Original Article

Relationship between Modulator Recognition Factor 2/AT-rich Interaction Domain 5B Gene Variations and Type 2 Diabetes Mellitus or Lipid Metabolism in a Northern Chinese Population

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Abstract

Background: Four single nucleotide polymorphisms (SNPs) in the modulator recognition factor 2/AT-rich interaction domain 5B (*MRF2/ARID5B*) gene located at chromosome 10q21.2 have been shown to be associated with both type 2 diabetes mellitus (T2DM) and coronary artery disease in a Japanese cohort. This study aimed to investigate the relationship between these SNPs (rs2893880, rs10740055, rs7087507, rs10761600) and new-onset T2DM and lipid metabolism in a Northern Chinese population.

Methods: This was a case-control study. The rs2893880, rs10740055, rs7087507, and rs10761600 genetic variants were genotyped by SNPscan and analyzed in relation to T2DM susceptibility in 2000 individuals (999 with newly diagnosed T2DM and 1001 controls without diabetes mellitus). Associations between the *MRF2/ARID5B* genetic models and T2DM were determined by multivariate logistic regression.

Results: Regarding the rs10740055 SNP, AA was associated with a higher risk of T2DM compared with codominant-type CC (adjusted by sex, age, and body mass index [BMI], $P = 0.041$, odds ratio [*OR*] = 1.421, 95% confidence interval [*CI*] 1.014–1.991). Meanwhile, AA individuals were at increased risk of presenting with T2DM compared with individuals with CC or a single C (adjusted by sex, age, and BMI, $P = 0.034$, *OR* = 1.366, 95% *CI* 1.023–1.824). With respect to rs10761600, AT contributed to a higher risk of T2DM compared with AA (adjusted by sex, age, and BMI, $P = 0.013$, *OR* = 1.585, 95% *CI* 1.101–2.282), while TT also increased the risk of presenting with T2DM compared with AA or A (adjusted by sex, age, and BMI, $P = 0.004$, *OR* = 1.632, 95% *CI* 1.166–2.284). High-density lipoprotein cholesterol (HDL-C) levels were significantly different among the three genotypes of rs7087507 in the controls ($P = 0.048$) (GG>GA).

Conclusions: The present results identified *MRF2/ARID5B* as a potential susceptibility gene for new-onset T2DM in a Northern Chinese population, while the rs7087507 SNP was associated with HDL-C levels. Further larger studies are required to validate these findings.

Key words: Diabetes Mellitus Type 2; Lipid Metabolism; Polymorphism; Single-nucleotide

Introduction

Type 2 diabetes mellitus (T2DM) is a major public health concern. Approximately 382 million (8.3%) patients worldwide are thought to suffer from T2DM, and this number is expected to increase to 592 million by 2035.[1] T2DM is a complex metabolic disorder characterized by hyperglycemia resulting from pancreatic beta-cell dysfunction and insulin resistance. It is associated with abnormal glucose metabolism and is also usually accompanied by abnormal lipid metabolism, both of which significantly increase the risk of coronary artery disease.

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At least 75 genetic variants have been identified as associated with T2DM thanks to the advent of genome-wide association studies (GWASs).[2] However, the effects of ethnicity and/or

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environmental factors mean that the same genetic variants may have different correlations with T2DM in different populations.

Modulator recognition factor-2 (MRF2) is a member of the AT-rich interaction domain (ARID) family of transcription factors (also known as *ARID5B* or *Desrt*). The *MRF2/ARID5B* gene is located at chromosome 10q21.2, which has been identified by GWAS as the most susceptible region for serum lipid levels.[3] Targeted disruption of *MRF2/ARID5B* in mice resulted in stunted growth and reduced lipid accumulation.[4-6] Furthermore, both *in vivo* and *in vitro* studies have indicated that the *MRF2/ARID5B* gene may affect adipogenesis.[7-9] Research in a Norwegian cohort identified an association between DNA methylation of the *ARID5B* gene in cord blood and birth weight.[10] These results suggest that *MRF2/ARID5B* may play an important role in growth and lipid metabolism.

