The effect of *CYP7B1* polymorphisms on the risk of coronary heart disease in Hainan Han population

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Abstract

Background Coronary heart disease (CHD) is the leading cause of human death worldwide. Genetic factors play an important role in the occurrence of CHD. Our study is designed to investigate the influence of *CYP7B1* polymorphisms on CHD risk.

Methods In this case-control study, 508 CHD patients and 510 healthy individuals were recruited to determine the correlation between *CYP7B1* polymorphisms (rs7836768, rs6472155, and rs2980003) and CHD risk. The associations were evaluated by computing odds ratios (OR) and 95% confidence intervals (CI) with logistic regression analysis. The association between SNP-SNP interaction and CHD susceptibility was carried out by multifactor dimensionality reduction analyses.

Results Our study found that rs6472155 is significantly associated with an increased risk of CHD in age > 60 years (OR 2.20, 95 % CI = 1.07–4.49, $p = 0.031$), women (OR 3.17, 95 % CI = 1.19–8.44, $p = 0.021$), and non-smokers (3.43, 95 % CI = 1.16–10.09, $p = 0.025$). Rs2980003 polymorphism has a lower risk of CHD in drinkers (OR 0.47, 95 % CI = 0.24–0.91, $p = 0.025$). Besides, it was found that rs6472155 polymorphism was associated with uric acid level ($p = 0.034$).

Conclusion Our study indicated that *CYP7B1* polymorphisms are related to the risk of CHD, which provides a new perspective for prevent of CHD.

Introduction

Coronary heart disease (CHD) is a kind of heart disease caused by myocardial ischemia, hypoxia or necrosis caused by structural and functional changes of coronary artery, also known as atherosclerotic heart disease, coronary disease or ischemic heart disease. It is the most common type of cardiovascular disease in the world (1–3). In America, about 620,000 people develop new coronary artery disease every year, and about 295,000 people have recurrent attacks (4). The incidence and mortality of CHD are increasing year by year in China, and the incidence population is also getting younger and younger. Deaths caused by CHD rank the second among deaths caused by other diseases (5). The World Health Organization (WHO) predicts that the number of CHD deaths will increase to 23.3 million by 2030, becoming the leading cause of human death (6). The pathophysiological basis of CHD is atherosclerosis caused by a variety of pathogenic factors, resulting in stenosis or complete occlusion of blood vessels, reduction, or complete interruption of coronary blood flow, and eventually ischemia or necrosis of cardiomyocytes (7). Although the exact pathogenesis of coronary heart disease is not very clear, the most widely accepted view is that coronary heart disease is a polygenetic disease, which is the result of the interaction between multiple genes and environmental factors (8). In addition to the classic risk factors such as age, smoking, drinking, hypertension, diabetes, and hypercholesterolemia (9), a large number of studies have confirmed that genetic factors play an important role in the occurrence and development of CHD (10). Besides, many genetic variants such as *APOB*, *GSTT1*, *CYP11B2*, and *MT2A* can significantly affect the susceptibility of CHD (11–14).

Cytochrome P450 (CYP), a superfamily of cystine-heme enzymes, its central role in the pathogenesis, progression, and prognosis of CHD has been determined (15). *CYP7B1* (Cytochrome P450 Family 7 Subfamily B Member 1) is the subfamily member of CYP enzymes, involved in the metabolism of endogenous hydroxysterols and steroids such as neurosteroids (16, 17). The *CYP7B1* gene mainly in human liver, brain, and reproductive tract (17). 27-hydroxycholesterol (27HC) is a rich cholesterol metabolite, which increases in hypercholesterolemia and is found to be a competitive estrogen receptor antagonist in vascular system in atherosclerotic lesions. Umetani et al found that increasing 27HC levels in mice caused by genetic manipulation (by knockout of the *CYP7B1* gene) could decrease estrogen-dependent vascular nitric oxide synthase expression and inhibited carotid artery endothelialization (18). This finding indicated that

CYP7B1 may play an important role in the occurrence of CHD. It is well accepted that genetics polymorphism can significantly influence the gene expression. However, it is not clear whether *CYP7B1* genetic polymorphism affects the risk of CHD.

Thus, we performed a case-control study to determine the potential role of *CYP7B1* genetic variants in CHD patients. We obtained the SNPs in the *CYP7B1* gene in accordance with 1000 Human Genomic Projects. We further investigated the association between the *CYP7B1* polymorphisms and CHD risk in the Chinese Han population. Stratified analyses were carried out to evaluate the association. We also detect the relationship of SNP-SNP interaction in CHD with clinical indicators among genotypes. This study will give new horizon for the molecular mechanism in the CHD.

Materials And Methods

Study population

In this case-control study, we recruited 1018 unrelated Chinese individuals including 508 patients with CHD and 510 age/gender-matched healthy subjects at Affiliated Haikou Hospital of Xiangya Medical College.

