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Original Article

Association of rs7903146 and rs7085532 polymorphisms in TCF7L2 gene with type 2 diabetes mellitus in a Chinese Uygur population

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Abstract: The transcription factor 7-like 2 gene (TCF7L2) has been reported to be strongly associated with an increased risk of type 2 diabetes mellitus (T2DM) in different ethnic populations. The aim of the present study was to investigate whether rs7903146 and rs7085532 polymorphisms in TCF7L2 are associated with susceptibility to T2DM in Chinese Uygur population. We analyzed two single-nucleotide polymorphisms (SNPs) (rs793146, and rs7085532) in 927 Uygur subjects with T2DM and 955 control subjects. Genotyping was performed by Sequenom Massarray platform. The distribution of rs7903146 genotypes showed a significant difference between patients with T2DM and control participants ($P = 0.008$). In logistic regression analysis, the rs7903146 polymorphism was associated with the risk of developing T2DM in the dominant model after adjustment for potential confounders. General linear model analysis showed rs7903146 was independently associated with increased glucose levels by analyses of the dominant model and additive model after adjustment for covariates ($P = 0.005$ and $P = 0.018$, respectively). Our study indicates that rs7903146 polymorphism may play a role in T2DM susceptibility in Chinese Uygur population and is functionally associated with glucose levels, suggesting a mechanistic link between rs7903146 polymorphism and T2DM susceptibility.

Keywords: TCF7L2, Chinese Uygur population, single nucleotide polymorphism, type 2 diabetes mellitus, haplotype

Introduction

Type 2 diabetes mellitus (T2DM) is a group of metabolic diseases characterized by hyperglycemia due to a progressive insulin secretory defect with insulin resistance [1]. To date, T2DM affects 285 million individuals around the world, whereas its prevalence is increasing rapidly attributable to increase age of the population and surge of obesity in several countries [2]. T2DM is a complex metabolic disease that results from the combination of genetic and environmental factors [3]. The precise mechanisms of T2DM are not clear, but genetic susceptibility plays an important role in the etiology and manifestation of the T2DM [4, 5]. Recently, several genetic polymorphisms have found to be significantly associated with T2DM in different ethnic populations. These contain the Pro12Ala polymorphism in the PPARG gene, the E23K polymorphism in the KCNJ11 gene and the genetic variants of the CAPN10 gene [6-8]. Among them, the transcription factor 7-like 2 gene (TCF7L2) has shown the strongest association with T2DM. It has been demonstrated that TCF7L2 encodes for a transcription factor that is expressed in fetal pancreas and involved in a Wnt signaling pathway through regulation of the hormone glucagon-like peptide (GLP-1), which has an essential role in glucose homeostasis [9]. In 2006, Grant et al. [10] identified a microsatellite marker DG10S478 in intron 3 of the TCF7L2 that confirmed a significant association with T2DM in Icelandic, Danish, and American cohorts. The authors then genotyped the five SNPs (rs12255372, rs7903146, rs7901695, rs11196205, and rs7895340) and found association between all

five SNPs and T2DM in all three cohorts. After the original research, numerous studies have demonstrated the association between TCF7L2 and T2DM in European, Asian and American populations [11-22]. Furthermore, the polymorphisms in TCF7L2 gene have been investigated in epidemiology studies of Chinese Han population, the research has been mainly in the south of China. However, the relationship between TCF7L2 gene polymorphisms and T2DM remains unclear in the Uygur populations of western China. In this context, it is reasonable to estimate that TCF7L2 may also play a critical role in genetic susceptibility to T2DM in Chinese Uygur population.

Therefore, the aim of the present study was to investigate whether rs7903146 and rs7085532 polymorphisms in TCF7L2 are associated with susceptibility to T2DM in Chinese Uygur population.