Recent Japanese studies showed that four single nucleotide polymorphisms (SNPs) in the *MRF2/ARID5B* gene (rs2893880, rs10740055, rs7087507, and rs10761600) associated with susceptibility to coronary artery disease were also associated with T2DM.[11,12] However, the relationships between these four SNPs and T2DM have only been reported in a Japanese population.

Based on the results of previous studies, we conducted a case-control study to evaluate the associations between these four *MRF2/ARID5B* SNPs and new-onset T2DM and lipid metabolism in a Northern Chinese population.

Methods

Ethical approval

The study protocol was reviewed and approved by the Ethics Committee of Harbin Medical University. All patients and controls received adequate information about this study and provided written informed consent.

Study population

Using a case-control approach, a total of 2000 participants (aged 20–79 years) including 999 patients with newly diagnosed T2DM and 1001 controls were included in our study. None of the participants were genetically related to each other. The patients were recruited from the Endocrinology and Metabolism Department of the Second Affiliated Hospital of Harbin Medical University from March 2013 to May 2015. T2DM was diagnosed in accordance with the World Health Organization criteria.[13] The duration of newly diagnosed T2DM was <6 months. Patients who used oral medications or insulin injections to achieve adequate glucose control and individuals with type 1 diabetes, gestational diabetes, or other special types of diabetes were excluded from the case group. Patients with acute diabetic complications or other serious metabolic diseases that might raise glucose levels were also excluded from the study.

Controls with a fasting plasma glucose (FPG) concentration <5.1 mmol/L and hemoglobin A1c (HbA1c) <6.0% and no family history of T2DM were enrolled from the physical examination center or outpatient clinics at the same hospital. Control individuals were required to meet the following criteria: no heart disease, liver dysfunction, malignancy, or other serious systemic disease, and no history of drugs known to influence glucose or lipid metabolism.

Anthropometric and clinical measurements

Anthropometric measurements including height, weight, waist and hip circumferences, and systolic and diastolic blood pressures were measured using standardized procedures. Waist-hip ratio was calculated as waist circumference (cm) divided by hip circumference (cm). Body mass index (BMI) was calculated as body weight in kilograms divided by the square of height in meters. The homeostasis model assessment (HOMA) was used to assess individual insulin resistance (HOMA-IR). HOMA-IR = (FPG [mmol/L] × FIN [mU/L])/22.5, FIN represents fasting insulin. HOMA-β was used to assess islet beta-cell secretion function. HOMA-β = FIN × 20/(FPG − 3.5).[14,15]

Peripheral venous blood samples were collected in tubes from all participants during the fasting state. Plasma insulin levels were measured by double-antibody radioimmunoassay. FPG was quantified by the glucose oxidase-peroxidase procedure (Modular DPP, Roche Diagnostics GmbH, Mannheim, Germany). Serum total cholesterol (TC), triglycerides (TGs), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein (LDL) levels were measured using an automatic biochemical analyzer (Modular DPP, Roche Diagnostics GmbH). HbA1c levels were measured using a high-performance liquid chromatography system (Bio-Rad DIA-MAT glycosylated hemoglobin analyzer system, Bio-Rad, Hercules, CA, USA).

Genotyping

Peripheral blood samples were collected in tubes containing Na_2EDTA and stored at −20°C for further analysis. Genomic DNA was extracted from peripheral blood leukocytes using a TIANamp Genomic DNA Kit (Tiangen Biotech Co., Ltd., Beijing, China) according to the standard procedure. Four SNPs were genotyped, rs2893880, rs10740055, rs7087507, and rs10761600, using a custom-by-design 48-Plex SNPscan™ kit (Genesky Biotechnologies Inc., Shanghai, China). This kit was developed based on multiplex fluorescence polymerase chain reaction according to patented technology elaborated by Genesky Biotechnologies Inc. The accuracy of the genotyping using the SNPscan™ Kit was validated by genotyping of a random sample of 5% of cases and controls twice for all SNPs, by different people. Specifically, 100 pairs of blind duplicates were performed with a concordance rate >98%.[16] Moreover, these variants exhibited allelic frequencies >15% in Han Chinese (National Center for Biotechnology Information dbSNP database: https://www.ncbi.nlm.nih.gov/snp/).