All participants were told the purpose of study and signed written informed consents.

Patients were firstly diagnosed and confirmed to be CHD by experienced cardiologists in accordance with coronary angiography (19). The patients with congenial and rheumatic heart disease, a family history of CHD, a history of any atherosclerotic vascular diseases and other comorbidity such as chronic renal failure, malignancy, and chronic infections should be excluded.

The healthy controls were selected from the CHD-free participants who take a normal physical examination in the same hospital.

And the controls must meet the following inclusion criterion: 1) without a family history of CHD; 2) no diabetes and hypertension; 3) no cardiovascular and cerebrovascular diseases. Basic characteristics included age, gender, smoking status, drinking status, diabetes and hypertension status were acquired by medical records and questionnaire survey.

Our study has been approved by the Medical Ethics Committee of Affiliated Haikou Hospital of Xiangya Medical College.

Experiments in this study were performed following the protocol of Helsinki's Declaration.

Selection And Genotyping For SNP

In our study, rs7836768, rs62519827, rs62519841, rs10808739, rs13276608, rs6472155, and rs2980003 in the *CYP7B1* gene were obtained from 1000 Genomes Project database with a minor allele frequency (MAF) > 5 % for further studying. Genomic DNA from peripheral blood samples of all participants was extracted following the protocol of DNA extraction kit (Xi'an GoldMag Co. Ltd., Xi'an, China).

We further designed the primers for PCR amplification according to Agena Bioscience Assay Design software.

All SNPs were genotyped by an Agena MassARRAY iPLEX platform (Agena Bioscience Inc., CA, USA). Finally, the data of SNP genotyping was management and analyzed by Agena Bioscience TYPER 4.0 software.

Statistical analysis

We performed SPSS version 17.0 software for statistical analyses. The *p*-value with two-tailed test lower than 0.05 means statistically significant. Data distributions are firstly evaluated for normality using the Kolmogorov-Smirnov test. Differences of age and clinical indicators between cases and controls were respectively tested by the student's *t*-test and Mann-whitney test. And the comparisons of gender between the cases and controls was compared by Pearson's χ^2 test.

The Hardy-Weinberg equilibrium (HWE) for SNPs in the controls was evaluated by a Chi-squared test. The χ^2 test or exact test was used to analyze the Allele and genotype distributions of SNPs in the case and control groups. Association of *CYP7B1* polymorphisms with CHD susceptibility was determined by computing ORs and 95 % Cis using logistic regression analysis under five multiple genetic models. Besides, we also detected the associations stratified by age, gender, smoking, and drinking status, and diabetes status. In the end, we explored the relationship of SNP interaction and CHD susceptibility with multifaceted dimensionality reduction (MDR) method. The comparisons between clinical indicators and SNPs were detected by ANOVA test and one-way analysis.

Results

Study subjects

Our study included in 508 CHD patients and 510 healthy individuals. The basic information of the cases and controls were shown in Table 1. The average age was 62.17 ± 10.34 years in the cases and 61.12 ± 9.02 years in the controls. There was no significant differences in age, gender, and urea between the cases and controls ($p = 0.084$, $p = 0.964$, $p = 0.759$, respectively). There were significant comparisons in creatinine, uric acid, total-cholesterol, and apolipoprotein AI between the cases and controls (all $p < 0.05$).

Table 1
Basic characteristics of CHD patients and controls

Characteristics	Cases (n = 508)	Controls (n = 510)	<i>p</i>
Age, years (mean ± SD) ^a	62.17 ± 10.34	61.12 ± 9.02	0.084
> 60	282 (55.5%)	284 (55.7%)	
≤ 60	226 (45.5%)	226 (44.3%)	
Gender ^b			0.964
Male	334 (65.7%)	336 (65.9%)	
Female	174 (34.3%)	174 (34.1%)	
Urea (mmol/l) ^c	462.01 ± 4.50	456.63 ± 2.67	0.759
Creatinine (umol/l) ^c	384.73 ± 3.40	456.07 ± 3.50	< 0.001
Uric acid (umol/l) ^c	431.26 ± 1.08	489.10 ± 2.07	< 0.001
Total-cholesterol (mmol/l) ^c	369.60 ± 1.00	548.04 ± 1.05	< 0.001
Apolipoprotein AI (g/l) ^c	247.13 ± 5.74	351.08 ± 0.70	< 0.001
Smoking status			
Smoker	231 (45.5%)	115 (22.5%)	
Nonsmoker	186 (36.6%)	167 (32.7%)	
Missing	91 (17.9%)	228 (44.8%)	
Drinking status			
Drinking	52 (10.2%)	124 (24.3%)	
Nondrinking	306 (60.2%)	135 (26.5%)	
Missing	150 (29.6%)	251 (49.2%)	
Diabetes	190 (37.4%)		
Non diabetes	318 (62.6%)		
Hypertension	362 (71.3%)		
Normal tension	146 (28.7%)		
<i>p</i> ^a value was calculated by Student's <i>t</i> -test. <i>p</i> ^b value was calculated by Pearson's χ^2 test.			
<i>p</i> ^c Mann-whitney test is used.			

