Genetics and Molecular Research

Association between transcription factor 7-like 2 genetic polymorphisms and development of type 2 diabetes in a Chinese population

H.Y. Jia, Q.Z. Li and L.F. Lv

Department of Endocrinology, Henan Provincial People's Hospital, Zhengzhou, China

Corresponding author: H.Y. Jia
E-mail: jiahaiyya@163.com

Genet. Mol. Res. 15 (2): gmr.15028223
Received December 7, 2015
Accepted April 4, 2016
Published June 10, 2016
DOI http://dx.doi.org/10.4238/gmr.15028223

ABSTRACT. We conducted a hospital-based case-control study to evaluate the relationship between the transcription factor 7-like 2 (*TCF7L2*) rs7903146 polymorphism and type 2 diabetes mellitus risk in a Chinese population. Genotyping of *TCF7L2* rs7903146 was carried out using the polymerase chain reaction-restriction fragment length polymorphism method. A chi-square test revealed a statistically significant difference between the distributions of rs7903146 genotypes in type 2 diabetes mellitus patient and control groups (chi-square = 10.49, P = 0.005). Using unconditional logistic regression analysis, we observed that the TT genotype of this polymorphism was significantly correlated with increased risk of developing type 2 diabetes mellitus compared to the CC genotype [odds ratio (OR) = 2.31, 95% confidence interval (CI) = 1.33-4.04]. Furthermore, we found that the rs7903146 sequence variation was also significantly associated with susceptibility to this disease under dominant (OR = 1.58, 95%CI = 1.09-2.28) and recessive models

 ©FUNPEC-RP www.funpecrp.com.br

(OR = 2.11, 95%CI = 1.25-3.62). We conclude that the *TCF7L2* rs7903146 genetic polymorphism is independently associated with the risk of developing type 2 diabetes mellitus under co-dominant, dominant, and recessive models.

Key words: *TCF7L2*; rs7903146; Type 2 diabetes mellitus

INTRODUCTION

Type 2 diabetes mellitus is a chronic and complex disease characterized by insulin resistance and impaired insulin secretion, and represents a serious global health problem associated with high morbidity. It is estimated that there are approximately 92.4 million adults with type 2 diabetes mellitus and 148.2 million with prediabetes in China (Yang et al., 2010; Cai et al., 2013; Fu et al., 2013; Jiang et al., 2013). The etiology of this disease is not well understood. Environmental and genetic factors both contribute to the development of type 2 diabetes mellitus (Mrozikiewicz-Rakowska et al., 2015; Pearson, 2015). Previous genome-wide association studies have implicated many loci in the onset, prognosis, and severity of type 2 diabetes mellitus (Adeyemo et al., 2015; Wei et al., 2015).

Transcription factor 7-like 2 (*TCF7L2*) is located on chromosome 10q25.3 and is 216.86 kb in length. *TCF7L2* expression is related to its proposed function in insulin secretion. Several previous studies have examined the role of the *TCF7L2* rs7903146 polymorphism in the development of type 2 diabetes mellitus, but their results have been inconclusive (Assmann et al., 2014; Barros et al., 2014; Allahdini et al., 2015; Guewo-Fokeng et al., 2015; Liu et al., 2015; Nanfa et al., 2015; Yao et al., 2015; Yang et al., 2015). In this study, we conducted a hospital-based case-control investigation to evaluate the relationship between *TCF7L2* rs7903146 and risk of developing type 2 diabetes mellitus in a Chinese population.

MATERIAL AND METHODS

Subjects

Between January 2013 and December 2014, 248 patients with type 2 diabetes mellitus and 267 control subjects without the disease were consecutively recruited from the Department of Endocrinology of Henan Provincial People's Hospital. Type 2 diabetes mellitus was diagnosed based on the World Health Organization-International Diabetes Federation criteria (WHO-IDF, 2006). Control subjects consisted of individuals who attended our hospital for a regular health examination, and all were confirmed as not having type 2 diabetes mellitus. Individuals with a history of diabetes, metabolic diseases or cancers were excluded.

Lifestyle and dietary data were collected using a self-designed questionnaire, and included sex, age, body mass index (BMI), and tobacco and alcohol consumption. Clinical data were gathered from medical record including hypertension, blood glucose, insulin and etc. The study was performed with the permission of the Institutional Review Board of Henan Provincial People's Hospital. Written informed consent was given by all subjects before participation. This study conformed to the Declaration of Helsinki.