Statistical analysis

Deviation from the Hardy–Weinberg equilibrium was assessed by exact tests (http://ihg.gsf.de/). Continuous parameters were compared using Student's *t*-tests (normal distribution) or nonparametric tests (nonnormal distribution), and categorical variables were compared using Chi-square tests. Data were described as mean ± standard deviation for normally distributed data or median (interquartile range) for nonnormally distributed data. A $P < 0.05$ was considered statistically significant for all data. Statistical evaluations of the associations between the case–control status and each individual SNP were measured as odds ratios (*OR*s) and 95% confidence intervals (*CI*s), estimated using unconditional logistic regression after adjusting for age, gender, and BMI. Quantitative traits among different genotypes were examined by analysis of variance (ANOVA). Associated statistical analyses were performed using SPSS software (version 17.0; SPSS, Chicago, IL, USA). We examined the degree of linkage disequilibrium (LD) between the polymorphisms and determined if the haplotype block associated with T2D using Haploview version 4.2 (http://www.broad.mit.edu/mpg/haploview).

Results

Clinical and biochemical characteristics of the study participants

The study participants comprised 999 new-onset T2DM patients and 1001 controls. The clinical and biochemical characteristics of the study groups are summarized in Table 1. There were no significant differences between the cases and controls with respect to sex ($P = 0.232$) and serum LDL cholesterol levels ($P = 0.742$). However, the new-onset T2DM patients had higher age, BMI, waist-hip ratio, FPG, HbA1c, fasting insulin, HOMA-IR, TC, TG, and blood pressure but lower serum HDL-C levels and HOMA-β (all $P < 0.001$) compared with controls.

Associations between modulator recognition factor 2/AT-rich interaction domain 5B variants and type 2 diabetes mellitus

The genotypes of some samples were not detected because of problems with the SNP detection technology or the quality of the experimental samples. Genotyping of SNP rs2893880, rs10740055, and rs10761600 was successful for 996 T2DM patients and 977 controls and of rs7087507 for 995 T2DM patients and 975 controls. The genotype distributions of the four SNPs were in Hardy–Weinberg equilibrium in both cases and controls. The genotype distributions of the four variants of *MRF2/ARID5B* are shown in Table 2. Analysis of rs2893880 found no differences in the frequency distributions of the GC and CC genotypes compared with the GG genotype between the cases and controls ($P > 0.05$). Likewise, rs10740055, rs7087507, and rs10761600 were not associated with T2DM.

We further analyzed the effects of the four SNPs under three different genetic models by logistic tests [Table 3]. rs10740055 and rs10761600 were associated with T2DM risk according to both the additive and recessive models after adjusting for sex, age, and BMI ($P < 0.05$). For the rs10740055 SNP, AA seemed to increase the risk of T2DM compared with wild-type CC (adjusted $P = 0.041$, *OR* = 1.421, 95% *CI* 1.014–1.991). Meanwhile, AA individuals were at increased risk of T2DM compared with those with CC or a single C (recessive model, Akaike information criterion value = 1839; adjusted $P = 0.034$, *OR* = 1.366, 95% *CI* 1.023–1.824). With respect to rs10761600, TT seemed to increase the risk of presenting with T2DM compared with AA (adjusted $P = 0.013$, *OR* = 1.585, 95% *CI* 1.101–2.282) and carrying TT increased the risk of T2DM compared with AA or a single A (recessive model Akaike information criterion value = 1843; adjusted $P = 0.004$, *OR* = 1.632, 95% *CI* 1.166–2.284). However,

