 10 × buffer solutions, and 10 U of '*Mob* II' enzyme in a total volume of 20 μl digested at 37°C overnight. After restriction enzyme digestion of the amplified DNA, the digestive products were separated by electrophoresis on 2% agarose gel. The length of each digested DNA fragment was determined by comparing migration of a sample with that of standard DNA marker. Genotypes were scored by an experienced reader blinded to the epidemiological and lipid results. Six samples (each genotype in two; respectively) detected by the PCR-RFLP were also confirmed by direct sequencing. The PCR products were purified by low melting point gel electrophoresis and phenol extraction, and then the DNA sequences were analyzed using an ABI Prism 3100 (Applied Biosystems) in Shanghai Sangon Biological Engineering Technology & Services Co., Ltd., People's Republic of China.

Diagnostic criteria

The normal values of serum TC, TG, HDL-C, LDL-C, ApoA1 and ApoB levels, and the ratio of ApoA1 to ApoB in our Clinical Science Experiment Center were 3.10-5.17, 0.56-1.70, 1.16-1.42, 2.70-3.10 mmol/L, 1.20-1.60, 0.80-1.05 g/L, and 1.00-2.50; respectively [38, 39].

Statistical analysis

The statistical analyses were performed with the statistical software package SPSS 17.0 (SPSS Inc., Chicago, Illinois). The quantitative variables were presented as mean ± standard deviation (serum TG levels were presented as medians and interquartile ranges). Allele frequency was determined via direct counting, and the Hardy-Weinberg equilibrium was verified with the standard goodness-of-fit test. The

Table 1. Comparison of demographic, lifestyle characteristics and serum lipid levels between the Mulao and Han populations

Parameter	Mulao	Han	t (X^2)	P
Number	879	844		
Male/female	434/445	417/427	0.000	0.989
Age (years)	52.59 ± 14.74	52.43 ± 14.01	0.223	0.824
Height (cm)	155.09 ± 7.93	155.19 ± 7.91	-0.252	0.801
Weight (kg)	52.67 ± 9.47	54.29 ± 9.27	-3.582	0.000
Body mass index (kg/m^2)	21.83 ± 3.13	22.51 ± 3.33	-4.345	0.000
waist circumference (cm)	75.21 ± 8.56	75.84 ± 8.03	-1.582	0.114
Cigarette smoking (n %)				
Nonsmoker	675 (76.8)	574 (68.0)		
< 20 cigarettes/day	74 (8.4)	102 (12.1)	16.763	0.000
≥ 20 cigarettes/day	130 (14.8)	168 (19.9)		
Alcohol consumption [n (%)]				
Nondrinker	679 (77.2)	631 (74.8)		
< 25 g/day	86 (9.8)	61 (7.2)	10.733	0.005
≥ 25 g/day	114 (13.0)	152 (18.0)		
Systolic blood pressure (mmHg)	129.62 ± 21.95	130.61 ± 18.37	-1.011	0.312
Diastolic blood pressure (mmHg)	81.03 ± 11.56	82.71 ± 10.68	-3.146	0.002
Pulse pressure (mmHg)	48.60 ± 16.95	47.89 ± 14.18	0.937	0.349
Glucose (mmol/L)	6.04 ± 1.69	6.23 ± 1.98	-2.112	0.035
Total cholesterol (mmol/L)	5.02 ± 1.28	5.15 ± 1.23	-2.016	0.044
Triglyceride (mmol/L)	1.04 (0.76)	1.12 (0.82)	-3.712	0.000
HDL-C (mmol/L)	1.78 ± 0.50	1.72 ± 0.53	2.236	0.025
LDL-C (mmol/L)	2.92 ± 0.92	2.89 ± 0.92	0.589	0.556
Apolipoprotein (Apo)A1 (g/L)	1.30 ± 0.45	1.36 ± 0.27	-3.174	0.002
ApoB (g/L)	1.02 ± 0.63	0.88 ± 0.22	6.326	0.000
ApoA1/ApoB	1.59 ± 1.17	1.63 ± 0.52	-1.040	0.298

HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol. The value of triglyceride was presented as median (interquartile range), the difference between the two ethnic groups was determined by the Wilcoxon-Mann-Whitney test.

genotype distribution between the groups was analyzed by the chi-square test. General characteristics between two ethnic groups were compared by the Student's unpaired *t*-test. The association between genotypes and serum lipid parameters was tested by analysis of covariance (ANCOVA). Age, sex, BMI, cigarette smoking, and alcohol consumption were adjusted for the statistical analysis. Multivariable linear regression analyses with stepwise modeling were used to determine the correlation between genotypes (GG = 1, GC = 2, CC = 3) or alleles (the C allele non-carrier = 1, the C allele carrier = 2) and several environmental factors with serum lipid levels in males and females of Mulao and Han populations. Two sided P value < 0.05 was considered statistically significant.

Results

General and biochemical characteristics of the subjects

The general characteristics and serum lipid levels between the Mulao and Han populations are summarized in **Table 1**. The levels of body weight, BMI, waist circumference, systolic blood pressure, diastolic blood pressure, pulse pressure and blood glucose were lower in Mulao than in Han (P < 0.05-0.001), whereas the percentage of alcohol consumption and the levels of ApoB was higher in Mulao than in Han (P < 0.05-0.001). There were no significant differences in the gender ratio, age structure, body height, the percentage of cigarette smoking, serum TC, TG, HDL-C, LDL-C and ApoA1 lev-

Table 2. Comparison of the genotype and allele frequencies of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene between males and females of the Mulao and Han populations

Group	n	Genotype			Allele	
		GG	GC	CC	G	C
Mulao	879	546 (62.1)	218 (24.8)	115 (13.1)	1310 (74.5)	448 (25.5)
Han	844	553 (65.5)	193 (22.9)	98 (11.6)	1299 (77.0)	389 (23.0)
X^2	–		2.212		2.784	
P	–		0.331		0.095	
Mulao						
Male	434	288 (66.4)	96 (22.1)	50 (11.5)	672 (77.4)	196 (22.6)
Female	445	258 (58.0)	122 (27.4)	65 (14.6)	638 (71.7)	252 (28.3)
X^2	–		6.569		7.608	
P	–		0.037		0.006	
Han						
Male	417	283 (67.9)	96 (23.0)	38 (9.1)	662 (79.4)	172 (20.4)
Female	427	270 (63.2)	97 (22.7)	60 (14.1)	637 (74.6)	217 (25.4)
X^2	–		5.132		5.451	
P	–		0.077		0.020	

els and the ApoA1/ApoB ratio between the two ethnic groups ($P > 0.05$ for all).