Association analyses between *CYP7B1* polymorphisms and CHD risk

A total of seven SNPs (rs7836768, rs62519827, rs62519841, rs10808739, rs13276608, rs6472155, rs2980003) in the *CYP7B1* gene were successfully detected in our study. The information of each SNP was presented in Table 2. The MAF for rs62519827, rs62519841, rs10808739, and rs13276608 were lower than 0.05, these polymorphisms were deleted in

the current study. SNPs including rs7836768, rs6472155, and rs2980003 in the control followed HWE ($p > 0.05$). We investigated the relationship of *CYP7B1* genetic variants and the risk of CHD under five genetic models, and our result showed that there are no significant associations (Table 3).

Table 2
Allele frequencies among *CYP7B1* SNPs

SNP ID	Chromosome position	Alleles (minor/major)	MAF		O (HET)	E (HET)	p -HWE
			Case	Control			
rs7836768	chr8: 64474910	G/A	0.413	0.396	0.514	0.478	0.115
rs62519827	chr8: 64569390	C/T	0.003	0.005	0.010	0.010	1.000
rs62519841	chr8: 64588948	A/G	0.005	0.005	0.010	0.010	1.000
rs10808739	chr8: 64727703	A/G	0.029	0.042	0.084	0.081	1.000
rs13276608	chr8: 64769294	T/C	0.001	0.003	0.006	0.006	1.000
rs6472155	chr8: 64817650	G/A	0.286	0.264	0.422	0.388	0.067
rs2980003	chr8: 65087728	T/C	0.357	0.346	0.452	0.452	1.000
SNP, Single nucleotide polymorphisms; MAF, Minor allele frequency; HWE, Hardy–Weinberg equilibrium; O (HET), Observed heterozygosity; E (HET), Expected heterozygosity; OR, Odds ratio; 95 % CI, 95% confidence intervals.							
p^a values were calculated by exact test.							

Table 3
Association of *CYP7B1* polymorphisms with CHD risk

SNP ID	Model	Allele/Genotype	Case N	Control N	OR (95% CI)	<i>p</i>
rs7836768	Allele	A	595	616	1	
		G	419	404	1.07 (0.90–1.28)	0.431
	Codominant	AG	239	71	0.90 (0.69–1.19)	0.464
		GG	90	262	1.27 (0.87–1.84)	0.220
		AA	178	177	1	
		AG-GG	329	333	0.98 (0.76–1.27)	0.878
	Recessive	AA-AG	417	248	1	
		GG	90	262	1.34 (0.96–1.89)	0.090
	Log-additive	–	–	–	1.08 (0.90–1.29)	0.427
rs6472155	Allele	A	725	751	1	
		G	291	269	1.12 (0.92–1.36)	0.252
	Codominant	AG	219	215	1.08 (0.83–1.39)	0.575
		GG	36	27	1.39 (0.82–2.35)	0.226
		AA	253	268	1	
		AG-GG	255	242	1.11 (0.87–1.42)	0.404
	Recessive	AA-AG	472	483	1	
		GG	36	27	1.34 (0.80–2.25)	0.265
	Log-additive	–	–	–	1.12 (0.92–1.38)	0.260
rs2980003	Allele	C	653	666	1	
		T	363	352	1.05 (0.88–1.26)	0.587
	Codominant	TC	233	230	1.05 (0.81–1.37)	0.722
		TT	65	61	1.09 (0.73–1.63)	0.663

CI, confidence interval; OR, odds ratio; SNP: single nucleotide polymorphism; OR, Odds ratio, 95 % CI; 95% confidence intervals.

p-values were calculated by unconditional logistic regression analysis with adjustment for age and gender.

p < 0.05 indicates statistical significance.

SNP ID	Model	Allele/Genotype	Case N	Control N	OR (95% CI)	<i>p</i>
		CC	210	218	1	
	Dominant	TC-TT	298	291	1.06 (0.82–1.36)	0.657
	Recessive	CC-TC	443	448	1	
		TT	65	61	1.07 (0.73–1.55)	0.738
	Log-additive	–	–	–	1.05 (0.87–1.26)	0.626
CI, confidence interval; OR, odds ratio; SNP: single nucleotide polymorphism; OR, Odds ratio, 95 % CI; 95% confidence intervals.						
<i>p</i> -values were calculated by unconditional logistic regression analysis with adjustment for age and gender.						
<i>p</i> < 0.05 indicates statistical significance.						