Table 1: Clinical and laboratory characteristics of cases and controls

Characteristics	Case (n = 999)	Control (n = 1001)	Statistics	*P*
Male, *n* (%)	612 (61.3)	587 (58.6)	1.429*	0.232
Age (years)	46.11 ± 12.55	42.87 ± 11.75	−5.969†	<0.001
BMI (kg/m²)	25.76 ± 3.59	23.32 ± 3.34	−15.717†	<0.001
WHR	0.94 ± 0.06	0.85 ± 0.07	−25.553†	<0.001
FBG (mmol/L)	10.03 ± 3.40	4.82 ± 0.33	−48.171†	<0.001
HbA1c (%)	9.30 ± 2.36	5.12 ± 0.47	−54.852†	<0.001
FI (μU/ml)	12.86 ± 7.59	7.86 ± 4.40	−18.045†	<0.001
HOMA-IR	4.80 (2.73–7.48)	1.51 (1.01–2.17)	−30.233‡	<0.001
HOMA-β	38.70 (22.05–69.60)	104.50 (70.01–149.00)	−25.615‡	<0.001
TC (mmol/L)	5.00 ± 1.29	4.89 ± 1.08	−2.013†	0.044
TG (mmol/L)	2.38 ± 2.25	1.43 ± 1.01	−12.114†	<0.001
HDL (mmol/L)	1.21 ± 0.32	1.48 ± 0.37	16.958†	<0.001
LDL (mmol/L)	2.91 ± 0.96	2.93 ± 0.90	0.330†	0.742
SBP (mmHg)	130.08 ± 17.56	121.10 ± 15.04	−12.148†	<0.001
DBP (mmHg)	84.58 ± 11.22	79.16 ± 9.64	−11.597†	<0.001

Data are presented as means ± SD or median (interquartile range). *Means χ^2 value; †Means *t* value; ‡Means *Z* value. BMI: Body mass index; WHR: Waist-hip ratio; FBG: Fasting blood glucose; HbA1c: Hemoglobin A1c; FI: Fasting insulin; HOMA-IR: Homeostasis model assessment of insulin resistance; HOMA-β: Homeostasis model assessment of β cell function; TC: Total cholesterol; TG: Triglyceride; HDL: High-density lipoprotein; LDL: Low-density lipoprotein; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; SD: Standard deviation.

Table 2: The genotype distributions of the four variants in *MRF2/ARID5B* (*n* (%))

SNPs	Location	Genotype	T2DM (*n* = 999)	Control (*n* = 1001)	χ^2	*P*
rs2893880	chr10:61937857		*n* = 996	*n* = 977	2.208	0.332
		GG	414 (41.6)	408 (41.8)		
		GC	464 (46.6)	433 (44.3)		
		CC	118 (11.8)	136 (13.9)		
rs10740055	chr10:61958720		*n* = 996	*n* = 977	2.621	0.270
		CC	266 (26.7)	291 (29.8)		
		CA	518 (52.0)	496 (50.8)		
		AA	212 (21.3)	190 (19.4)		
rs7087507	chr10:61985930		*n* = 995	*n* = 975	0.056	0.972
		AA	384 (38.6)	372 (38.1)		
		GA	469 (47.1)	461 (47.1)		
		GG	142 (14.3)	142 (14.5)		
rs10761600	chr10:61997361		*n* = 996	*n* = 977	3.061	0.217
		AA	368 (36.9)	374 (38.3)		
		AT	471 (47.3)	476 (48.7)		
		TT	157 (15.8)	127 (13.0)		

SNPs: Single nucleotide polymorphisms; T2DM: Type 2 diabetes mellitus; *MRF2*: Modulator recognition factor 2; *ARID5B*: AT-rich interaction domain 5B.