Materials and methods

Subjects

A total of 1882 unrelated subjects were enrolled in this case-control study. All participants were of Western Uygur Chinese ancestry and were recruited among local inhabitants of Xinjiang Province. The patient population consists of 927 subjects (578 men and 349 women) with T2DM. The T2DM patients were recruited from the outpatient clinic of the First Affiliated Hospital of Xinjiang Medical University. T2DM patients were diagnosed based on the criteria of the American Diabetes Association with fasting plasma glucose ≥ 126 mg/dl or 2-h plasma glucose ≥ 200 mg/dl during an oral glucose tolerance test. The control group included 955 healthy subjects (604 men and 351 women) were recruited from routine health examinations. The study consists of interviews, physical examinations, and data from blood sample analyses. All information and data regarding hypertension, coronary artery disease, hyperlipidemia were collected from all participants. Anthropometric measurements including weight and height were obtained using standardized techniques. The body mass index (BMI) was calculated as the weight in kilograms divided by the square of height in meters (kg/m^2). We excluded participants with impaired renal function, malignancy, connective-tissue disease, or chronic inflammatory disease. The present study was approved by the ethics committee of the First Affiliated Hospital of Xinjiang Medical University and informed consent was obtained from all participants before data collection, and the study was conducted according to the standards of the Declaration of Helsinki.

DNA extraction and genotyping

Blood samples were collected from the study participants with informed consent after overnight fasting. Genomic DNA was extracted from blood samples using the DNA extraction Kit (Beijing Biotech Company Limited, Beijing, China) according to the manufacturer's protocol and stored at -80°C until utilized. The quantity and quality of the extracted DNA were measured using NanoDrop2000 Spectrophotometer (Thermo Scientific, USA). Based on International HapMap Project Phase II database of the Chinese population (http://www.hapmap.org) and haploview version 4.2 software, we selected two tagSNPs (rs7903146, rs7085532) in the TCF7L2 gene. Haploview version 4.2 software was selected to determine the tagSNPs with the criteria of minor allele frequency (MAF) ≥ 0.05 in the Chinese populations and a pairwise linkage disequilibrium squared correlation coefficient (r^2) ≥ 0.80. The genotyping for single nucleotide polymorphisms (SNPs) was carried out using the Sequenom MassARRAY system according to the manufacturer's protocol. To ensure the accuracy of the genotyping, 10% samples were randomly selected and genotyped in duplicate with 100% concordance. Call rate for genotyping were 100%.

Biochemical measurements

The plasma concentrations of glucose, triglyceride (TG), total cholesterol (TC), high density lipoprotein (HDL), low density lipoprotein (LDL), uric acid (UA), creatinine (Cr) and blood urea nitrogen (BUN) were detected using standard methods by the Central Laboratory Department of the First Affiliated Hospital of Xinjiang Medical University.

Statistical analysis

Continuous data were expressed as means ± standard deviations. Categorical data are described with number (percentage). The chi-square test was used to determine whether the genotype distributions were in Hardy-Weinberg equilibrium. Clinical and laboratory characteris-

Table 1. Demographic and clinical characteristics of study participants

	Total			Men			Women		
	T2DM	Control	*p* value	T2DM	Control	*p* value	T2DM	Control	*p* value
Number (n)	927	955		578	604		349	351	
age (years)	51.19±9.77	50.41±9.82	0.083	51.10±9.94	50.56±9.92	0.348	51.34±9.49	50.15±9.64	0.100
BMI (kg/m^2)	27.70±5.04	27.05±3.94	0.027*	27.79±4.96	27.00±3.56	0.041*	27.56±5.17	27.14±4.56	0.449
SBP (mmHg)	126.85±19.11	122.73±17.62	0.015*	125.11±16.53	123.93±18.83	0.541	129.61±22.37	120.81±15.46	< 0.001*
DBP (mmHg)	79.30±11.97	81.29±14.05	0.074	78.79±10.90	82.86±15.12	0.003*	80.11±13.47	78.67±11.72	0.455
Glu (mmol/L)	9.59±3.45	5.00±0.93	< 0.001*	9.66±3.38	4.99±0.88	< 0.001*	9.47±3.57	5.03±1.01	< 0.001*
TG (mmol/L)	2.43±2.18	2.44±2.06	0.928	2.68±2.50	2.64±2.21	0.787	2.03±1.41	2.09±1.72	0.579
TC (mmol/L)	4.68±1.38	4.31±1.69	< 0.001*	4.65±1.46	4.24±1.67	< 0.001*	4.73±1.24	4.42±1.72	0.009*
HDL (mmol/L)	0.96±0.34	1.25±0.33	< 0.001*	0.91±0.29	1.18±0.30	< 0.001*	1.03±0.41	1.38±0.33	< 0.001*
LDL (mmol/L)	2.89±1.50	3.00±0.82	0.065	2.89±1.78	3.00±0.78	0.209	2.88±0.85	2.99±0.89	0.102
UA (μmol/L)	267.17±84.03	283.08±70.51	< 0.001*	285.93±85.00	307.79±61.57	< 0.001*	236.43±72.71	240.44±64.41	0.456
Cr (μmol/L)	68.22±41.08	70.95±18.08	0.083	74.39±40.14	76.13±13.90	0.357	58.06±40.64	62.14±20.78	0.117
BUN (mmol/L)	5.20±2.33	4.95±1.44	0.011*	5.42±2.39	5.08±1.36	0.005*	4.84±2.18	4.74±1.56	0.519