Results of genotyping

After the genomic DNA of the samples was amplified by PCR, the purpose gene of 496-bp nucleotide sequences could be seen in all samples. The genotypes identified were labeled according to the presence or absence of the enzyme restriction sites. Thus, GG genotype is homozygote for the absence of the site (496-bp), GC genotype is heterozygote for the presence and absence of the site (496-, 288- and 208-bp) and CC genotype is homozygote for the presence of the site (288- and 208-bp). The GG, GC and CC genotypes detected by PCR-RFLP were also confirmed by direct sequencing.

Genotypic and allelic frequencies

As shown in **Table 2**, there was no significant difference in the genotypic and allelic frequencies of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene between the Mulao and Han populations ($P > 0.05$ for each). The genotype frequencies of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene agreed with the Hardy-Weinberg equilibrium in both populations ($P > 0.05$ for each). Gender-subgroup analysis showed that the genotype and allele frequencies were different between Mulao males and females (GG, 66.4% *vs.* 58.0%; GC, 22.1% *vs.* 27.4%, CC, 11.5% *vs.* 14.6%, $P = 0.037$; G, 77.4% *vs.* 71.7%; C, 22.6% *vs.* 28.3%, $P = 0.006$). There was no significant difference in the genotypic frequencies of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene between the Han males and females ($P > 0.005$). The allele frequencies were significantly different between Han males and females (G, 79.4% *vs.* 74.6%; C, 20.4% *vs.* 25.4%, $P = 0.020$).

Genotypes and serum lipid levels

Tables 3 and **4** describe the association between genotypes and serum lipid levels. The levels of TC, LDL-C and ApoA1 in Mulao, and TG, HDL-C, ApoA1 and the ratio of ApoA1 to ApoB in Han were different among the genotypes ($P < 0.05$-0.01). Gender-subgroup analysis showed that the levels of TC, TG, LDL-C, and ApoA1 in Mulao males, ApoA1 in Mulao females; TC, TG, HDL-C and ApoB and the ratio of ApoA1 to ApoB in Han males, and HDL-C, ApoA1 and the ratio of ApoA1 to ApoB in Han females were associated with the genotypes ($P < 0.05$-0.001).

Relative factors for serum lipid parameters

Several environmental factors such as age, gender, height, weight, waist circumference, alcohol consumption and cigarette smoking, and traditional cardiovascular risk factors such as BMI, fasting blood glucose and blood pressure levels were also correlated with serum lipid parameters in the Mulao and Han populations and in males and females of both ethnic groups ($P < 0.05$-0.001, **Tables 5** and **6**).

Discussion

It is a truth universally acknowledged that serum lipid levels are highly genetically determined. Family studies state that genetic factors

Table 3. Comparison of the genotypes and serum lipid levels in the Mulao and Han populations

Ethnic/Genotype	N	TC (mmol/L)	TG (mmol/L)	HDL-C (mmol/L)	LDL-C (mmol/L)	Apo A1 (g/L)	ApoB (g//L)	ApoA1/ApoB
Mulao								
GG	546	5.08 ± 1.21	1.10 (0.81)	1.74 ± 0.43	2.98 ± 0.90	1.21 ± 0.48	1.35 ± 0.39	1.65 ± 1.30
GC	218	4.96 ± 1.16	0.97 (0.73)	1.84 ± 0.56	2.89 ± 0.87	1.24 ± 0.58	1.21 ± 0.48	1.43 ± 0.86
CC	115	4.87 ± 1.71	0.97 (0.63)	1.84 ± 0.64	2.71 ± 1.09	1.35 ± 0.39	1.24 ± 0.58	1.55 ± 0.97
F		4.337	3.013	1.916	7.415	9.832	1.081	2.498
P		0.013	0.050	0.148	0.001	0.000	0.340	0.083
Han								
GG	553	5.22 ± 1.37	1.14 (0.86)	1.68 ± 0.43	2.92 ± 1.00	1.34 ± 0.27	0.89 ± 0.23	1.60 ± 0.49
GC	193	5.02 ± 0.87	1.14 (0.78)	1.72 ± 0.41	2.90 ± 0.75	1.36 ± 0.27	0.88 ± 0.19	1.63 ± 0.51
CC	98	4.95 ± 0.92	0.98 (0.70)	1.97 ± 0.98	2.74 ± 0.71	1.41 ± 0.27	0.82 ± 0.21	1.83 ± 0.60
F		2.479	4.038	11.435	0.462	3.652	1.263	5.272
P		0.084	0.018	0.000	0.630	0.026	0.283	0.005

TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA1, apolipoprotein A1; ApoB, apolipoprotein B; ApoA1/ApoB, the ratio of apolipoprotein A1 to apolipoprotein B. The value of TG was presented as median (interquartile range). The difference between the genotypes was determined by the Kruskal-Wallis test.

Table 4. Comparison of the Trp316Ser variant (rs1801690) genotypes near the apolipoprotein H (β2-Glycoprotein-I) gene and serum levels in the males and females of the Mulao and Han populations

