Int J Clin Exp Pathol 2016;9(1):267-274
www.ijcep.com /ISSN:1936-2625/IJCEP0017808

Original Article

Association of genetic polymorphisms with pulmonary tuberculosis in a Chinese Tibetan population: a case-control study

Qunying Hu[1,2,3*], Mingxia Zhang[4*], Na He[1,2,3], Guinian Liang[5], Shilin Xu[5], Yuhe Wang[5], Fangping Mo[5], Hengxun Zhang[5], Longli Kang[1,2,3], Tianbo Jin[1,2,3,4]

[1]Key Laboratory of High Altitude Environment and Genes Related to Diseases of Tibet Autonomous Region, School of Medicine, Xizang Minzu University, Xianyang, Shaanxi, China; [2]Key Laboratory for Basic Life Science Research of Tibet Autonomous Region, School of Medicine, Xizang Minzu University, Xianyang, Shaanxi 712082, China; [3]Key Laboratory for Molecular Genetic Mechanisms and Intervention Research on High Altitude Disease of Tibet Autonomous Region, School of Medicine, Xizang Minzu University, Xianyang, Shaanxi 712082, China; [4]School of Life Sciences, Northwest University, Xi'an, Shaanxi, China; [5]Affiliated Hospital of Xizang Minzu University, Xianyang, Shaanxi, China. [*]Co-first authors.

Received October 13, 2015; Accepted November 28, 2015; Epub January 1, 2016; Published January 15, 2016

Abstract: Background: Pulmonary tuberculosis (TB) is a complex chronic infectious disease with both environmental and genetic factors contributing to its development. However, the genetic background of pulmonary TB remains unclear. Our study aimed to investigate the association between pulmonary tuberculosis genetic susceptibility variants and pulmonary tuberculosis in Chinese Tibetan population. Materials and methods: 20 single nucleotide polymorphisms (SNPs) were selected from previous genome-wide association studies. A case-control study was conducted to evaluate whether these variants are related to pulmonary tuberculosis susceptibility among 217 cases and 383 healthy controls. All these SNPs were genotyped using Sequenom Mass-ARRAY technology. For each SNP, genotypic frequencies in controls were tested for departure from Hardy-Weinberg Equilibrium (HWE) using an exact test. A *p*-value of 0.05 was considered the threshold for statistical significance. We compared the allele frequencies of cases and controls using the chi-squared (χ^2) test. Associations between the gene and the risk of pulmonary tuberculosis were tested using various genetic models (co-dominant, dominant, recessive, over-dominant, and additive) and analysis by SNP stats. Odds ratios and 95% confidence intervals (CIs) were calculated by unconditional logistic regression with adjustments for age and gender. Results: Using the χ^2 test, we found five SNPs had an increased risk of pulmonary tuberculosis, including rs6676375 (OR = 1.48; 95% CI = 1.04-2.10; *P* = 0.030), rs7821565 (OR = 1.51, 95% CI = 1.12-2.05; *P* = 0.007), rs17217757 (OR = 1.66; 95% CI = 1.21-2.27; *P* = 0.001), rs1900442 (OR = 1.28, 95% CI = 1.01-1.62; *P* = 0.041), and rs712039 (OR = 1.34, 95% CI = 1.06-1.70; *P* = 0.014). Additionally, we found five SNPs were significantly associated with reduce risk of pulmonary tuberculosis, including rs2505675 (OR = 0.78; 95% CI = 0.61-1.00; *P* = 0.048), rs586716 (OR = 0.67; 95% CI = 0.47-0.96; *P* = 0.027), rs1434579 (OR = 0.63; 95% CI = 0.47-0.84; *P* = 0.002), rs2837857 (OR = 0.66; 95% CI = 0.49-0.89; *P* = 0.006), and rs6538140 (OR = 0.64; 95% CI = 0.50-0.82; *P* = 0.000). Conclusion: Our study provides the first reported data of a possible association between the SNPs rs2505675, rs6538140, rs5867716, rs586716, and rs6676375 and pulmonary TB in a Chinese Tibetan population.