*$P < 0.05$.

Table 2. Genotype and allele distributions in patients with T2DM and control participants

Variants	Total			Men			Women		
	T2DM n (%)	Control n (%)	*p* value	T2DM n (%)	Control n (%)	*p* value	T2DM n (%)	Control n (%)	*p* value
rs7903146									
Genotyping									
CC	495 (53.4)	576 (60.3)		306 (52.9)	380 (62.9)		189 (54.2)	196 (55.8)	
CT	311 (33.5)	265 (27.8)		195 (33.7)	170 (28.1)		116 (33.2)	95 (27.1)	
TT	121 (13.1)	114 (11.9)	0.008*	77 (13.3)	54 (8.9)	0.001*	44 (12.6)	60 (17.1)	0.097
Dominant model									
CC	495 (53.4)	576 (60.3)		306 (52.9)	380 (62.9)		189 (54.2)	196 (55.8)	
CT+TT	432 (46.6)	379 (39.7)	0.002*	272 (47.1)	224 (37.1)	0.001*	160 (45.8)	155 (44.2)	0.654
Recessive model									
TT	121 (13.1)	114 (11.9)		77 (13.3)	54 (8.9)		44 (12.6)	60 (17.1)	
CC+CT	806 (86.9)	841 (88.1)	0.464	501 (86.7)	550 (91.1)	0.016*	305 (87.4)	291 (82.9)	0.095
Additive model									
CC	495 (53.4)	576 (60.3)		306 (52.9)	380 (62.9)		189 (54.2)	196 (55.8)	
CT	311 (33.5)	265 (27.8)		195 (33.7)	170 (28.1)		116 (33.2)	95 (27.1)	
TT	121 (13.1)	114 (11.9)	0.014*	77 (13.3)	54 (8.9)	< 0.001*	44 (12.6)	60 (17.1)	0.613
Allele									
C	1301 (70.2)	1417 (74.2)		807 (69.8)	930 (77.0)		494 (70.8)	487 (69.4)	
T	553 (29.8)	493 (25.8)	0.006*	349 (30.2)	278 (23.0)	< 0.001*	204 (29.2)	215 (30.6)	0.567
rs7085532									
Genotyping									
AA	363 (39.2)	358 (37.5)		248 (42.9)	231 (38.2)		115 (33.0)	127 (36.2)	
AG	424 (45.7)	444 (46.5)		243 (42.0)	276 (45.7)		181 (51.9)	168 (47.9)	
GG	140 (15.1)	153 (16.0)	0.720	87 (15.1)	97 (16.1)	0.263	53 (15.2)	56 (16.0)	0.561
Dominant model									
AA	363 (39.2)	358 (37.5)		248 (42.9)	231 (38.2)		115 (33.0)	127 (36.2)	
AG+GG	564 (60.8)	597 (62.5)	0.456	330 (57.1)	373 (61.8)	0.103	234 (67.0)	224 (63.8)	0.369
Recessive model									
GG	140 (15.1)	153 (16.0)		87 (15.1)	97 (16.1)		53 (15.2)	56 (16.0)	
AA+AG	787 (84.9)	802 (84.0)	0.583	491 (84.9)	507 (83.9)	0.633	296 (84.8)	295 (84.0)	0.779
Additive model									
AA	363 (39.2)	358 (37.5)		248 (42.9)	231 (38.2)		115 (33.0)	127 (36.2)	
AG	424 (45.7)	444 (46.5)		243 (42.0)	276 (45.7)		181 (51.9)	168 (47.9)	
GG	140 (15.1)	153 (16.0)	0.421	87 (15.1)	97 (16.1)	0.168	53 (15.2)	56 (16.0)	0.633
Allele									
A	1150 (62.0)	1160 (60.7)		739 (63.9)	738 (61.1)		411 (58.9)	422 (60.1)	
G	704 (38.0)	750 (39.3)	0.415	417 (36.1)	470 (38.9)	0.155	287 (41.1)	280 (39.9)	0.639

*$P < 0.05$.