The Association Analyses Stratified By Age And Gender

We then determined the effect of *CYP7B1* polymorphisms on the risk of CHD stratified by age and gender. Table 4 showed the associations under age-based stratification. We found that rs6472155 is significantly associated with an increased risk of CHD in age > 60 years (allele model: G vs A, OR 1.43, 95 % CI = 1.10–1.86, *p* = 0.008; co-dominant model: GG vs AA, OR 2.20, 95 % CI = 1.07–4.49, *p* = 0.031; log-additive: OR 1.39, 95 % CI = 1.06–1.83, *p* = 0.018). On gender stratification (Table 5), our data indicated that rs6472155 can increase the susceptibility to CHD in women under allele model (G vs A, OR 1.48, 95 % CI = 1.06–2.07, *p* = 0.022), co-dominant model (GG vs AA, OR 3.17, 95 % CI = 1.19–8.44, *p* = 0.021), and recessive model (GG vs AA-AG, OR 2.91, 95 % CI = 1.12–7.58, *p* = 0.029).

Table 4
Association between *CYP7B1* polymorphisms and CHD risk stratified by age

SNP ID	Model	Genotype	Case	Control	OR (95% CI)	<i>p</i>	Case	Control	OR (95% CI)	<i>p</i>
			> 60 years				≤ 60 years			
rs7836768	Allele	A	326	342	1		269	274	1	
		G	238	226	1.11 (0.87– 1.40)	0.410	181	178	1.04 (0.79– 1.35)	0.796
	Codominant	AG	140	156	0.91 (0.89– 1.32)	0.624	99	106	0.92 (0.61– 1.39)	0.693
		GG	49	35	1.52 (0.89– 2.58)	0.123	41	36	1.13 (0.66– 1.93)	0.669
		AA	93	93	1		85	84	1	
	Dominant	AG-GG	189	191	1.02 (0.71– 1.45)	0.920	140	142	0.97 (0.66– 1.43)	0.890
	Recessive	AA-AG	233	249	1		184	190	1	
		GG	49	35	1.61 (1.00– 2.59)	0.052	41	36	1.18 (0.72– 1.93)	0.519
	Log-additive	–	–	–	1.15 (0.90– 1.48)	0.266	–	–	1.03 (0.80– 1.34)	0.806
rs6472155	Allele	A	388	431	1		337	320	1	
		G	176	137	1.43 (1.10– 1.86)	0.008	115	132	0.83 (0.62– 1.11)	0.205
	Codominant	AG	124	111	1.31 (0.92– 1.86)	0.130	95	104	0.82 (0.56– 1.19)	0.293
		GG	26	13	2.20 (1.07– 4.49)	0.031	10	14	0.64 (0.27– 1.50)	0.300
	Dominant	AA	132	160	1		121	108	1	
		AG-GG	150	124	1.40 (1.00– 1.95)	0.048	105	118	0.79 (0.55– 1.15)	0.222
	Recessive	AA-AG	256	271	1		216	212	1	

OR, Odds ratio, 95 % CI; 95% confidence intervals.

p values were calculated by logistic regression adjusted by age and gender.

p < 0.05 indicates statistical significance.

SNP ID	Model	Genotype	Case	Control	OR (95% CI)	<i>p</i>	Case	Control	OR (95% CI)	<i>p</i>
			> 60 years		≤ 60 years					
		GG	26	13	1.94 (0.97– 3.90)	0.062	10	14	0.70 (0.30– 1.61)	0.403
	Log-additive	–	–	–	1.39 (1.06– 1.83)	0.018	–	–	0.81 (0.59– 1.10)	0.180
rs2980003	Allele	C	350	369	1		303	297	1	
		T	214	197	1.15 (0.90– 1.46)	0.273	149	155	0.94 (0.72– 1.24)	0.673
	Codominant	TC	126	137	0.97 (0.68– 1.39)	0.856	107	93	1.20 (0.81– 1.77)	0.368
		TT	44	30	1.52 (0.89– 2.61)	0.126	21	31	0.70 (0.38– 1.31)	0.268
		CC	112	116	1		98	102	1	
	Dominant	TC-TT	170	167	1.07 (0.76– 1.50)	0.708	128	124	1.08 (0.74– 1.56)	0.705
	Recessive	CC-TC	238	253	1		205	195	1	
		TT	44	30	1.55 (0.94– 2.57)	0.087	21	31	0.64 (0.36– 1.16)	0.142
	Log-additive	–	–	–	1.15 (0.90– 1.48)	0.261	–	–	0.94 (0.72– 1.24)	0.574
OR, Odds ratio, 95 % CI; 95% confidence intervals.										
<i>p</i> values were calculated by logistic regression adjusted by age and gender.										
<i>p</i> < 0.05 indicates statistical significance.										