Table 3: Association between *MRF2/ARID5B* genetic models and T2DM with multivariable logistic regression

SNPs	Genotype	Additive model				Dominant model*			
		β	*P*	*OR* (95% *CI*)	AIC	β	*P*	*OR* (95% *CI*)	AIC
rs2893880	GG	Reference			1842	−0.067	0.560	0.934 (0.742–1.176)	1843
	GC	−0.028	0.811	0.971 (0.762–1.237)					
	CC	−0.218	0.246	0.802 (0.553–1.164)					
rs10740055	CC	Reference			1839	0.132	0.283	1.148 (0.892–1.479)	1842
	CA	0.056	0.655	1.062 (0.814–1.386)					
	AA	0.340	0.041	1.421 (1.014–1.991)					
rs7087507	AA	Reference			1841	−0.168	0.147	0.841 (0.665–1.063)	1840
	GA	−0.165	0.169	0.841 (0.657–1.076)					
	GG	−0.175	0.340	0.840 (0.587–1.202)					
rs10761600	AA	Reference			1836	0.060	0.626	1.060 (0.838–1.341)	1836
	AT	−0.047	0.687	0.951 (0.743–1.217)					
	TT	0.454	0.013	1.585 (1.101–2.282)					

SNPs	Recessive model†			
	β	*P*	*OR* (95% *CI*)	AIC
rs2893880	−0.203	0.251	0.815 (0.574–1.156)	1842
rs10740055	0.304	0.034	1.366 (1.023–1.824)	1839
rs7087507	−0.083	0.647	0.926 (0.665–1.288)	1841
rs10761600	0.481	0.004	1.632 (1.166–2.284)	1843

P values and *OR* values were adjusted for age, sex, body mass index. *Dominant model: rs2893880, GC+CC versus GG; rs10740055, CA+AA versus CC; rs7087507, GA+GG versus AA; rs10761600, AT+TT versus AA; †Recessive model: rs2893880, CC versus GC+GG; rs10740055, AA versus CA+CC; rs7087507, GG versus GA+AA; rs10761600, TT versus AT+AA. SNPs: Single nucleotide polymorphisms; AIC: Akaike information criterion; *OR*: Odds ratio; *CI*: Confidential interval; *ARID5B*: AT-rich interaction domain 5B; T2DM: Type 2 diabetes mellitus.

there was no association in any genetic models for the SNPs rs7087507 and rs2893880 ($P > 0.05$).

Linkage disequilibrium analysis of the constructed haplotype block with type 2 diabetes mellitus

The combined effect of the four SNPs was evaluated by haplotype analysis using Haploview. Pairwise LD D' values between SNPs and the reconstructed LD plots of the four SNPs are shown in Figure 1. Two SNPs (rs7087507and rs10761600) were in strong LD with each other and therefore formed a haplotype block [Figure 1]. However, the haplotype distributions were not significantly different between the cases and controls (data not shown).

Associations between modulator recognition factor 2/AT-rich interaction domain 5B single nucleotide polymorphisms and serum lipid levels in controls

We further analyzed the relationships between serum lipid metabolism and SNPs in the controls. Serum HDL-C levels were significantly different among the three genotypes in normal individuals (P = 0.048) as shown by ANOVA [Table 4], and serum HDL-C levels were significantly higher in individuals harboring the rs7087507 GG genotype compared with the GA genotype (P = 0.037). However, none of the SNPs were significantly associated with quantitative traits related to glucose or other lipid-related traits in the control population (P > 0.05).

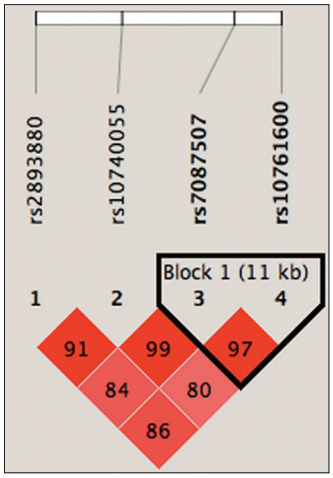

Figure 1: LD patterns of four genotyped SNPs in a Northern Chinese population. The LD between the SNPs is measured as D' and shown in the diamond at the intersection of the diagonals from each SNP. Numbers in the red square indicating strong LD (|D'| > 0.8) and a logarithm of odds score ≥2.0. The analysis track at the top shows the SNPs according to chromosomal location. One haplotype block (outlined in bold black line) indicating markers that are in high LD are shown. SNPs: Single nucleotide polymorphisms; LD: Linkage disequilibrium.