Table 3. Multiple logistic regression analysis for T2DM patients and control subjects (rs7903146)

	Total			Men			Women		
	OR	95% CI	*p* value	OR	95% CI	*p* value	OR	95% CI	*p* value
Dominant model (CC vs CT+TT)	1.376	1.103-1.715	0.005*	1.604	1.209-2.127	0.001*	1.021	0.711-1.467	0.909
TG	0.894	0.841-0.951	< 0.001*	0.896	0.836-0.960	0.002*	0.961	0.822-1.123	0.615
TC	1.514	1.347-1.701	< 0.001*	1.470	1.285-1.681	< 0.001*	1.674	1.307-2.144	< 0.001*
HDL	0.030	0.020-0.047	< 0.001*	0.017	0.009-0.032	< 0.001*	0.037	0.019-0.071	< 0.001*
LDL	0.756	0.631-0.905	0.002*	0.827	0.671-1.019	0.074	0.634	0.437-0.920	0.016*

*$P < 0.05$.

Table 4. Multiple logistic regression analysis for T2DM patients and control subjects (rs7085532)

	Total			Men			Women		
	OR	95% CI	*p* value	OR	95% CI	*p* value	OR	95% CI	*p* value
Dominant model (AA vs AG+GG)	0.869	0.695-1.087	0.218	0.752	0.567-1.012	0.161	1.014	0.693-1.483	0.944
TG	0.890	0.837-0.947	< 0.001*	0.890	0.830-0.954	0.001*	0.961	0.821-1.123	0.613
TC	1.511	1.345-1.698	< 0.001*	1.461	1.278-1.670	< 0.001*	1.673	1.306-2.143	< 0.001*
HDL	0.030	0.020-0.047	< 0.001*	0.017	0.009-0.031	< 0.001*	0.037	0.019-0.071	< 0.001*
LDL	0.758	0.633-0.908	0.003*	0.833	0.677-1.024	0.082	0.635	0.438-0.921	0.017*

*$P < 0.05$.

Table 5. Glucose levels and TCF7L2 genotypes

SNP Genotypes	Unadjusted model		Adjusted model§	
	Glucose (mmol/L) Mean ± SD	*p* value	Glucose (mmol/L) Mean ± SE	*p* value
rs7903146				
Additive model				
CC (n = 1071)	7.05±3.39		6.95±0.10	
CT (n = 576)	7.52±3.48		7.43±0.14	
TT (n = 235)	7.35±3.10	0.027*	7.32±0.22	0.018*
Dominant model				
CC (n = 1071)	7.05±3.39		6.95±0.10	
CT+TT (n = 811)	7.47±3.38	0.009*	7.40±0.12	0.005*
Recessive model				
TT (n = 235)	7.35±3.10		7.32±0.22	
CC+CT (n = 1647)	7.22±3.43	0.584	7.12±0.08	0.394
rs7085532				
Additive model				
AA (n = 721)	7.16±3.29		7.06±0.13	
AG (n = 868)	7.34±3.52		7.26±0.12	
GG (n = 293)	7.12±3.26	0.477	7.02±0.20	0.417
Dominant model				
AA (n = 721)	7.16±3.29		7.06±0.13	
AG+GG (n = 1161)	7.28±3.45	0.443	7.20±0.10	0.395
Recessive model				
GG (n = 293)	7.12±3.26		7.02±0.20	
AA+AG (n = 1589)	7.25±3.41	0.529	7.17±0.09	0.507

§Analysis of covariance adjusted for TG, TC, HDL and LDL levels; *$P < 0.05$.