Genotype	n	TC (mmol/L)	TG (mmol/L)	HDL-C (mmol/L)	LDL-C (mmol/L)	ApoA1 (g/L)	ApoB (g/L)	ApoA/ApoB
Mulao/Male								
GG	288	5.11 ± 1.24	1.14 (0.83)	1.73 ± 0.44	2.92 ± 0.86	1.18 ± 0.47	1.04 ± 0.65	1.60 ± 1.24
GC	96	4.98 ± 1.76	1.05 (0.67)	1.77 ± 0.65	2.80 ± 0.88	1.35 ± 0.42	1.10 ± 0.76	1.37 ± 0.80
CC	50	4.83 ± 1.24	0.97 (0.68)	1.82 ± 0.69	2.56 ± 1.09	1.39 ± 0.55	0.99 ± 0.63	1.71 ± 0.93
F		3.079	4.297	0.140	4.171	4.881	0.623	1.679
P		0.047	0.014	0.869	0.016	0.028	0.537	0.188
Mulao/Female								
GG	258	5.05 ± 1.19	1.02 (0.79)	1.76 ± 0.40	3.04 ± 0.93	1.34 ± 0.36	0.96 ± 0.51	1.71 ± 1.36
GC	122	5.07 ± 1.09	0.97 (0.76)	1.90 ± 0.46	2.96 ± 0.86	1.23 ± 0.50	1.05 ± 0.67	1.48 ± 0.91
CC	65	4.78 ± 1.67	0.88 (0.61)	1.85 ± 0.61	2.84 ± 1.08	1.13 ± 0.57	1.05 ± 0.71	1.42 ± 0.99
F		2.614	1.833	3.002	2.987	6.627	1.158	1.832
P		0.074	0.161	0.051	0.051	0.001	0.315	0.161
Han/Male								
GG	283	5.46 ± 1.42	1.32 (0.94)	1.64 ± 0.42	2.96 ± 1.05	1.38 ± 0.29	0.95 ± 0.23	1.53 ± 0.48
GC	96	5.08 ± 0.82	1.20 (0.78)	1.73 ± 0.42	2.90 ± 0.69	1.40 ± 0.29	0.90 ± 0.18	1.61 ± 0.52
CC	38	5.09 ± 0.76	1.02 (0.79)	1.80 ± 0.50	2.85 ± 0.62	1.43 ± 0.28	0.87 ± 0.20	1.75 ± 0.60
F		5.485	4.669	3.061	0.541	0.490	4.130	3.211
P		0.004	0.010	0.048	0.582	0.613	0.017	0.041
Han/Female								
GG	270	4.97 ± 1.27	1.05 (0.71)	1.71 ± 0.40	2.88 ± 0.95	1.31 ± 0.26	0.83 ± 0.21	1.88 ± 0.61
GC	97	4.96 ± 0.91	0.98 (0.78)	1.73 ± 0.44	2.91 ± 0.82	1.33 ± 0.23	0.86 ± 0.19	1.67 ± 0.49
CC	60	4.87 ± 1.00	0.87 (0.64)	2.07 ± 1.19	2.67 ± 0.76	1.41 ± 0.26	0.80 ± 0.21	1.64 ± 0.51
F		0.199	0.228	9.489	0.409	5.082	1.164	3.087
P		0.819	0.796	0.000	0.665	0.007	0.313	0.047

TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA1, apolipoprotein A1; ApoB, apolipoprotein B; ApoA1/ApoB, the ratio of apolipoprotein A1 to apolipoprotein B. The values of triglyceride were presented as median (interquartile range), and the difference among the genotypes was determined by the Kruskal-Wallis test.

are the major determinants of the familial resemblance in plasma lipids and lipoprotein.

Mulao people have been abided with their special customs especially in case of marriage. Their marriages were family-arranged since childhood. However, divorce and remarriage were also allowed. Although the marriage was arranged at childhood, it was celebrated only when the girl reached puberty. Traditionally, until before a Mulao girl delivered her first child, she stayed with her parents and was unrestricted to join the young people social activities such as responsive singing, flirtations, and

Table 5. Relationship between serum lipid parameters and relative factors in the Mulao and Han populations

Lipid parameter	Relative factor	*B*	*Std. error*	*Beta*	*t*	*P*
Mulao and Han						
TC	Genotype	-0.126	0.042	-0.071	-3.016	0.003
	Age	0.011	0.003	0.128	4.101	0.000
	Cigarette smoking	0.111	0.047	0.068	2.338	0.019
	Waist circumference	0.016	0.006	0.108	2.759	0.006
	Diastolic blood pressure	0.007	0.003	0.064	2.575	0.010
	Glucose	0.046	0.017	0.068	2.800	0.005
TG	Ethnic group	0.484	0.137	0.083	3.533	0.000
	Cigarette smoking	0.436	0.109	0.115	3.983	0.000
	Body mass index	-0.280	0.135	-0.312	-2.069	0.039
	Waist circumference	0.065	0.014	0.187	4.773	0.000
	Diastolic blood pressure	0.013	0.006	0.050	2.037	0.042
	Pulse pressure	0.011	0.005	0.059	2.304	0.021
	Glucose	0.150	0.038	0.094	3.899	0.000
HDL-C	Genotype	0.077	0.017	0.107	4.759	0.000
	Gender	0.095	0.036	0.093	2.676	0.008
	Age	0.003	0.001	0.083	2.696	0.007
	Alcohol consumption	0.089	0.019	0.129	4.627	0.000
	Height	0.020	0.008	0.314	2.636	0.008
	Weight	-0.031	0.010	-0.576	-3.016	0.003
	Body mass index	0.050	0.024	0.318	2.112	0.035
	Waist circumference	-0.006	0.002	-0.091	-2.333	0.020
LDL-C	Genotype	-0.102	0.031	-0.078	-3.294	0.001
	Age	0.008	0.002	0.123	3.917	0.000
	Cigarette smoking	-0.093	0.035	-0.078	-2.657	0.008
ApoA1	Ethnic group	0.051	0.018	0.068	2.857	0.004
	Alcohol consumption	0.095	0.014	0.188	6.646	0.000
	Pulse pressure	0.003	0.001	0.113	4.342	0.000
ApoB	Ethnic group	-0.152	0.023	-0.157	-6.695	0.000
	Waist circumference	0.010	0.002	0.173	4.438	0.000
	Pulse pressure	0.004	0.001	0.119	4.616	0.000
	Glucose	0.019	0.006	0.073	3.000	0.003
ApoA1/ApoB	Age	-0.005	0.002	-0.074	-2.328	0.020
	Alcohol consumption	0.079	0.035	0.064	2.254	0.024
	Waist circumference	-0.013	0.004	-0.115	-2.884	0.004
Mulao						
TC	Genotype	-0.177	0.060	-0.099	-2.944	0.003
	Age	0.010	0.003	0.127	2.978	0.003
	Cigarette smoking	0.141	0.070	0.080	2.015	0.044
TG	Waist circumference	0.052	0.014	0.204	3.794	0.000
HDL-C	Age	0003	0.001	0.086	2.000	0.046
	Alcohol consumption	0.098	0.027	0.139	3.603	0.000
LDL-C	Genotype	-0.164	0.043	-0.128	-3.800	0.000
	Age	0.008	0.003	0.135	3.108	0.002
	Alcohol consumption	-0.106	0.051	-0.080	-2.052	0.040
ApoA1	Genotype	-0.079	0.021	-0.126	-3.773	0.000
	Alcohol consumption	0.138	0.025	0.216	5.578	0.000