Discussion

We evaluated the association between four SNPs of the *MRF2/ARID5B* gene and susceptibility to new-onset T2DM in a hospital-based case-control study of a Northern Chinese population. Two variants (rs10740055, rs10761600) were significantly associated with T2DM risk, and SNP rs7087507 was associated with serum HDL-C levels.

Previous studies by Japanese researchers found that rs2893880, rs10740055, rs7087507, and rs10761600 variants in the *MRF2/ARID5B* gene were associated with T2DM, with C, A, A, and T being the respective risk alleles. The four variants were also associated with higher HbA1c and FPG levels.[11] In the present study, participants who harbored the *MRF2/ARID5B* rs10740055AA genotype had a higher incidence of T2DM, and the risk of T2DM was also increased in subjects harboring the T allele of rs10761600, consistent with the results of the previous study. The rs10740055 and rs10761600 SNPs thus appear to be common loci for T2DM susceptibility among Asian populations. However, we found no association between either rs2893880 or rs7087507 SNP and T2DM in the current sample of a Northern Chinese population. This apparent inconsistency may be the result of genetic heterogeneity, different geographic locations, different genetic origins, or differences in gene structure

Table 4: The association analysis of *MRF2/ARID5B* variants and serum lipid level in control subjects (n = 1001)

SNPs	Genotype	TC (mmol/L)	TG (mmol/L)	HDL (mmol/L)	LDL (mmol/L)
rs2893880	CC	4.83 ± 1.11	1.34 ± 1.01	1.52 ± 0.38	2.90 ± 0.94
	GC	4.92 ± 1.08	1.50 ± 1.11	1.45 ± 0.34	2.95 ± 0.91
	GG	4.86 ± 1.02	1.41 ± 0.90	1.48 ± 0.38	2.90 ± 0.83
	F	0.501	1.457	2.150	0.353
	P	0.606	0.233	0.177	0.703
rs10740055	AA	4.93 ± 1.04	1.39 ± 0.92	1.51 ± 0.38	2.94 ± 0.87
	CA	4.91 ± 1.05	1.51 ± 1.10	1.45 ± 0.36	2.95 ± 0.88
	CC	4.80 ± 1.08	1.35 ± 0.91	1.49 ± 0.36	2.88 ± 0.90
	F	1.219	2.369	2.203	0.495
	P	0.296	0.094	0.111	0.610
rs7087507	GG	4.83 ± 1.06	1.36 ± 1.00	1.52 ± 0.37	2.88 ± 0.85
	GA	4.89 ± 1.08	1.51 ± 1.11	1.44 ± 0.39	2.94 ± 0.92
	AA	4.88 ± 1.02	1.39 ± 0.89	1.49 ± 0.37	2.92 ± 0.84
	F	0.199	1.957	3.055	0.242
	P	0.820	0.142	0.048	0.785
rs10761600	TT	4.94 ± 0.98	1.39 ± 0.94	1.51 ± 0.36	2.92 ± 0.82
	AT	4.87 ± 1.04	1.49 ± 1.07	1.45 ± 0.37	2.93 ± 0.87
	AA	4.88 ± 1.11	1.39 ± 0.96	1.48 ± 0.36	2.92 ± 0.93
	F	0.243	1.123	1.731	0.005
	P	0.785	0.326	0.178	0.995

SNPs: Single nucleotide polymorphisms; TC: Total cholesterol; TG: Triglyceride; HDL: High-density lipoprotein; LDL: Low-density lipoprotein; *MRF2*: Modulator recognition factor 2; *ARID5B*: AT-rich interaction domain 5B.

caused by gene–environment interactions. We also found no significant associations between these SNPs and fasting glucose levels or HbA1C in our study, suggesting that these genetic variations may affect the incidence but not severity of T2DM in Chinese.