tics were compared between the T2DM patients and the control groups by using independent samples *t* test. The differences between the T2DM patients and control groups in terms of genotype distribution or allele frequency were compared using chi-square test. We also performed multiple logistic regression analysis adjusted for TG, TC, HDL, and LDL under a dominant model. General linear model (GLM) analysis was performed to test for associations between SNP genotypes and serum glucose after adjustment for confounding variables. The statistical analyses for haplotype and linkage disequilibrium coefficients were performed using the software SHEsis [23]. SPSS 16.0 software for Windows (SPSS Institute, Chi-

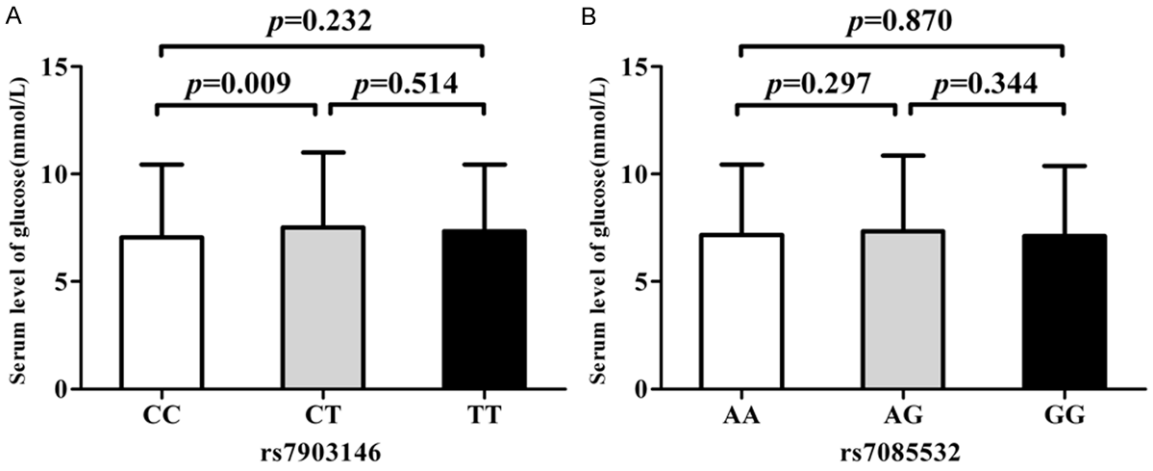

Figure 1. Association of glucose levels with rs7903146 and rs7085532 genotypes in study participants. A. Mean plasma levels of glucose in subjects according to rs7903146 genotypes. CC vs CT: 7.05±3.39 vs 7.52±3.48, P = 0.009. CC vs TT: 7.05±3.39 vs 7.35±3.10, P = 0.232. CT vs TT: 7.52±3.48 vs 7.35±3.10, P = 0.514. B. Mean plasma levels of glucose in subjects according to rs7085532 genotypes. AA vs AG: 7.16±3.29 vs 7.34±3.52, P = 0.297. AA vs GG: 7.16±3.29 vs 7.12±3.26, P = 0.870. AG vs GG: 7.34±3.52 vs 7.12±3.26, P = 0.344.

Table 6. Haplotype analysis in patients with T2DM and in control subjects

Haplotype	rs7903146	rs7085532	Frequency in Total			Frequency in Men			Frequency in Women		
			T2DM	Control	*p* value	T2DM	Control	*p* value	T2DM	Control	*p* value
H1	C	A	0.420	0.422	0.173	0.432	0.454	0.277	0.399	0.419	0.459
H2	C	G	0.282	0.300	0.221	0.266	0.315	0.008*	0.309	0.275	0.169
H3	T	A	0.201	0.166	0.006*	0.207	0.157	0.001*	0.190	0.183	0.731
H4	T	G	0.098	0.093	0.588	0.095	0.074	0.063	0.103	0.124	0.212

*P < 0.05.

cago, IL, USA) was used for all statistical analysis. A P value < 0.05 was considered statistically significant.