Genotyping methods

A peripheral blood sample (5 mL) was obtained from each participan. DNA was extracted from these samples using a QIAGEN Blood Mini Kit (QIAGEN, Hilden, Germany). Genotyping of *TCF7L2* rs7903146 was carried out using the polymerase chain reaction (PCR)-restriction fragment length polymorphism method with the following primers: 5'-CTG AAC AAT TAG AGA GCT AAG CAC TTT TTA GGT A-3' (forward) and 5'-TTT CAC TAT GTA TTG TTG CCA GTC AGC AAA CAC-3' (reverse). The length of the amplified fragment was 266 bp. The cycling conditions were as follows: 94°C for 5 min; 35 cycles of 94°C for 5 s, 63°C for 30 s, and 72°C for 30 s; and a final extension step at 72°C for 10 min. The amplified products were digested with *Rsa*I restriction enzyme. To determine purity and integrity, PCR products were separated using 3% agarose gel electrophoresis and visualized by ultraviolet light.

Statistical analysis

Demographic, lifestyle, and clinical characteristics of type 2 diabetes mellitus patients and control subjects were compared by independent samples *t*-tests or chi-square tests. The distribution of *TCF7L2* rs7903146 genotypes in the two groups was analyzed using chi-square tests. Deviation of genotype distributions from Hardy-Weinberg equilibrium (HWE) was assessed in the patient and control groups using the goodness-of-fit chi-square test. The association between *TCF7L2* rs7903146 and type 2 diabetes mellitus was examined using logistic regression analyses, and odds ratios (OR) and 95% confidence intervals (95%CI) were calculated. Statistical analysis was conducted using SPSS 16.0 (SPSS Inc., Chicago, IL, USA). P values less than 0.05 were considered statistically significant.

RESULTS

The demographic and lifestyle characteristics of type 2 diabetes mellitus patients and control subjects are shown in Table 1. Chi-square tests showed that patients and controls were comparable in terms of age (chi-square = 0.15, P = 0.70), sex (chi-square = 1.51, P = 0.22), BMI (chi-square = 3.33, P = 0.07), tobacco smoking (chi-square = 0.45, P = 0.50), and alcohol consumption (chi-square = 0.28, P = 0.60). However, individuals with type 2 diabetes mellitus were more likely to develop hypertension (chi-square = 4.39, P = 0.04) and have higher levels of fasting glucose ($t = 5.73$, $P < 0.05$), fasting insulin ($t = 38.39$, $P < 0.05$), TC ($t = 23.46$, $P < 0.05$), TG ($t = 31.43$, $P < 0.05$), HDL-C ($t = 2.34$, $P < 0.05$), and LDL-C ($t = 17.30$, $P < 0.05$).

The distribution of *TCF7L2* rs7903146 genotypes in the two groups is presented in Table 2. In the type 2 diabetes mellitus patient group, 125 (50.40%), 73 (29.44%), and 50 (20.16%) individuals carried the CC, CT, and TT genotypes, respectively. In the control group, these genotypes were found in 165 (61.80%), 74 (27.72%), and 28 (10.49%) subjects, respectively. A chi-square test revealed a statistically significant difference between type 2 diabetes mellitus patients and control subjects (chi-square = 10.49, P = 0.005) regarding these distributions. *TCF7L2* rs7903146 genotype frequencies deviated from HWE in both study groups, with P values less than 0.001.

Table 1. Demographic and lifestyle characteristics of type 2 diabetes mellitus patients and control subjects.

Characteristic	Patients (N = 248)	%	Controls (N = 267)	%	Chi-square	P
Age, years						
<50	137	55.24	143	53.56		
≥50	111	44.76	124	46.44	0.15	0.70
Gender						
Female	90	36.29	111	41.57		
Male	158	63.71	156	58.43	1.51	0.22
BMI						
<24	148	59.68	180	67.42		
≥24	100	40.32	87	32.58	3.33	0.07
Tobacco smoking						
No	165	66.53	185	69.29		
Yes	83	33.47	82	30.71	0.45	0.50
Alcohol consumption						
No	157	63.31	175	65.54		
Yes	91	36.69	92	34.46	0.28	0.60
Hypertension						
No	192	77.42	226	84.64		
Yes	56	22.58	41	15.36	4.39	0.04
Fasting glucose (mM)		8.43 ± 2.47		4.82 ± 1.35	5.73	<0.05
Fasting insulin (mM)		59.12 ± 14.65		51.6 ± 15.11	38.39	<0.05
TC (mg/dL)		184.55 ± 17.35		154.53 ± 11.25	23.46	<0.05
TG (mg/dL)		174.26 ± 14.12		131.50 ± 16.55	31.43	<0.05
HDL-C (mg/dL)		47.82 ± 1.81		46.10 ± 11.42	2.34	<0.05
LDL-C (mg/dL)		134.26 ± 11.64		118.71 ± 8.63	17.30	<0.05

BMI = body mass index, TC = total cholesterol, TG = triglycerides, HDL-C = high-density lipoprotein cholesterol, LDL-C = low-density lipoprotein cholesterol.