Table 5
Association between *CYP7B1* polymorphisms and CHD risk stratified by gender

Association between C777D1 polymorphisms and CHD risk stratified by gender										
SNP ID	Model	Genotype	Case	Control	OR (95% CI)	<i>p</i>	Case	Control	OR (95% CI)	<i>p</i>
			Women				Men			
rs7836768	Allele	A	205	223	1		390	393	1	
		G	141	125	1.23 (0.90–1.67)	0.191	278	279	1.00 (0.81–1.25)	0.971
	Codominant	AG	89	95	1.00 (0.63–1.59)	0.989	150	167	/	/
		GG	26	15	1.87 (0.90–3.89)	0.096	64	56	1.08 (0.69–1.68)	0.734
	Dominant	AA	58	64	1		120	113	1	
		AG-GG	115	110	1.12 (0.72–1.75)	0.616	214	223	0.90 (0.66–1.24)	0.537
	Recessive	AA-AG	147	159	1		270	280	1	
		GG	26	15	1.86 (0.94–3.67)	0.073	64	56	1.19 (0.80–1.77)	0.392
	Log-additive	–	–	–	1.24 (0.89–1.73)	0.201	–	–	1.01 (0.81–1.25)	0.961
	rs6472155	Allele	A	239	266	1		486	485	1
G			109	82	1.48 (1.06–2.07)	0.022	182	187	0.97 (0.76–1.23)	0.812
Codominant		AG	73	70	1.21 (0.78–1.89)	0.393	146	145	/	/

OR, Odds ratio, 95 % CI; 95% confidence intervals.

p values were calculated by logistic regression adjusted by age and gender.

p < 0.05 indicates statistical significance.

SNP ID	Model	Genotype	Case	Control	OR (95% CI)	<i>p</i>	Case	Control	OR (95% CI)	<i>p</i>
			Women				Men			
		GG	18	6	3.17 (1.19–8.44)	0.021	18	21	1.37 (0.83–2.26)	0.652
		AA	83	98	1		170	170	1	
	Dominant	AG-GG	91	76	1.37 (0.89–2.10)	0.148	164	166	0.99 (0.73–1.34)	0.937
	Recessive	AA-AG	156	168	1		316	315	1	
		GG	18	6	2.91 (1.12–7.58)	0.029	18	21	0.86 (0.45–1.64)	0.637
	Log-additive	–	–	–	1.45 (1.02–2.06)	0.037	–	–	0.97 (0.75–1.25)	0.803
rs2980003	Allele	C	225	215	1		428	451	1	
		T	123	133	0.88 (0.65–1.20)	0.432	240	219	1.16 (0.92–1.45)	0.212
	Codominant	TC	81	81	0.92 (0.58–1.46)	0.732	152	149	/	/
		TT	21	26	0.73 (0.37–1.43)	0.362	44	35	1.37 (0.83–2.26)	0.220
		CC	72	67	1		138	151	1	
	Dominant	TC-TT	102	107	0.88 (0.57–1.35)	0.551	196	184	1.16 (0.86–1.58)	0.334
	Recessive	CC-TC	153	148	1		290	300	1	
		TT	21	26	0.76 (0.41–1.43)	0.398	44	35	1.29 (0.81–2.08)	0.286
	Log-additive	–	–	–	0.87 (0.64–1.19)	0.394	–	–	1.15 (0.92–1.45)	0.220
OR, Odds ratio, 95 % CI; 95% confidence intervals.										
<i>p</i> values were calculated by logistic regression adjusted by age and gender.										
<i>p</i> < 0.05 indicates statistical significance.										

CYP7B1 polymorphisms related to CHD susceptibility under smoking, drinking, and diabetes status subgroups

We also detected the associations stratified by smoking, drinking, and diabetes status. As was shown in Table 6, rs2980003 polymorphism has a lower risk of CHD in drinkers under allele model (T vs C, OR 0.57, 95 % CI = 0.35–0.95, $p = 0.031$), co-dominant model (TC vs CC, OR 0.48, 95 % CI = 0.23–0.97, $p = 0.042$), dominant model (TC-TT vs CC, OR 0.47, 95 % CI = 0.24–0.91, $p = 0.025$), and log-additive model (OR 0.59, 95 % CI = 0.36–0.98, $p = 0.041$). In non-drinkers, we observed that rs6472155 significantly increased a risk of CHD (co-dominant model: GG vs AA, OR 3.16, 95 % CI = 1.05–9.48, $p = 0.040$; log-additive model: OR 3.43, 95 % CI = 1.16–10.09, $p = 0.025$). We further evaluated the relationship of polymorphisms with and without diabetes in CHD patients, no significant association were found (Table 7).