Results

The demographic and clinical characteristics of the T2DM patients (n = 927) and control participants (n = 955) are summarized in **Table 1**. In the total participants, the following variables were significantly different between T2DM patients and control subjects: BMI, SBP, the serum levels of glucose, TC, BUN, HDL, BUN and UA (P < 0.05). There was no significant difference in age, DBP, the serum levels of TG, LDL, creatinine between the two groups (all P > 0.05). In the men participants, the following variables were significantly different between T2DM patients and control subjects: BMI, DBP, the serum levels of glucose, TC, HDL, UA and BUN (P < 0.05). There was no significant difference in age, SBP, the serum levels of TG, LDL, creatinine between the two groups (all P > 0.05). In the women participants, the following variables were significantly different between T2DM patients and control subjects: SBP, the serum levels of glucose, TC and HDL (P < 0.05). There was no significant difference in age, BMI, DBP, the serum levels of TG, LDL, UA, creatinine and BUN between the two groups (all P > 0.05).

The distribution of genotypes and alleles of rs7903146 and rs7085532 of TCF7L2 gene analysis are shown in **Table 2**. The genotypic distribution in both the control and the T2DM groups was in Hardy-Weinberg equilibrium (data not shown). For total participants, the distribution of rs7903146 genotypes showed a significant difference between patients with T2DM and control participants (P = 0.008). In addition, the distribution of the dominant model (CC vs CT+TT) and T allele frequency of rs7903146 were significantly higher among T2DM patients when compared to control subjects (46.6% vs 39.7%, 29.8% vs 25.8%). For men, the distribu-

tion of rs7903146 genotypes showed a significant difference between patients with T2DM and control participants (P = 0.001). Moreover, the distribution of the dominant model (CC vs CT+TT), recessive model (TT vs CC+CT), and T allele frequency of rs7903146 were significantly higher among T2DM patients when compared to control subjects (47.1% vs 37.1%, 13.3% vs 8.9%, 30.2% vs 23.0%). However, no difference was found in women. For total, men, and women subjects, the distribution of rs7085532 genotypes, dominant model (AA vs AG+GG), recessive model (GG vs AA+AG), additive model (AA vs AG vs GG) and allele frequency did not show a significant difference between the T2DM patients and control subjects (P > 0.05, respectively).

Logistic regression analysis with adjustment for the serum concentration of TG, TC, HDL, LDL was performed using T2DM as the dependent variable and the genotypes as the independent variables. For total participant, the rs7903146 polymorphism was associated with the risk of developing T2DM in the dominant model after adjust ment for potential confounders, individuals with CT+TT genotype in rs7903146 of the TCF7L2 gene had 1.376-fold increased risk of developing T2DM than individuals with CC genotypes (OR = 1.376, 95% CI 1.103 to 1.715, P = 0.005). For men, individuals with CT+TT genotype had 1.604-fold increased risk of developing T2DM than individuals with CC genotypes (OR = 1.604, 95% CI 1.209 to 2.127, P = 0.001) (**Table 3**). However, no significant differences were observed in the association between rs7085532 polymorphisms and T2DM among total, men, and women subjects (**Table 4**).

Table 5 shows that rs7903146 was associated with glucose levels by analyses of an additive model (P = 0.027) and a dominant model (P = 0.009), and the difference remained significant after adjustment for TG, TC, HDL and LDL levels (P = 0.018, P = 0.005, respectively). However, these associations were not found in rs7085532 before and after adjustment of confounders. When we compared plasma glucose levels according to the genotypes of rs7903146 in study participants, the mean plasma concentration of glucose was significantly higher in subjects with CT genotype than in subjects with CC genotype (7.52±3.48 mmol/L vs 7.05±3.39 mmol/L, P = 0.009) (**Figure 1A**). However, these differences were not observed in the rs7085532 (**Figure 1B**).

Table 6 shows the result of haplotype analysis. In the haplotype-based case-control analysis of the Uygur population, the haplotypes were established through the use of rs7903146 and rs7085532. For total participants and men, the frequency of the T-A haplotype was significantly higher for the T2DM patients as compared to the control subjects (total: P = 0.006; men: P = 0.001). For men, the frequency of the C-G haplotype was significantly lower for T2DM patients than for control participants (P = 0.008). For women, the overall distribution of haplotypes were not significantly different between the T2DM patients and the control subjects (P > 0.05).

Discussion

The important finding of present study is that the rs7903146 polymorphism was found to be significantly associated with T2DM in this Uygur population in western China.