Table 2. Distribution of *TCF7L2* rs7903146 polymorphism genotypes in type 2 diabetes mellitus patient and control groups.

Genotype	Patients	%	Controls	%	Chi-square	P value	P value for HWE	
							Patients	Controls
CC	125	50.40	165	61.80				
CT	73	29.44	74	27.72				
TT	50	20.16	28	10.49	10.49	0.005	<0.001	<0.001

HWE = Hardy-Weinberg equilibrium.

Using unconditional logistic regression analysis, we established that the TT genotype of *TCF7L2* rs7903146 was significantly correlated with increased risk of developing type 2 diabetes mellitus compared to the CC genotype, with an adjusted OR (and 95%CI) of 2.31 (1.33-4.04; Table 3). Furthermore, we observed that the *TCF7L2* rs7903146 genetic polymorphism was significantly associated with susceptibility to this disease under dominant (OR = 1.58, 95%CI = 1.09-2.28) and recessive models (OR = 2.11, 95%CI = 1.25-3.62).

Table 3. Association between the *TCF7L2* rs7903146 polymorphism and risk of type 2 diabetes mellitus.

Model	Patients	%	Controls	%	OR (95%CI)[1]	P value
Co-dominant						
CC	125	50.4	165	61.8	1.0 (Ref.)	-
CT	73	29.44	74	27.72	1.30 (0.86-1.98)	0.19
TT	50	20.16	28	10.49	2.31 (1.33-4.04)	0.001
Dominant						
CC	125	50.4	165	61.8	1.0 (Ref.)	-
CT+TT	123	49.6	102	38.2	1.58 (1.09-2.28)	0.01
Recessive						
CC+CT	198	79.84	239	89.51	1.0 (Ref.)	-
TT	50	20.16	28	10.49	2.11 (1.25-3.62)	0.003

[1]Adjusted for age, sex, hypertension, fasting glucose, fasting insulin, total cholesterol ($t = 23.46$, $P < 0.05$), triglycerides, and high- and low-density lipoprotein cholesterol. OR = odds ratio; CI = confidence interval, Ref. = reference.

DISCUSSION

In the present study, we investigated the role of the *TCF7L2* rs7903146 polymorphism in the pathogenesis of type 2 diabetes mellitus. Our findings indicated that the TT genotype of this variant significantly influences the development of this disease compared to the CC genotype, and that the *TCF7L2* rs7903146 polymorphism is associated with increased risk of type 2 diabetes mellitus under the dominant and recessive models in the Chinese population examined.

TCF7L2 is a member of Wnt signaling pathway, and plays an important role in the development of embryonic cellular (Cauchi et al., 2006). A previous experimental study has indicated that this pathway is closely associated with insulin secretion (Papadopoulou et al., 2011). Moreover, the *TCF7L2* gene is involved in modifying the expression of insulin and glucagon protein through Wnt signaling. A previous study has demonstrated a significant association between high expression of *TCF7L2* in pancreatic beta cells and glucose metabolism (Takamoto et al., 2014). Zhou et al. (2014) reported that the T allele of rs7903146, associated with diabetes risk, correlates with increased *TCF7L2* expression and reduced insulin levels and secretion, thus the affected protein may regulate insulin production and the pathogenesis of type 2 diabetes mellitus. In a study of a healthy Chinese population, Wang et al. (2015) found that *TCF7L2* genetic variation is significantly associated with fasting plasma glucose and triglyceride indexes.