	Waist circumference	-0.008	0.003	-0.159	-2.994	0.003
	Pulse pressure	0.004	0.001	0.138	3.770	0.000
ApoB	Waist circumference	-0.011	0.004	0.154	2.860	0.004
	Diastolic blood pressure	-0.004	0.002	-0.072	-2.056	0.040
	Pulse pressure	0.006	0.001	-.149	4.038	0.000
ApoA1/ApoB	Waist circumference	0.004	0.004	0.037	1.047	0.295
Han						
TC	Genotype	-0.177	0.060	-0.099	-2.944	0.003
	Age	0.013	0.004	0.152	3.481	0.001
	Cigarette smoking	0.141	0.070	0.080	2.015	0.044
TG	Genotype	-0.446	0.167	-0.089	-2.664	0.008
	Cigarette smoking	0.748	0.184	0.173	4.072	0.000
	Alcohol consumption	-0.447	0.182	0.100	-2.459	0.014
	Waist circumference	0.079	0.026	0.181	3.070	0.002
	Diastolic blood pressure	0.025	0.012	0.077	2.185	0.029
	Glucose	0.274	0.061	0.156	4.493	0.000
HDL-C	Genotype	0.111	0.026	0.146	4.328	0.000
	Gender	0.129	0.059	0.122	2.175	0.030
	Age	0.004	0.002	0.104	2.248	0.025
	Alcohol consumption	0.080	0.028	0.118	2.882	0.004
	Height	0.028	0.011	0.417	2.609	0.009
	Weight	-0.041	0.014	-0.721	2.993	0.003
	Body mass index	0.074	0.030	0.466	2.488	0.013
LDL-C	Gender	-0.237	0.103	-0.129	-2.287	0.022
	Age	0.008	0.003	0.120	2.575	0.010
	Cigarette smoking	-0.241	0.049	-0.211	-4.895	0.000
	Glucose	0.035	0.016	0.076	2.165	0.031
ApoA1	Genotype	0.034	0.013	0.087	2.618	0.009
	Cigarette smoking	0.049	0.014	0.144	3.406	0.001
	Alcohol consumption	0.067	0.014	0.193	4.762	0.000
	Height	0.016	0.005	0.478	3.033	0.003
	Weight	-0.026	0.007	-0.875	-3.684	0.000
	Body mass index	0.035	0.015	0.430	2.324	0.020
	Waist circumference	0.004	0.002	0.131	2.223	0.027
	Pulse pressure	0.002	0.001	0.084	2.256	0.024
ApoB	Gender	-0.068	0.023	-0.154	-2.928	0.004
	Waist circumference	0.007	0.002	0.252	4.503	0.000
	Diastolic blood pressure	0.003	0.001	0.123	3.689	0.000
	Glucose	0.018	0.004	0.163	4.979	0.000
ApoA1/ApoB	Genotype	0.073	0.024	0.098	3.045	0.002
	Gender	0.226	0.055	0.219	4.082	0.000
	Cigarette smoking	0.089	0.026	0.139	3.388	0.001
	Alcohol consumption	0.057	0.026	0.086	2.190	0.029
	Height	0.028	0.010	0.427	2.788	0.005
	Weight	-0.036	0.013	-0.638	-2.766	0.006
	Diastolic blood pressure	-0.004	0.002	-0.074	-2.169	0.030

TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA1, apolipoprotein A1; ApoB, apolipoprotein B; ApoA1/ApoB, the ratio of apolipoprotein A1 to apolipoprotein B.

courtships at festival times. The age of the wife was usually four or five years older than that of the husband. Engagement and marriage were socially accepted with bride-wealth payment.

Table 6. Relationship between serum lipid parameters and relative factors in the males and females of the Mulao and Han populations

Lipid parameter	Relative factor	*B*	*Std. error*	*Beta*	*t*	*P*
Mulao/male						
TC	Genotype	-0.183	0.093	-0.096	-1.975	0.049
	Age	0.014	0.005	0.165	2.830	0.005
TG	Waist circumference	0.051	0.023	0.179	2.275	0.023
HDL-C	Age	0.004	0.002	0.124	2.184	0.029
	Alcohol consumption	0.109	0.030	0.176	3.599	0.000
LDL-C	Genotype	-0.182	0.064	-0.141	-2.858	0.004
	Age	0.009	0.004	0.148	2.525	0.012
ApoA1	Alcohol consumption	0.136	0.026	0.254	5.201	0.000
ApoB	Pulse pressure	0.007	0.002	0.162	3.017	0.003
ApoA1/ApoB	Waist circumference	-0.015	0.006	-0.119	-2.484	0.013
Mulao/female						
TC	Genotype	-0.157	0.080	-0.093	-1.975	0.049
	Age	0.018	0.004	0.212	4.515	0.000
	Body mass index	0.044	0.018	0.113	2.447	0.015
TG	Alcohol consumption	1.976	0.455	0.200	4.348	0.000
	Waist circumference	0.035	0.010	0.157	3.408	0.001
HDL-C	Genotype	0.062	0.029	0.099	2.158	0.032
	Weight	-0.013	0.003	-0.245	-5.343	0.000
LDL-C	Genotype	-0.139	0.059	-0.109	-2.359	0.019
	Age	0.011	0.003	0.163	3.229	0.001
	Body mass index	0.054	0.013	0.187	4.103	0.000
	Pulse pressure	0.006	0.003	0.104	2.072	0.039
ApoA1	Genotype	-0.115	0.028	-0.191	-4.155	0.000
	Waist circumference	-0.009	0.003	-0.163	-3.529	0.000
	Pulse pressure	0.004	0.001	0.151	3.239	0.001
ApoB	Waist circumference	0.014	0.003	0.191	4.144	0.000
	Cigarette smoking	0.624	0.255	0.112	2.453	0.015
	Pulse pressure	0.006	0.002	0.167	3.618	0.000
ApoA1/ApoB	Age	-0.011	0.004	-0.126	-2.721	0.007
	Waist circumference	-0.029	0.007	-0.186	-4.015	0.000
Han/male						
TC	Genotype	-0.225	0.089	-0.116	-2.528	0.012
	Diastolic blood pressure	0.029	0.005	0.251	5.404	0.000
	Glucose	0.101	0.032	0.147	3.192	0.002
TG	Genotype	-0.752	0.300	-0.114	-2.507	0.013
	Cigarette smoking	0.823	0.227	0.169	3.630	0.000
	Alcohol consumption	-0.575	0.226	-0.123	-2.550	0.011
	Waist circumference	0.108	0.025	0.204	4.303	0.000
	Glucose	0.428	0.110	0.183	3.874	0.000
HDL-C	Genotype	0.076	0.030	0.114	2.501	0.013
	Alcohol consumption	0.082	0.023	0.173	3.628	0.000
	Weight	-0.017	0.002	-0.352	-7.378	0.000
LDL-C	Cigarette smoking	-0.258	0.050	-0.241	-5.119	0.000
	Systolic blood pressure	0.005	0.002	0.103	2.149	0.032
	Body mass index	0.029	0.012	0.111	2.321	0.021
ApoA1	Cigarette smoking	0.052	0.015	0.160	3.418	0.001