Keywords: Single nucleotide polymorphism, susceptibility, pulmonary tuberculosis, case-control study

Introduction

Pulmonary tuberculosis (TB) is one of the most devastating chronic infectious diseases, and remains the leading cause of death in developing countries. It is a significant cause of morbidity and mortality and causes almost 10 million new cases and 2 million deaths annually [1, 2].

As in the case of many other diseases, pulmonary TB is a complex disease with both environmental and genetic factors have been implicated in the infection and spread of the disease [3].

Human immunodeficiency virus (HIV) and *Mycobacterium tuberculosis* (MTB) infection were

two major risk factors for pulmonary TB incidence, and annually cause 3 million and 2 million deaths, respectively [4]. It has been reported that extensive exposure to mycobacteria in general does not result in the disease [5], suggesting that genetic factors also play a vital role in susceptibility to this disease.

Epidemiological studies have revealed that the environmental factors that influence pulmonary TB susceptibility include socio-economic conditions, smoking and acute infection. However, the genetic background of pulmonary TB remains unclear. Recently, a number of studies revealed statistically significant associations between PTB susceptibility and polymorphisms of some genes or gene variants, including the vitamin D receptor (VDR) gene [6], the rs3808607 of SP110 gene [7], the CYP7A1 gene [8], and the TLR8 gene [9].

The previous genetic polymorphisms studies of the pulmonary TB susceptibility mainly focused on the Africans and Asians in Genome-wide association study [10-13]. Although genome-wide association studies (GWAS) have successfully identified many common variants associated with pulmonary TB susceptibility, we did not found any data regarding a Chinese Tibetan population. In this study we want to find the pulmonary TB susceptibility loci for this ethnic group. So we choose 20 high frequency SNPs from previous GWAS study to estimate their association with pulmonary TB risk.

Materials and methods

Study population

All subjects were members of Chinese Tibetan population living in the Tibet Autonomous Region of China. The cases were recruited between October 2012 and September 2013 at the Department of Respiratory Physicians of Tangdu Hospital affiliated with the Fourth Military Medical University. All patients were newly diagnosed with pulmonary tuberculosis and were characterized histologically. None of the patients had previous history of other cancers, chemotherapy, or radiotherapy. Control subjects were randomly selected from the medical examination center of the Tang Du Hospital based on standard recruitment and exclusion criteria during the same period, and their medical history and a physical examination showed that they were in good health. Participants were chosen without restrictions of age, sex, or disease stage. As a result, a total of 217 cases and 383 controls were included in the study.

Clinical data and demographic information

We used a standard epidemiological questionnaire and in-person interview to collect personal data, including residential regions, age, gender, education status, and family history of cancer. The case information was collected through consultation with treating physicians or from medical chart review. All of the participants signed an informed consent agreement. The Human Research Committee for Approval of Research Involving Human Subjects, Tangdu Hospital, approved the use of human tissue in this study.

Selection of SNPs and methods of genotyping

We selected 20 SNPs from GWA studies that were previously reported to be associated with pulmonary tuberculosis. Minor allele frequencies of all SNPs were > 5%, in the HapMap of the Chinese Han Beijing (CHB) population. Genomic DNA was extracted from whole blood using the GoldMag-Mini Whole Blood Genomic DNA Purification Kit (GoldMag Co. Ltd., Xi'an City, People's Republic of China), and DNA concentration was measured with a NanoDrop 2000 spectrophotometer. We used Sequenom MassARRAY Assay Design 3.0 Software (San Diego, California, USA) to design a Multiplexed SNP MassEXTEND assay [14]. Genotyping was done with the Sequenom MassARRAY RS1000 system using the standard protocol recommended by the manufacturer. Data management and analysis was done using Sequenom Typer 4.0 Software [14, 15].

Statistical analysis

The SPSS18.0 statistical software and Microsoft Excel were used for statistical analysis. All p values presented in this study were two sided, and $p \leq 0.05$ was used as the cutoff value for statistical significance. An exact test was used to assess the variation in each SNP frequency from Hardy-Weinberg equilibrium (HWE) in the control subjects [16]. We tested odds ratios (ORs) and constructed 95% confidence intervals (CIs) using unconditional logistic regression analysis with adjustments for age and gender [17].