To date, several studies have tested the association between *TCF7L2* genetic variations and risk of developing type 2 diabetes mellitus (Assmann et al., 2014; Barros et al., 2014; Ouhaibi-Djellouli et al., 2014; Guewo-Fokeng et al., 2015; Nanfa et al., 2015; Yang et al., 2015; Yao et al., 2015). Assmann et al. (2014), Guewo-Fokeng et al. (2015), and Ouhaibi-Djellouli et al. (2014) conducted investigations to assess the role of the *TCF7L2* rs7903146 polymorphism in the pathogenesis of this disease, reporting that this sequence variant is associated with susceptibility to type 2 diabetes mellitus in Caucasians. In studies of Chinese populations, Yao et al. (2015) and Yang et al. (2015) determined that the *TCF7L2* rs12255372 polymorphism may also contribute to the risk of developing this condition. However, some investigations have reached inconsistent conclusions (Barros et al., 2014; Pourahmadi et al., 2015). For instance, Barros et al. (2014) failed to establish a significant correlation between the *TCF7L2* rs7903146 variant and development of type 2 diabetes mellitus in Brazilian individuals. In addition, Pourahmadi et al. (2015) conducted a study of 200 patients and 200 control subjects from an Iranian population, in which they found no influence of the *TCF7L2* rs7903146 and rs12255372 polymorphisms on the incidence of this disease. The current work suggests that the *TCF7L2* rs7903146 variation may affect susceptibility to type 2 diabetes mellitus in a Chinese population.

In conclusion, we observed that the *TCF7L2* rs7903146 genetic polymorphism is independently associated with risk of developing type 2 diabetes mellitus under co-dominant, dominant, and recessive models. Further studies involving a greater number of ethnicities and larger sample sizes are greatly required to verify our findings.

Conflicts of interest

The authors declare no conflict of interest.

ACKNOWLEDGMENTS

We thank for the help from staffs for helping us collecting the blood samples in Henan Provincial People's Hospital.

REFERENCES

Adeyemo AA, Tekola-Ayele F, Doumatey AP, Bentley AR, et al. (2015). Evaluation of genome wide association study associated type 2 diabetes susceptibility loci in Sub Saharan Africans. *Front. Genet.* 6: 335. http://dx.doi.org/10.3389/fgene.2015.00335

Allahdini M, Kamalidehghan B, Akbari L, Azadfar P, et al. (2015). Prevalence of the rs7903146C>T polymorphism in *TCF7L2* gene for prediction of type 2 diabetes risk among Iranians of different ethnicities. *Drug Des. Devel. Ther.* 9: 5835-5841.

Assmann TS, Duarte GC, Rheinheimer J, Cruz LA, et al. (2014). The *TCF7L2* rs7903146 (C/T) polymorphism is associated with risk to type 2 diabetes mellitus in Southern-Brazil. *Arq. Bras. Endocrinol. Metabol.* 58: 918-925.

Barros CM, Araujo-Neto AP, Lopes TR, Barros MA, et al. (2014). Association of the rs7903146 and rs12255372 polymorphisms in the *TCF7L2* gene with type 2 diabetes in a population from northeastern Brazil. *Genet. Mol. Res.* 13: 7889-7898. http://dx.doi.org/10.4238/2014.September.29.1

Cai H, Cai J and Tao G (2013). Association of toll-like receptor 4 polymorphisms with type 2 diabetes mellitus. *APMIS* 121: 605-611. http://dx.doi.org/10.1111/apm.12027

Cauchi S, Meyre D, Dina C, Choquet H, et al. (2006). Transcription factor *TCF7L2* genetic study in the French population: expression in human beta-cells and adipose tissue and strong association with type 2 diabetes. *Diabetes* 55: 2903-2908. http://dx.doi.org/10.2337/db06-0474

Fu XD, Sun XQ, Wang HY, Guo JF, et al. (2013). Genetic polymorphisms of the *TLR4* gene and their association with susceptibility to type 2 diabetes mellitus in the Chinese population. *Genet. Mol. Res.* 12: 3813-3820. http://dx.doi.org/10.4238/2013.September.19.13

Guewo-Fokeng M, Sobngwi E, Atogho-Tiedeu B, Donfack OS, et al. (2015). Contribution of the *TCF7L2* rs7903146 (C/T) gene polymorphism to the susceptibility to type 2 diabetes mellitus in Cameroon. *J. Diabetes Metab. Disord.* 14: 26. http://dx.doi.org/10.1186/s40200-015-0148-z

Jiang ZS, Wang SX, Jia HX, Wang J, et al. (2013). Association of toll-like receptor 4 polymorphisms with type 2 diabetes mellitus. *Inflammation* 36: 251-257. http://dx.doi.org/10.1007/s10753-012-9541-7

Liu XH, Xie CG, An Y, Zhang XX, et al. (2015). Meta-analysis of the association between the rs7903146 polymorphism at the *TCF7L2* locus and type 2 diabetes mellitus susceptibility. *Genet. Mol. Res.* 14: 16856-16862. http://dx.doi.org/10.4238/2015.December.14.12

Mrozikiewicz-Rakowska B, Maroszek P, Nehring P, Sobczyk-Kopciol A, et al. (2015). Genetic and environmental predictors of chronic kidney disease in patients with type 2 diabetes and diabetic foot ulcer: a pilot study. *J. Physiol. Pharmacol.* 66: 751-761.