The precise role of *MRF2/ARID5B* with regard to T2DM risk is still under investigation. The *MRF2/ARID5B* gene encodes a member of a novel class of DNA-binding proteins known as the ARID family.[17] rs10761600 and rs10740055 are located within the second and third intron regions of *MRF2/ARID5B*, which region may give rise to alternatively spliced mRNAs and thus affect mRNA stability or processing,[18] with possible impacts on *MRF2/ARID5B* function. Gene variants in this region may affect gene function and thus increase disease risk.

Several studies, including in mice lacking the *MRF2/ARID5B* gene[4-6] and other *in vivo* and *in vitro* research,[7-9] as well as evidence from epigenetic results,[10] implicate the *MRF2/ARID5B* gene in diabetes through its regulation of adipogenesis. These results support a direct relationship between *MRF2/ARID5B* and lipid metabolism and insulin resistance and a critical role in the pathogenesis of T2DM.

MRF2/ARID5B is located at chromosome region 10q21.2, which has been identified by GWAS as the most susceptible region for serum lipid levels.[3] Although we found no association between rs7087507 and T2DM, our results revealed that the rs7087507 SNP was related to serum HDL-C levels (P = 0.048, F = 3.055) (GG>GA) and that HDL-C levels were lower in diabetic patients compared with controls (mean HDL-C 1.21 ± 0.32 vs. 1.48 ± 0.37). Furthermore, a Japanese haplotype analysis revealed that the haplotype G (rs2893880)-C (rs10740055)-G (rs7087507)-A (rs10761600) was negatively associated with susceptibility to coronary artery disease (P = 0.049).[12] Several studies of genetic animal models have proven that increased serum HDL-C levels may protect against atherosclerosis,[19-21] while other researchers have also demonstrated that low HDL-C levels are a well-defined risk factor for the development of cardiovascular diseases.[22]

Wang *et al*. showed that individuals with haplotype CAAT constructed from the four tested *MRF2/ARID5B* SNPs were associated with a 1.86-fold increase in the prevalence of T2DM compared with individuals with GCGA (*OR* 1.86, 95% *CI* 1.43–2.41).[11] Nevertheless, although two SNPs (rs7087507 and rs10761600) were in strong LD with each other and formed a haplotype block in our study, we found no significant difference in haplotype distributions between cases and controls. It is possible that some causal variants may be ethnic-specific or could be present elsewhere in the same or nearby genes. Differences in the patterns of LD between these SNPs and functional variants at these loci could underlie these disparate findings.

The strengths of our study included the fact that all individuals were of the same ethnicity. Most inhabitants of Harbin have lived there for at least three generations,[23,24] and the gene pool of the Harbin Han Chinese population is thus relatively stable. It has been suggested that the strong LD in the haplotype block constructed in our study reflected the characteristics of our cohort. The current study also had some limitations. First, we did not classify both T2DM and low HDL as necessary distinctions between cases and controls, which may have resulted in selection bias. However, the fact that all the controls had attended for routine physical examinations and were not hospitalized patients with specific diseases increased the likelihood that the controls were more representative of the general population. Further well-designed investigations with larger sample sizes are warranted to confirm our findings.

In summary, our results indicate that the *MRF2/ARID5B* gene is associated with new-onset T2DM in a Northern Chinese population. Among the four *MRF2/ARID5B* SNPs screened, rs10761600 and rs10740055 are associated with T2DM, while rs7087507 is associated with serum HDL-C levels. Further well-designed studies with larger samples are required to validate these findings.

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Conflicts of interest

There are no conflicts of interest.

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