Table 1. Primers used for this study

SNP_ID	2nd-PCRP	1st-PCRP	UEP sequences
rs958617	ACGTTGGATGGCCAGTTTATAGTGTCACTC	ACGTTGGATGGAGTGTTGAATAGGTCTAAG	ACTCACTCCACTTTGTT
rs1434579	ACGTTGGATGCGTATAGGGCTTTTTTCCCG	ACGTTGGATGCGAGAAACCTTACAGATGCC	TGTGCACCTAAAGCCCTTTC
rs160441	ACGTTGGATGGAATTGTGCTTTCAGAATG	ACGTTGGATGGCTGCATTCCAGTTTGCTAT	TTGCAGCTTTATAGCTTTTGCT
rs17175227	ACGTTGGATGACAGTGAACTACTCAACATC	ACGTTGGATGCATTTAAAGAGGCATACAGC	CTCAACATCTCAGTAGACA
rs17217757	ACGTTGGATGCCAAGTATAAGTTCAGGAGC	ACGTTGGATGAGAGTTCAGTGACAGACTGC	ggggATAAGTTCAGGAGCATAACC
rs1900442	ACGTTGGATGAGCAGATATGTAAGATGACC	ACGTTGGATGGGTTTGGCAAAAGCAGCTA	tcATAAAACTGTACTGTGAAATTGTA
rs1925714	ACGTTGGATGGTGCAGGTTTGTTACATGG	ACGTTGGATGGATAGGATCACTCAGATCCC	tTTACATGGATTTATTGCATGAC
rs2505675	ACGTTGGATGACTCCTCGCTGAAATGATGC	ACGTTGGATGAAAGAGGGTTGGATCAGGTG	gggtCTGAAATGATGCTGTTGG
rs2837857	ACGTTGGATGGGAACACATGTGTAGGCATC	ACGTTGGATGTTAGGGAACATGCGGAGATG	gagCTCTACATTTCTTAGGAGGT
rs3218255	ACGTTGGATGCAGTTTCAGAGTCATGAACG	ACGTTGGATGAGGACCTCGAGTTTGAGCAC	ATGAACGACAGGGCA
rs40363	ACGTTGGATGAAGCGAACTACTCTGTTGGG	ACGTTGGATGGTTTTCACTCTGCCAAGTCC	ATGAGTGACACAAAAGTGTA
rs586716	ACGTTGGATGGGACTTAAGAAGGGATTAAAC	ACGTTGGATGTCAGACGATTCTCATCCATC	ATTAAACTACGTAGTCTCACA
rs6538140	ACGTTGGATGCTGGCCAAAGCATGTATCAC	ACGTTGGATGTTTCCCCAACAAAATAGTCC	cCCTTGTGAGCTTGGTTT
rs6545883	ACGTTGGATGGTTGTACTGCCAAATCATGC	ACGTTGGATGCTGAAAATTGCCACAGCTCG	ggATCATGCTAGACCACTC
rs6575836	ACGTTGGATGCAATAGGGTAGCCAACAAGC	ACGTTGGATGCTGGAAAGATGTTGGTTCGC	cACAAAGCTGGGGAATGA
rs6676375	ACGTTGGATGGGATTTAAAGTGCCAGGGAG	ACGTTGGATGTGTAGCAGAATGAGCCGTTG	GAGAAGCTGAGGGAGATAC
rs712039	ACGTTGGATGAAGACAGATGTTAGCACCGC	ACGTTGGATGTTTCAGCGATGCCAAGTGTC	aacaGCGCACACCCATCTACCACCGCA
rs7821565	ACGTTGGATGTACTCCCATATGTCAGCAAC	ACGTTGGATGTTCCCTCATCAATGTGCTCC	gccACAAACATACAGAAAGCATA
rs8005962	ACGTTGGATGTGATGGTACTAGAAGATGGG	ACGTTGGATGTAAGGGCACTAATCCCTCTG	ggcaTATGGGAGGTGATCAGGTC
rs9373523	ACGTTGGATGGACATGTACAATAAAAACTTC	ACGTTGGATGTGTGGATACTGCCACTTAAC	GAAACTAAGATGTAAGTCAGT

UEP unextended minisequencing primer.