Nanfa D, Sobngwi E, Atogho-Tiedeu B, Noubiap JJ, et al. (2015). Association between the *TCF7L2* rs12255372 (G/T) gene polymorphism and type 2 diabetes mellitus in a Cameroonian population: a pilot study. *Clin. Transl. Med.* 4: 17. http://dx.doi.org/10.1186/s40169-015-0058-1

Ouhaibi-Djellouli H, Mediene-Benchekor S, Lardjam-Hetraf SA, Hamani-Medjaoui I, et al. (2014). The *TCF7L2* rs7903146 polymorphism, dietary intakes and type 2 diabetes risk in an Algerian population. *BMC Genet.* 15: 134. http://dx.doi.org/10.1186/s12863-014-0134-3

Papadopoulou A, Lynch KF, Shaat N, Håkansson R, et al.; DiPiS Study Group (2011). Gestational diabetes mellitus is associated with *TCF7L2* gene polymorphisms independent of HLA-DQB1*0602 genotypes and islet cell autoantibodies. *Diabet. Med.* 28: 1018-1027. http://dx.doi.org/10.1111/j.1464-5491.2011.03359.x

Pearson ER (2015). Dissecting the etiology of type 2 diabetes in the Pima Indian population. *Diabetes* 64: 3993-3995. http://dx.doi.org/10.2337/dbi15-0016

Pourahmadi M, Erfanian S, Moradzadeh M and Jahromi AS (2015). Non-association between rs7903146 and rs12255372 polymorphisms in transcription factor 7-like 2 gene and type 2 diabetes mellitus in Jahrom City, Iran. *Diabetes Metab. J.* 39: 512-517. http://dx.doi.org/10.4093/dmj.2015.39.6.512

Takamoto I, Kubota N, Nakaya K, Kumagai K, et al. (2014). *TCF7L2* in mouse pancreatic beta cells plays a crucial role in glucose homeostasis by regulating beta cell mass. *Diabetologia* 57: 542-553. http://dx.doi.org/10.1007/s00125-013-3131-6

Wang J, Yan G, Zhang J, Gao K, et al. (2015). Association of LRP5, TCF7L2, and GCG variants and type 2 diabetes mellitus as well as fasting plasma glucose and lipid metabolism indexes. *Hum. Immunol.* 76: 339-343. http://dx.doi.org/10.1016/j.humimm.2015.03.005

Wei FJ, Cai CY, Yu P, Lv J, et al. (2015). Quantitative candidate gene association studies of metabolic traits in Han Chinese type 2 diabetes patients. *Genet. Mol. Res.* 14: 15471-15481. http://dx.doi.org/10.4238/2015.November.30.25

WHO-IDF (2006). Definition and diagnosis of diabetes mellitus and intermediate hyperglycemia: report of a WHO/IDF

consultation. Available at [http://www.idf.org/webdata/docs/WHO_IDF_definition_diagnosis_of_diabetes.pdf]. Accessed October 1, 2014.

Yang SH, Dou KF and Song WJ (2010). Prevalence of diabetes among men and women in China. *N. Engl. J. Med.* 362: 1090-1101. http://dx.doi.org/10.1056/NEJMoa0908292

Yang Y, Xu JR, Wang YJ and Liu XM (2015). Association of *TCF7L2* gene polymorphisms with susceptibility to type 2 diabetes mellitus in a Chinese Hui population. *Genet. Mol. Res.* 14: 10064-10071. http://dx.doi.org/10.4238/2015.August.21.13

Yao H, Wang Z, Wang T, Ma Y, et al. (2015). Association of *TCF7L2* genetic polymorphisms with type 2 diabetes mellitus in the Uygur population of China. *Int. J. Environ. Res. Public Health* 12: 11797-11814. http://dx.doi.org/10.3390/ijerph120911797

Zhou Y, Park SY, Su J, Bailey K, et al. (2014). *TCF7L2* is a master regulator of insulin production and processing. *Hum. Mol. Genet.* 23: 6419-6431. http://dx.doi.org/10.1093/hmg/ddu359