	Alcohol consumption	0.066	0.015	0.210	4.301	0.000
	Weight	-0.008	0.002	-0.248	-5.130	0.000
ApoB	Genotype	-0.039	0.015	-0.116	2.659	0.008
	Waist circumference	0.007	0.001	0.241	5.273	0.000
	Diastolic blood pressure	0.004	0.001	0.210	4.591	0.000
	Glucose	0.024	0.005	0.199	4.441	0.000
ApoA1/ApoB	Genotype	0.091	0.034	0.118	2.661	0.008
	Cigarette smoking	0.091	0.026	0.159	3.532	0.000
	Alcohol consumption	0.071	0.026	0.128	2.743	0.006
	Body mass index	-0.030	0.008	-0.212	-3.683	0.000
	Waist circumference	-0.012	0.003	-0.188	-3.380	0.001
Han/female						
TC	Age	0.029	0.004	0.332	6.827	0.000
	Body mass index	0.044	0.018	0.112	2.464	0.014
	Glucose	0.073	0.026	0.134	2.839	0.005
TG	Glucose	0.147	0.054	0.132	2.756	0.006
HDL-C	Genotype	0.152	0.040	0.183	3.799	0.000
	Age	0.007	0.003	0.148	2.718	0.007
	Diastolic blood pressure	-0.006	0.003	-0.110	-2.160	0.031
LDL-C	Age	0.020	0.003	0.291	5.919	0.000
	Waist circumference	0.019	0.006	0.158	3.447	0.001
	Glucose	0.042	0.020	0.100	2.093	0.037
ApoA1	Genotype	0.050	0.017	0.145	3.024	0.003
	Age	0.002	0.001	0.129	2.304	0.022
	Cigarette smoking	0.124	0.055	0.107	2.240	0.026
	Body mass index	-0.010	0.004	-0.114	-2.387	0.017
	Systolic blood pressure	-0.004	0.001	-0.248	-2.829	0.005
	Pulse pressure	0.006	0.002	0.302	3.546	0.000
ApoB	Age	0.004	0.001	0.232	5.077	0.000
	Waist circumference	0.007	0.001	0.236	5.208	0.000
	Glucose	0.013	0.005	0.134	2.868	0.004
ApoA1/ApoB	Age	-0.006	0.002	-0.162	-3.198	0.001
	Height	0.010	0.004	0.105	2.134	0.033
	Cigarette smoking	0.242	0.114	0.102	2.115	0.035
	Waist circumference	-0.014	0.003	-0.203	-4.314	0.000

TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; ApoA1, apolipoprotein A1; ApoB, apolipoprotein B; ApoA1/ApoB, the ratio of apolipoprotein A1 to apolipoprotein B.

Interestingly, there was a preference of marriage to cousins from maternal side (mother's brother's daughter). Therefore, it is compelling us to believe that some hereditary characteristics and genotypes of lipid metabolism-related genes in this population may be different from those in Han nationality [32-35]. In additions, different environmental and genetic factors might also contribute to variable levels of association with serum lipid levels in different populations.

In the present study, we showed that the levels of TC, LDL-C and ApoA1 in Mulao, and TG, HDL-C, ApoA1 and the ratio of ApoA1 to ApoB in Han were different among the genotypes of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene ($P < 0.05$-0.01). Subgroup analyses according to the sex showed that the levels of TC, TG, LDL-C, and ApoA1 in Mulao males, ApoA1 in Mulao females; TC, TG, HDL-C and ApoB and the ratio of ApoA1 to ApoB in Han males, and HDL-C and ApoA1, and the ratio of ApoA1 to ApoB in Han females were associated with the genotypes of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene.

Serum lipid parameters were also associated with several environmental factors.

It is well known that environmental factors such as dietary patterns, lifestyle, obesity, physical activity, and hypertension are all strongly related with serum lipid levels [40]. In the present study, we also showed that serum lipid parameters were correlated with age, sex, alcohol consumption, cigarette smoking, BMI, fasting blood glucose levels, and blood pressure in both ethnic groups. These data suggested that the environmental factors also played an important role in determining serum lipid levels in our populations. Although rice and corn are the staple foods in both ethnic groups, Mulao people prefer to eat cold foods along with acidic and spicy dishes, so bean soy sauce and pickled vegetables are among their most popular dishes. They also favor to eat animal offals which contain abundant saturated fatty acid. It has been widely accepted that high-fat diets, particularly those containing large quantities of saturated fatty acids raise blood cholesterol concentrations and predispose individuals to CAD [41]. Many studies also stated that diatery habits can impart a strong influence on serum levels of ApoB, and which in turn can effects the risk of CAD [42-44]. In the current study, we found that the level of ApoB was higher in Mulao than in Han. This might be partly due to the difference in dietary habit between the Mulao and Han populations.

The Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-Glycoprotein-I) gene explain a small proportion of the variance in ApoH expression, thus the ability of the SNP to influence plasma β2GPI levels may be obscured by the strong effects of other factors (undefined elements which are in strong LD with the SNP and other regulatory factors that affect in vivo gene expression) in aggregate. However, given the reporter gene expression data indicating possible binding to transcription factors, there is clearly a functional effect of the polymorphism on *ApoH* regulation that are worthy of further investigation. However, haplotype analysis including *ApoH* promoter SNPs alone or in conjunction with previously known coding SNPs affecting plasma β2GPI levels (Trp316Ser) gave us no new insights into determining the genetic basis of plasma β2GPI levels. The significant haplotypes were defined predominantly by the minor alleles at the coding SNPs, which are already known to have a major effect on β2GPI levels. Therefore, given both the method and also the small sample size, further studies are warranted in larger cohorts using improvised methods that will help better to delineate the goals of this study that to assess the association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and several environmental factors with serum lipid profiles in the Mulao and Han populations.

In addition, it is reported that some of the identified genes are plausible biological candidates, some SNP in the *ApoH* showed suggestive association with the β2-GPI phenotype, as they are actually or potentially involved in inflammatory processes or some unknown processes [45, 46]. Our results represent a first step towards identifying common variants reflecting the genetic architecture influencing plasma β2-GPI levels, and warrant further validation by functional experiments, as the functions of some of the discovered loci are still unknown.

Although we observe significant association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and serum lipid levels, the unmeasured interactions of gene-gene, gene-environment, and environment-environment on serum lipid levels were remained to be determined. Furthermore, the impact of sex hormones was not evaluated in both males and females of the two ethnic groups respectively due to the relatively small samples. Hence, we must recognize the limited power to provide an understanding of full impact of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene on lipid metabolism since there are none of published documents involved in this aspect directly. The association of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene and serum lipid levels should be detected in further investigations.

In conclusion, the C allele frequency of the Trp316Ser variant (rs1801690) near the apolipoprotein H (β2-glycoprotein-I) gene in both Mulao and Han is higher in females than in males. The minor C allele carriers in both ethnic groups have more favorable serum lipid profiles than the C allele non-carriers. These findings suggest that the association between the Trp316Ser variant (rs1801690) near the apoli-

poprotein H (β2-glycoprotein-I) gene and serum lipid levels might have ethnic-and/or sex-specificity.

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Disclosure of conflict of interest

None.

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References

[1] Wu L, Parhofer KG. Diabetic dyslipidemia. Metabolism 2014; S0026-0495: 00258-00263.

[2] Evans A, Van Baal GC, McCarron P, DeLange M, Soerensen TI, De Geus EJ, Kyvik K, Pedersen NL, Spector TD, Andrew T, Patterson C, Whitfield JB, Zhu G, Martin NG, Kaprio J, Boomsma DI. The genetics of coronary heart disease: the contribution of twin studies. Twin Res 2003; 6: 432-441.

[3] Draisma HH, Reijmers TH, Meulman JJ, van der Greef J, Hankemeier T, Boomsma DI. Hierarchical clustering analysis of blood plasma lipidomics profiles from mono- and dizygotic twin families. Eur J Hum Genet 2013; 21: 95-101.

[4] Roberts R. Personalized medicine: a reality within this decade. J Cardiovasc Transl Res 2008; 1: 11-16.

[5] Galli M, Comfurius P, Maassen C, Hemker HC, de Baets MH, van Breda-Vriesman PJ, Barbui T, Zwaal RF, Bevers EM. Anticardiolipin antibodies (ACA) directed not to cardiolipin but to a plasma protein cofactor. Lancet 1990; 335: 1544-1547.

[6] McNeil HP, Simpson RJ, Chesterman CN, Krilis SA. Anti-phospholipid antibodies are directed against a complex antigen that includes a lipid-binding inhibitor of coagulation: beta 2-glycoprotein I (apolipoprotein H). Proc Natl Acad Sci U S A 1990; 87: 4120-4124.

[7] Tsunoda K, Harihara S, Dashnyam B, Semjidmaa D, Yamaguchi Y, Tanabe Y, Sakai N, Sato A, Sato K. 2002 Apolipoprotein E and H polymorphisms in Mongolian Buryat: allele frequencies and relationship with plasma lipid levels. Hum Biol 2002; 74: 659-71.

[8] Chen Q, Reis SE, Kammerer C, Craig W, McNamara DM, Holubkov R, Sharaf BL, Sopko G, Pauly DF, Merz CN, Kamboh MI; WISE study group. Association of anti-oxidized LDL and candidate genes with severity of coronary stenosis in the Women's Ischemia Syndrome Evaluation study. J Lipid Res 2011; 52: 801-7.

[9] Xia J, Yang QD, Yang QM, Xu HW, Liu YH, Zhang L, Zhou YH, Wu ZG, Cao GF. Apolipoprotein H gene polymorphisms and risk of primary cerebral hemorrhage in a Chinese population. Cerebrovasc Dis 2004; 17: 197-203.

[10] Okkels H, Rasmussen TE, Sanghera DK, Kamboh MI, Kristensen T. Structure of the human beta2-glycoprotein I (apolipoprotein H) gene. Eur J Biochem 1999; 259: 435-440.

[11] Sheng Y, Sali A, Herzog H, Lahnstein J, Krilis SA. Site-directed mutagenesis of recombinant human beta 2-glycoprotein I identifies a cluster of lysine residues that are critical for phospholipid binding and anti-cardiolipin antibody activity. J Immunol 1996; 157: 3744-3751.

[12] Sanghera DK, Wagenknecht DR, McIntyre JA, Kamboh MI. Identification of structural mutations in the fifth domain of apolipoprotein H (beta 2-glycoprotein I) which affect phospholipid binding. Hum Mol Genet 1997; 6: 311-316.