Table 6
CYP7B1 polymorphisms related to CHD risk stratified by smoking and drinking status

SNP ID	Model	Genotype	Smoking		Non-smoking		Drinking		Non-drinking	
			OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>
rs7836768	Allele	A	1		1		1		1	
		G	0.89 (0.65– 1.23)	0.485	1.19 (0.87– 1.61)	0.275	1.07 (0.68– 1.71)	0.763	1.25 (0.93– 1.68)	0.139
	Codominant	AG	0.84 (0.50– 1.40)	0.495	0.96 (0.60– 1.54)	0.881	0.65 (0.31– 1.36)	0.250	1.18 (0.75– 1.85)	0.465
		GG	0.83 (0.45– 1.56)	0.565	1.51 (0.76– 2.99)	0.236	1.31 (0.54– 3.16)	0.546	1.71 (0.88– 3.29)	0.111
	Dominant	AA	1		1		1		1	
		AG-GG	0.84 (0.52– 1.35)	0.461	1.07 (0.68– 1.66)	0.781	0.81 (0.41– 1.60)	0.550	1.29 (0.84– 1.98)	0.247
	Recessive	AA-AG	1		1		1		1	
		GG	0.92 (0.53– 1.60)	0.774	1.07 (0.68– 1.66)	0.781	1.66 (0.75– 3.66)	0.209	1.55 (0.85– 2.84)	0.156
	Log-additive	–	0.90 (0.67– 1.23)	0.522	1.16 (0.84– 1.59)	0.370	1.07 (0.68– 1.68)	0.769	1.27 (0.94– 1.73)	0.119
rs6472155	Allele	A	1		1		1		1	
		G	0.83 (0.59– 1.18)	0.294	1.27 (0.90– 1.77)	0.171	0.99 (0.59– 1.66)	0.961	1.15 (0.83– 1.59)	0.387
	Codominant	AG	0.92 (0.57– 1.47)	0.724	0.93 (0.59– 1.44)	0.735	1.36 (0.70– 2.64)	0.371	0.84 (0.56– 1.29)	0.425
		GG	0.55 (0.23– 1.31)	0.175	2.68 (0.93– 7.71)	0.068	0.26 (0.03– 2.21)	0.220	3.16 (1.05– 9.48)	0.040
	Dominant	AA	1		1		1		1	
		AG-GG	0.85 (0.54– 1.33)	0.481	1.05 (0.68– 1.60)	0.841	1.18 (0.61– 2.26)	0.625	0.98 (0.65– 1.47)	0.907
	Recessive	AA-AG	1		1		1		1	

OR, Odds ratio, 95 % CI; 95% confidence intervals.

p values were calculated by logistic regression adjusted by age and gender.

p < 0.05 indicates statistical significance.

SNP ID	Model	Genotype	Smoking		Non-smoking		Drinking		Non-drinking	
			OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>
rs2980003		GG	0.57 (0.25– 1.32)	0.190	2.77 (0.98– 7.83)	0.055	0.23 (0.03– 1.18)	0.170	3.43 (1.16– 10.09)	0.025
	Log-additive	–	0.82 (0.57– 1.17)	0.268	1.19 (0.83– 1.69)	0.346	0.95 (0.55– 1.64)	0.863	1.16 (0.83– 1.63)	0.376
	Allele	C	1		1		1		1	
		T	0.97 (0.70– 1.36)	0.879	1.04 (0.77– 1.41)	0.795	0.57 (0.35– 0.95)	0.031	0.96 (0.72– 1.30)	0.810
	Codominant	TC	1.07 (0.66– 1.72)	0.790	1.06 (0.67– 1.69)	0.805	0.48 (0.23– 0.97)	0.042	0.98 (0.63– 1.53)	0.927
		TT	0.86 (0.41– 1.79)	0.682	1.03 (0.54– 1.96)	0.928	0.44 (0.15– 1.32)	0.143	0.93 (0.49– 1.53)	0.833
		CC	1		1		1		1	
	Dominant	TC-TT	1.02 (0.65– 1.60)	0.933	1.05 (0.68– 1.63)	0.818	0.47 (0.24– 0.91)	0.025	0.97 (0.64– 1.48)	0.882
	Recessive	CC-TC	1		1		1		1	
		TT	0.83 (0.41– 1.68)	0.603	1.00 (0.55– 1.81)	0.995	0.61 (0.21– 1.75)	0.359	0.94 (0.52– 1.71)	0.849
	Log-additive	–	0.97 (0.69– 1.36)	0.857	1.03 (0.75– 1.39)	0.875	0.59 (0.36– 0.98)	0.041	0.97 (0.72– 1.31)	0.840
OR, Odds ratio, 95 % CI; 95% confidence intervals.										
<i>p</i> values were calculated by logistic regression adjusted by age and gender.										
<i>p</i> < 0.05 indicates statistical significance.										