The TCF7L2 gene encodes a transcription factor involved in the Wnt signaling pathway, which plays a critical role in cell development and regulatory mechanisms [24]. Because of glucagon-like peptide 1 (GLP-1) and insulin plays an important role in blood glucose homeostasis, it was supposed that TCF7L2 polymorphisms may affect T2DM susceptibility by indirectly altering GLP-1 levels [25]. Little is known about the physiological mechanisms of TCF7L2 in glucose homeostasis. It has been suggested that TCF7L2 mainly affects the risk of T2DM by influencing insulin secretion. The identification of genetic polymorphisms influencing T2DM is an important focus of study to improve the understanding of the mechanisms underlying the pathogenesis of this disorder. In this scenario, Grant *et al.* [10] initially reported that a strong association with T2DM in a tetranucleotide microsatellite marker (DG10S478) located in intron 3 of TCF7L2. Additionally, five SNPs (rs12255372, rs7903146, rs7901695, rs11196205, and rs7895340) correlated with DG10S478 clustered within the same linkage disequilibrium (LD) block spanning from intron 3 to intron 4 showed a significant association with T2DM. Following the initial study, there have been a considerable number association studies among different populations. Kimber *et al.* [11] suggested that TCF7L2 polymorphisms may be associated with increased disease severity and therapeutic failure in UK, the CT genotype of rs7903146 having an OR of 1.36

for type 2 diabetes and the TT genotype having an OR of 2.03. Dahlgren *et al.* [12] confirmed that polymorphisms of the TCF7L2 gene are associated with beta cell dysfunction and confer an increased risk of T2DM in the cohort of Swedish elderly men. The highest significant odds ratio was found for rs7903146 was 2.15 (95% CI: 1.20-3.85). The odds ratio for rs12255372 was 1.69 (95% CI: 1.20-2.39). González-Sánchez *et al.* [13] reported that TCF7L2 gene is a major determinant of T2DM risk in Spain, the T allele of the rs7903146 was significantly associated with a greater OR for T2DM (OR = 1.29, 95% confidence interval [CI]: 1.06-1.57, *P* = 0.01). This T allele was also associated with an increased proinsulin: insulin ratio after OGTT shows that TCF7L2 may be involved in insulin synthesis and processing. Damcott *et al.* [14] found that polymorphisms in the TCF7L2 gene are associated with T2DM in the Amish, the genotype frequencies of rs7903146 differed significantly between T2DM subjects and control subjects (P = 0.008). Scott *et al.* [15] demonstrated that variants in TCF7L2 are associated with increased risk of T2DM in a Finnish Sample, the strongest results were for rs12255372 (OR = 1.36, 95% CI: 1.15-1.61, *P* = 0.00026) and rs7903146 (OR = 1.33, 95% CI: 1.14-1.56, *P* = 0.00042). Magellan *et al.* [16] suggested that rs7903146 polymorphism of the TCF7L2 was associated with T2DM in this Cameroonian population (*P* = 0.013 for alleles and *P* = 0.013 for genotypes). Regarding the Asian population. In the study of Horikoshi *et al.* [17] and Hayashi *et al.*, [18] rs7903146 showed a significant association with T2DM in the Japanese population, these results suggest that TCF7L2 may be a strong candidate for conferring susceptibility to T2DM across different ethnic groups. Chandak *et al.* [19] reported that TCF7L2 is an important gene for determining susceptibility to T2DM in the Indian population. They observed a strong association with all the polymorphisms, including rs12255372, rs4506565 and rs7903146. ($P = 4.0\times10^{-5}$, $P = 2.0\times10^{-5}$, $P = 3.0\times10^{-5}$, respectively). Unfortunately, contradictory results were observed in Chinese Han populations. Ren *et al.* [20] showed that no significant associations were found between the TCF7L2 and T2DM in Chinese Han population. Chang *et al.* [21] identified a novel association of the rs290487 polymorphism of the TCF7L2 gene with T2DM in a Chinese population. However, they did not find any association of the rs7904136 and rs12255372 with T2DM. In the study of Ng *et al.*, [22] the frequency of the risk alleles of rs11196205 and rs7903146 were rare in Chinese Han population (0.013 and 0.024, respectively), the association for rs7903146 was not significant in the case-control study. Interestingly, the rs1196205 and rs1196218 were associated with T2DM. Although T2DM association studies have reported that the T allele of the rs7903146 at the TCF7L2 is strongly associated with T2DM risk in different populations, it should be noted that the frequency of the T allele of the rs7903146 at the TCF7L2 was found to be different in the various populations. In addition, unlike European populations, the frequency of T allele of rs7903146 is relatively low in Asian populations, including the Japanese and Chinese Han populations, suggesting genetic heterogeneity among different ethnicities. These finding raised the question of whether these variants are major contributors to T2DM in the Chinese Uygur population.