[13] Mehdi H, Naqvi A, Kamboh MI. A hydrophobic sequence at position 313-316 (Leu-Ala-Phe-Trp) in the fifth domain of apolipoprotein H (beta2-glycoprotein I) is crucial for cardiolipin binding. Eur J Biochem 2000; 267: 1770-1776.

[14] Wang HH, Chiang AN. Cloning and characterization of the human beta2-glycoprotein I (beta2-GPI) gene promoter: roles of the atypical TATA box and hepatic nuclear factor-1alpha in regulating beta2-GPI promoter activity. Biochem J 2004; 380: 455-463.

[15] Averna M, Paravizzini G, Marino G, Lanteri E, Cavera G, Barbagallo CM, Petralia S, Cavallaro S, Magro G, Grasso S, Notarbartolo A, Travali S. Liver is not the unique site of synthesis of beta 2-glycoprotein I (apolipoprotein H): evidence for an intestinal localization. Int J Clin Lab Res 1997; 27: 207-212.

[16] Polz E, Kostner GM. The binding of beta 2-glycoprotein-I to human serum lipoproteins: distribution among density fractions. FEBS Lett 1979; 102: 183-186.

[17] Gambino R, Ruiu G, Pagano G, Cassader M. The binding of apolipoprotein H (beta2-glycoprotein I) to lipoproteins. Prostaglandins Other Lipid Mediat 1999; 57: 351-359.

[18] Ağar C, de Groot PG, Levels JH, Marquart JA, Meijers JC. Beta2-glycoprotein I is incorrectly named apolipoprotein H. J Thromb Haemost 2009; 7: 235-236.

[19] Kamboh MI, Manzi S, Mehdi H, Fitzgerald S, Sanghera DK, Kuller LH, Atson CE. Genetic variation in apolipoprotein H (beta2-glycoprotein I) affects the occurrence of antiphospholipid antibodies and apolipoprotein H concentrations in systemic lupus erythematosus. Lupus 1999; 8: 742-750.

[20] Rahgozar S, Giannakopoulos B, Yan X, Wei J, Cheng Qi J, Gemmell R, Krilis SA. Beta2-glycoprotein I protects thrombin from inhibition by heparin cofactor II: potentiation of this effect in the presence of anti-beta2-glycoprotein I autoantibodies. Arthritis Rheum 2008; 58: 1146-1155.

[21] Chen Q, Kamboh MI. Complete DNA sequence variation in the apolipoprotein H (beta-glycoprotein I) gene and identification of informative SNPs. Ann Hum Genet 2006; 70: 1-11.

[22] Kamboh MI, Wang X, Kao AH, Barmada MM, Clarke A, Ramsey-Goldman R, Manzi S, Demirci FY. Genome-Wide Association Study of Antiphospholipid Antibodies. Autoimmune Dis 2013; 2013: 761046.

[23] Lin KY, Pan JP, Yang DL, Huang KT, Chang MS, Ding PY, Chiang AN. Evidence for inhibition of low density lipoprotein oxidation and cholesterol accumulation by apolipoprotein H (b2- glycoprotein I). Life Sci 2001; 69: 707-719.

[24] Nakaya Y, Schaefer EJ, Brewer HB Jr. Activation of human post heparin lipoprotein lipase by apolipoprotein H (beta 2-glycoprotein I). Biochem Biophys Res Commun 1980; 95: 1168-1172.

[25] Mehdi H, Aston CE, Sanghera DK, Hamman RF, Kamboh MI. Genetic variation in the apolipoprotein H (beta2-glycoprotein I) gene affects plasma apolipoprotein H concentrations. Hum Genet 1999; 105: 63-71.

[26] Sepehrnia B, Kamboh MI, Adams-Campbell L, Bunker CH, Nwankwo M, Majumder P, Ferrell RE. Genetic studies of human apolipoproteins, VIII: role of the apolipoprotein H polymorphism in relation to serum lipoprotein concentrations. Hum Genet 1989; 82: 118-122.

[27] Cassader M, Ruiu G, Gambino R, Guzzon F, Pagano A, Veglia R, Pagano G. Influence of apolipoprotein H polymorphism on levels of triglycerides. Atherosclerosis 1994; 110: 45-51.

[28] Asselbergs FW, Guo Y, van Iperen EP, Sivapalaratnam S, Tragante V, Lanktree MB, Lange LA, Almoguera B, Appelman YE, Barnard J, Baumert J, Beitelshees AL, Bhangale TR, Chen YD, Gaunt TR, Gong Y, Hopewell JC, Johnson T, Kleber ME, Langaee TY, Li M, Li YR, Liu K, McDonough CW, Meijs MF, Middelberg RP, Musunuru K, Nelson CP, O'Connell JR, Padmanabhan S, Pankow JS, Pankratz N, Rafelt S, Rajagopalan R, Romaine SP, Schork NJ, Shaffer J, Shen H, Smith EN, Tischfield SE, van der Most PJ, van Vliet-Ostaptchouk JV, Verweij N, Volcik KA, Zhang L, Bailey KR, Bailey KM, Bauer F, Boer JM, Braund PS, Burt A, Burton PR, Buxbaum SG, Chen W, Cooper-Dehoff RM, Cupples LA, deJong JS, Delles C, Duggan D, Fornage M, Furlong CE, Glazer N, Gums JG, Hastie C, Holmes MV, Illig T, Kirkland SA, Kivimaki M, Klein R, Klein BE, Kooperberg C, Kottke-Marchant K, Kumari M, LaCroix AZ, Mallela L, Murugesan G, Ordovas J, Ouwehand WH, Post WS, Saxena R, Scharnagl H, Schreiner PJ, Shah T, Shields DC, Shimbo D, Srinivasan SR, Stolk RP, Swerdlow DI, Taylor HA Jr, Topol EJ, Toskala E, van Pelt JL, van Setten J, Yusuf S, Whittaker JC, Zwinderman AH; LifeLines Cohort Study, Anand SS, Balmforth AJ, Berenson GS, Bezzina CR, Boehm BO, Boerwinkle E, Casas JP, Caulfield MJ, Clarke R, Connell JM, Cruickshanks KJ, Davidson KW, Day IN, de Bakker PI, Doevendans PA, Dominiczak AF, Hall AS, Hartman CA, Hengstenberg C, Hillege HL, Hofker MH, Humphries SE, Jarvik GP, Johnson JA, Kaess BM, Kathiresan S, Koenig W, Lawlor DA, März W, Melander O, Mitchell BD, Montgomery GW, Munroe PB, Murray SS, Newhouse SJ, Onland-Moret NC, Poulter N, Psaty B, Redline S, Rich SS, Rotter JI, Schunkert H, Sever P, Shuldiner AR, Silverstein RL, Stanton A, Thorand B, Trip MD, Tsai MY, van der Harst P, van der Schoot E, van der Schouw YT, Verschuren WM, Watkins H, Wilde AA, Wolffenbuttel BH, Whitfield JB, Hovingh GK, Ballantyne CM, Wijmenga C, Reilly MP, Martin NG, Wilson JG, Rader DJ, Samani NJ, Reiner AP, Hegele RA, Kastelein JJ, Hingorani AD, Talmud PJ, Hakonarson H, Elbers CC, Keating BJ, Drenos F. Large-scale gene-centric meta-analysis across 32 studies identifies multiple lipid loci. Am J Hum Genet 2012; 91: 823-838.