Table 7
Association of *CYP7B1* polymorphisms in CHD patients with and without diabetes

SNP ID	Model	Genotype	With diabetes	Without diabetes	OR (95% CI)	<i>p</i>
rs7836768	Allele	A	185	410	1	
		G	105	314	0.74 (0.56–0.98)	0.036
	Codominant	AG				0.153
			65	174	0.73 (0.48–1.12)	
		GG	20	70	0.56 (0.31–1.02)	0.057
		AA	60	118	1	
	Dominant	AG-GG	85	244	0.69 (0.46–1.02)	0.063
	Recessive	AA-AG	125	292	1	
		GG	20	70	0.67 (0.39–1.15)	0.147
		–	–	–	0.75 (0.57–0.99)	0.042
rs6472155	Allele	A	214	511	1	
		G	78	213	0.87 (0.64–1.19)	0.388
	Codominant	AG	54	165	0.77 (0.51–1.17)	0.226
		GG	12	24	1.02 (0.56–1.86)	0.949
		AA	80	173	1	
	Dominant	AG-GG	66	189	0.73 (0.50–1.08)	0.118
	Recessive	AA-AG	134	338	1	
		GG	12	24	1.16 (0.56–2.42)	0.685
		–	–	–	0.84 (0.61–1.15)	0.276
	Log-additive	–	–	–		
rs2980003	Allele	C	190	463	1	
		T	102	261	0.95 (0.72–1.27)	0.736
	Codominant	TC	60	173	0.77 (0.51–1.17)	0.226
		TT	21	44	1.02 (0.56–1.86)	0.949
		CC	65	145	1	
	Dominant	TC-TT	81	217	0.82 (0.56–1.22)	0.332

OR, Odds ratio, 95 % CI; 95% confidence intervals.

p values were calculated by logistic regression adjusted by age and gender.

p < 0.05 indicates statistical significance.

SNP ID	Model	Genotype	With diabetes	Without diabetes	OR (95% CI)	<i>p</i>
	Recessive	CC-TC	125	318	1	
		TT	21	44	1.16 (0.66–2.04)	0.600
	Log-additive	–	–	–	0.94 (0.70–1.25)	0.657
OR, Odds ratio, 95 % CI; 95% confidence intervals.						
<i>p</i> values were calculated by logistic regression adjusted by age and gender.						
<i>p</i> < 0.05 indicates statistical significance.						

Effect Of SNP-SNP Interactions On CHD Susceptibility

MDR analyses were performed to determine the influence of SNP-SNP interactions. Our results were summarized in Table 8, the combinations of rs7836768, rs6472155, and rs2980003 are the best three-locus model to predict CHD (testing accuracy = 0.4764, CVC = 10/10, *p* = 0.002). The two-locus model included rs7836768 and rs6472155 (testing accuracy = 0.5079, CVC = 8/10, *p* = 0.021). Interestingly, the diagram analysis revealed that rs7836768 and rs10808739 had strong interactions (Fig. 1).

Table 8
The analysis of SNP-SNP interaction models using MDR method

Model	Training Bal. Acc.	Testing Bal. Acc.	CVC	OR (95% CI)	<i>p</i>
rs7836768	0.5236	0.4931	9/10	1.19 (0.93–1.52)	0.167
rs7836768,rs6472155	0.5336	0.5079	8/10	1.39 (1.05–1.83)	0.021
rs7836768,rs6472155,rs2980003	0.5454	0.4764	10/10	1.59 (1.19–2.13)	0.002
Bal. Acc., Balanced accuracy; CVC, Cross-validation consistently.					
<i>p</i> values were calculated by χ^2 test. <i>p</i> < 0.05 indicates statistical significance.					

The Association Between SNP Genotypes And Clinical Indicators

Finally, we studied the possible association of SNP with clinical indicators in CHD patients. As was showed in Table 9, the AG genotype (289.796 ± 84.427 umol/l) and GG genotype (296.096 ± 68.438 umol/l) of rs6472155 polymorphism were lower uric acid level than AA genotype (310.364 ± 86.950 umol/l) (*p* = 0.034).