In our study, it was found that there was a significant difference in the genotype distribution of rs7903146 between patients with T2DM and control subjects in the total population. When analyzing men and women separately, T allele frequency of rs7903146 was higher among men T2DM patients when compared to men control subjects. This indicates that the risk of T2DM was increased in men carrying the T allele. For men, the distribution of the dominant model (CC vs CT+TT) was significantly higher in patients with T2DM than that in control subjects. After multivariate adjustment of the confounding factors for T2DM, the significant difference was retained, which indicated that men carrying the CT+TT genotype of rs7903146 may have a higher risk of T2DM. In the present study, no significant differences in allele frequency or genotype distribution of rs7085532 were observed between patients with T2DM and control subjects. In addition, we hypothesized that haplotype analysis would be useful for the assessment of association between haplotypes and T2DM. For total participants, we found a susceptible haplotype T-A. For men, we found a susceptible haplotype T-A and a protective haplotype C-G, and these haplotypic analysis results were consistent with genotypic analysis results of rs7903146, which showed that the T allele could be a susceptible genetic marker and the C allele could be a protective genetic marker of T2DM. For women, the over-

all distribution of haplotypes was not significantly different between the T2DM patients and the control subjects ($P > 0.05$).

When we analyzed glucose according to the genotypes, we found that the rs7903146 polymorphism is associated with glucose concentration. Compared with CC genotype, individuals with CT genotype had significantly higher glucose levels. This association remains significant after adjustment for TG, TC, HDL and LDL levels, indicating the association of CT genotype with T2DM risk may be in part mediated by its impact on plasma glucose level which may provide a mechanistic link between rs7903146 polymorphism and T2DM susceptibility in Chinese Uygur population. There was no difference in plasma concentrations of glucose between different genotypes of rs7085532 in study participants, indicating that the association between glucose plasma levels and rs7085532 polymorphism may be non-causal.

We have found that rs7903146 of TCF7L2 are associated with susceptibility to T2DM in the Chinese Uygur population. Our results are consistent with previous reports for Caucasian [10-15] and Japanese populations [17, 18], but not with other reports for Chinese Han populations [20-22]. No significant associations were found between the rs7903146 and T2DM in Chinese Han populations. The T allele frequency of rs7903146 in the samples from a Chinese Uygur population (0.258) was substantially higher than that of the previously reported three samples from Chinese Han populations, in which T allele frequency ranged from 0.023 to 0.030 [20-22]. The apparent difference in the association of rs7903146 in Chinese Han populations could be due to the low frequencies of the rs7903146 and the relatively small sample sizes used in their studies. Moreover, differences in populations such as race, geographical and environment factors, might explain these results between Chinese Uygur and Han population.

There are several limitations in the present study that should be considered. First, selection bias may occur, because it was a hospital-based case-control study that the T2DM cases and controls were recruited from hospital. It is crucial to evaluate these observations in a population-based prospective study. Second, since our study was restricted to Uygur Chinese, our data were not representative of adults throughout China. Therefore, future studies in larger sample size and different ethnicities are warranted. Third, environmental factors, which may demonstrate the non-genetic factors on the risk of T2DM, further studies are warranted to confirm the contribution of gene-environment interaction on T2DM.

In summary, the present study demonstrated an association between the rs7903146 polymorphism and T2DM risk in Chinese Uygur population. Individuals with the CT and TT genotype may have a higher risk of T2DM than those with the CC genotype. Moreover, further studies with larger sample size should be performed in Uygur population to corroborate the precise mechanism of TCF7L2 gene in insulin secretion.

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Disclosure of conflict of interest

None.

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