[29] Xu L, Deng QY, Li SF, Zhou LN, Gong JC, Wei BY. Genetic analysis of Mulao nationality using 15 short tandem repeats. Zhonghua Yi Xue Yi Chuan Xue Za Zhi 2008; 25: 96-100.

[30] Cullen P. Evidence that triglycerides are an independent coronary heart disease risk factor. Am J Cardiol 2000; 86: 943-949.

[31] Meagher EA. Addressing cardiovascular disease in women: focus on dyslipidemia. J Am Board Fam Pract 2004; 17: 424-437.

[32] Guo T, Yin RX, Chen X, Bin Y, Nie RJ, Li H. Sex-specific association of the SPTY2D1 rs7934205 polymorphism and serum lipid levels. Int J Clin Exp Pathol 2015; 8: 665-81.

[33] Guo T, Yin RX, Lin QZ, Wu J, Shen SW, Sun JQ, Shi GY, Wu JZ, Li H, Wang YM. Polymorphism of rs873308 near the transmembrane protein 57 gene is associated with serum lipid levels. Biosci Rep 2014; 34: e00095.

[34] Lin QZ, Yin RX, Guo T, Wu J, Sun JQ, Shen SW, Shi GY, Wu JZ, Liu CW, Pan SL. Association of the ST3GAL4 rs11220462 polymorphism and

serum lipid levels in the Mulao and Han populations. Lipids Health Dis 2014; 13: 123.
[35] Aung LH, Yin RX, Wu DF, Wang W, Wu JZ, Liu CW. Sex-specific Association of the Zinc Finger Protein 259 rs2075290 Polymorphism and Serum Lipid Levels. Int J Med Sci 2014; 11: 471-478.
[36] Ruixing Y, Qiming F, Dezhai Y, Shuquan L, Weixiong L, Shangling P, Hai W, Yongzhong Y, Feng H, Shuming Q. Comparison of demography, diet, lifestyle, and serum lipid levels between the Guangxi Bai Ku Yao and Han populations. J Lipid Res 2007; 48: 2673-2681.
[37] Ruixing Y, Dezhai Y, Shuquan L, Yuming C, Hanjun Y, Qiming F, Shangling P, Weixiong L, Jing T, Yiyang, L. Hyperlipidaemia and its risk factors in the Guangxi Bai Ku Yao and Han populations. Public Health Nutr 2009; 12: 816-824.
[38] Aung LH, Yin RX, Wu JZ, Wu DF, Wang W, Li H. Association between the MLX interacting protein-like, BUD13 homolog and zinc finger protein 259 gene polymorphisms and serum lipid levels. Sci Rep 2014; 4: 5565.
[39] Aung LH, Yin RX, Wu DF, Wang W, Liu CW, Pan SL. Association of the variants in the BUD13-ZNF259 genes and the risk of hyperlipidaemia. J Cell Mol Med 2014; 18: 1417-1428.
[40] Ruixing Y, Fengping H, Shangling P, Dezhai Y, Weixiong L, Tangwei L, Yuming C, Jinzhen W, Limei Y, Jiandong H. Prevalence of hyperlipidemia and its risk factors for the middle-aged and elderly in the Guangxi Hei Yi Zhuang and Han populations. J Investig Med 2006; 54: 191-200.
[41] Yu-Poth S, Zhao G, Etherton T, Naglak M, Jonnalagadda S, Kris-Etherton PM. Effects of the National Cholesterol Education Program's Step I and Step II dietary intervention programs on cardiovascular disease risk factors: a meta-analysis. Am J Clin Nutr 1999; 69: 632-46.
[42] Sola R, Fito M, Estruch R, Salas-Salvado J, Corella D, de La Torre R, Munoz MA, Lopez-Sabater Mdel C, Martinez-Gonzalez MA, Aros F. et al. Effect of a traditional Mediterranean diet on apolipoproteins B, A-I, and their ratio: a randomized, controlled trial. Atherosclerosis 2011; 218: 174-180.
[43] Valente EA, Sheehy ME, Avila JJ, Gutierres JA, Delmonico MJ, Lofgren IE. The effect of the addition of resistance training to a dietary education intervention on apolipoproteins and diet quality in overweight and obese older adults. Clin Interv Aging 2011; 6: 235-241.
[44] Dominiczak MH, Caslake MJ. Apolipoproteins: metabolic role and clinical biochemistry applications. Ann Clin Biochem 2011; 48: 498-515.
[45] Athanasiadis G, Sabater-Lleal M, Buil A, Souto JC, Borrell M, Lathrop M, Watkins H, Almasy L, Hamsten A, Soria JM. Genetic determinants of plasma β_2-glycoprotein I levels: a genome-wide association study in extended pedigrees from Spain. J Thromb Haemost 2013; 11: 521-528.
[46] Suresh S, Demirci FY, Lefterov I, Kammerer CM, Ramsey-Goldman R, Manzi S, Kamboh MI. Functional and genetic characterization of the promoter region of apolipoprotein H (beta2-glycoprotein I). FEBS J 2010; 277: 951-963.