Table 9
Comparisons between clinical characteristics and SNP genotypes

SNP	Urea (mmol/l)	Creatinine (umol/l)	Uric acid (umol/l)	Total-cholesterol (mmol/l)	Apolipoprotein AI (g/l)
rs7836768					
AA	5.167 ± 1.422	2.350 ± 0.841	302.857 ± 82.083	4.024 ± 0.999	1.185 ± 0.225
AG	5.210 ± 1.592	2.370 ± 0.867	297.444 ± 87.826	4.066 ± 1.068	1.170 ± 0.238
GG	5.178 ± 1.556	2.430 ± 0.785	304.943 ± 84.697	4.186 ± 1.091	1.149 ± 0.255
<i>p</i>	0.958	0.761	0.718	0.501	0.518
rs6472155					
AA	5.209 ± 1.543	2.340 ± 0.825	310.364 ± 86.950	4.039 ± 1.083	1.179 ± 0.236
AG	5.160 ± 1.493	2.410 ± 0.888	289.796 ± 84.427	4.125 ± 1.011	1.162 ± 0.230
GG	5.186 ± 1.615	2.420 ± 0.753	296.096 ± 68.438	4.055 ± 1.045	1.165 ± 0.279
<i>p</i>	0.942	0.636	0.034	0.684	0.737
rs2980003					
TT	5.042 ± 1.387	2.300 ± 0.909	290.025 ± 86.984	3.885 ± 1.011	1.126 ± 0.223
CT	5.285 ± 1.599	2.370 ± 0.861	302.334 ± 81.741	4.075 ± 1.082	1.176 ± 0.224
CC	5.117 ± 1.474	2.410 ± 0.810	301.934 ± 88.426	4.142 ± 1.020	1.180 ± 0.254
<i>p</i>	0.384	0.639	0.569	0.239	0.261
<i>p</i> values were calculated by Kruskal-Wallis H test.					
<i>p</i> < 0.05 indicates statistical significance.					

Discussion

In our study, we determined the influence of *CYP7B1* polymorphisms on CHD susceptibility. Our study showed that *CYP7B1* genetic variants significantly relate to the risk of CHD in Chinese Han population. Besides, we found that rs6472155 polymorphism was associated with uric acid level. To our best known, our study is the first time to investigate the effect of *CYP7B1* genetic variants on CHD susceptibility.

CHD is a kind of multifactorial disease resulting from environmental and genetic factor. An increasing number studies strongly supported that genetic polymorphisms involve in the risk of CHD, such as *MMPs* (20), *CD36* (21), *MYH15* (22), and *SCARB1* (23). The *CYP7B1* gene is located on chromosome 8q21.3. *CYP7B1* is known to be involved in cholesterol

synthesis of bile acids, which contributes to the CHD progression. Whereas there have been no studies on the genetic polymorphism in CHD. Therefore, we tried to evaluate the association between *CYP7B1* polymorphisms and CHD risk. Age-based stratification showed that rs6472155 is significantly associated with an increased risk of CHD in age > 60 years. When stratified by gender, we found that rs6472155 can increase the susceptibility to CHD in women. In drinkers, rs2980003 polymorphism has a lower risk of CHD while rs6472155 significantly increased a risk of CHD in non-drinkers. Our results suggest that the *CYP7B1* polymorphism influencing the risk of CHD may rely on the age, gender, and drinking status.

The association of SNP-SPN interaction with CHD risk may help to discover the risk factors because of this disease caused by environmental and genetic interaction. We observed that the combinations of rs7836768, rs6472155, and rs2980003 are the best model to predict CHD.

Some clinical indicators including serum uric acid can be used as prognostic marker of CHD (24). Therefore, we detected the effect of SNPs on clinical indicators in CHD. Our data showed that rs6472155 polymorphism significantly correlated with uric acid level. We guess that the SNPs of *CYP7B1* are related to the clinical indicator and we'll verify the result in the next work.

Some limitations exist in our present study. First, we investigated the association between *CYP7B1* polymorphisms and CHD susceptibility, the relationship between *CYP7B1* SNPs and the gene expression should be tested in future. Second, the molecular mechanism of *CYP7B1* in CHD is needed to be performed in the next work. In spite the above limitations, our study gives available information for the molecular mechanism of CHD in Chinese Han population.

Conclusion

In summary, this study showed that rs6472155 is significantly related to an increased susceptibility to CHD in age > 60 years, women, and non-smokers. Rs2980003 polymorphism has a lower risk of CHD in drinkers. Besides, it was found that rs6472155 polymorphism was associated with uric acid level. These data may give a new potential biomarker for the prevention and management of CHD in Chinese Han population.

Declarations

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Authors' contributions

TB J and TB L conceived, designed the experiments. XB Z and AS L recruited and collected study samples. HQ W and Q W selected the SNPs and designed primers. J H and M L analyzed the data. TB L wrote the manuscript. All authors read and approved the final manuscript.

Ethics approval and consent to participate

All procedures performed in studies involving human participants were in accordance with the ethical standards of the Affiliated Haikou Hospital of Xiangya Medical College and Informed consent was obtained from all individual

participants included in the study. Our study has been approved by the Medical Ethics Committee of Affiliated Haikou Hospital of Xiangya Medical College. Experiments in this study were performed following the protocol of Helsinki's Declaration.

Patient consent for publication

Not applicable.

Competing interests

All authors declare that they have no competing interests.

Data Availability Statement

Participant informed consent statements did not seek consent for data to be made publicly available; however, data will be made available to individual researchers upon reasonable request. In this case, please contact the corresponding author.

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Figures

Figure 1

The tree diagram analysis among SNP interaction