Associations between SNPs and risk of pulmonary tuberculosis were tested using four different genetic models (co-dominant, dominant, recessive and log-additive) analysis by SNPtats, website software from http://bioinfo.iconcologia.net. We calculated ORs and 95% CIs by unconditional logistic regression analysis adjusted for age and gender [17]. Akaike's Information Criterion and Bayesian Information Criterion were applied to estimate the best-fit model for each SNP.

Results

A total of 217 cases and 383 controls were included in the study. The mean age for the case group was 60.7 ± 8.9 years and 49.4 ± 7.9 years for the control group. There were significant differences in age and gender distribution between the case and control groups ($P < 0.05$).

A total of twenty SNPs were selected for further genotyping. The primer sequences are presented in **Table 1**. **Table 2** summarizes the basic characteristics of the tested SNPs and their estimated association with pulmonary tuberculosis risk in crude analysis. The allelic frequency of other SNPs in the controls group was similar to those of the HapMap CHB population. Through the χ^2 test, we found five SNPs had an increased risk of pulmonary tuberculosis, including rs6676375 (OR = 1.48; 95% CI = 1.04-2.10; $P = 0.030$), rs7821565 (OR = 1.51, 95% CI = 1.12-2.05; $P = 0.007$), rs17217757 (OR = 1.66; 95% CI = 1.21-2.27; $P = 0.001$), rs1900442 (OR = 1.28, 95% CI = 1.01-1.62; $P = 0.041$), and rs712039 (OR = 1.34, 95% CI = 1.06-1.70; $P = 0.014$). Additionally, we found five SNPs were significantly associated with reduce risk of pulmonary tuberculosis, including rs2505675 (OR = 0.78; 95% CI = 0.61-1.00; $P = 0.048$), rs586716 (OR = 0.67; 95% CI = 0.47-0.96; $P = 0.027$), rs1434579 (OR = 0.63; 95% CI = 0.47-0.84; $P = 0.002$), rs2837857 (OR = 0.66; 95% CI = 0.49-0.89; $P = 0.006$), and rs6538140 (OR = 0.64; 95% CI = 0.50-0.82; $P = 0.000$) (**Table 2**).

Four genetic models (co-dominant, dominant, recessive, and log-additive) were applied to analyze the associations between the SNPs and pulmonary tuberculosis. The results showed that the rs7821565 was significantly associated with an increased risk of pulmonary tuberculosis, based on the results from the co-dominant model (OR = 3.26; 95% CI = 1.32-8.06, $P = 0.018$, dominant model (OR = 1.46; 95% CI = 1.02-2.09, $P = 0.037$), recessive model (OR = 3.01; 95% CI = 1.23-7.38, $P =$

Table 2. Allele frequencies in cases and controls and odds ratio estimates for pulmonary TB

SNP ID	Gene (s)	Band	Alleles	H-W	MAF		ORs	95% CI	*P*-value
			A[a]/B	*P*-value	Case	Control			
rs1925714	BTNL10-RNA5SP19	1q42.13	A/G	0.754	0.088	0.089	0.98	0.65-1.49	0.943
rs6676375	RPL10AP5-RSL24D1P4	1q43	C/T	0.158	0.145	0.103	1.48	1.04-2.10	0.030*
rs6545883	XPO1-RPS29P10	2p15	A/G	0.444	0.410	0.372	1.17	0.92-1.49	0.193
rs958617	CXCL13-CNOT6L	4q21.1	T/C	0.584	0.408	0.373	1.16	0.91-1.48	0.230
rs2505675	GMDS-AS1	6p25.2	G/A	0.198	0.332	0.389	0.78	0.61-1.00	0.048
rs9373523	STXBP5	6q24.3	T/G	0.529	0.472	0.415	1.26	1.00-1.60	0.055
rs7821565	CYCSP22-PXDNL	8q11.22	C/T	1.000	0.212	0.151	1.51	1.12-2.05	0.007
rs160441	RNA5SP272-RIPK2	8q21.3	T/C	0.552	0.248	0.223	1.14	0.87-1.51	0.341
rs17217757	ZFPM2	8q23.1	C/G	0.175	0.201	0.132	1.66	1.21-2.27	0.001
rs586716	DMRTA1-LINC01239	9p21.3	G/A	0.560	0.111	0.157	0.67	0.47-0.96	0.027
rs6538140	NAV3	12q21.2	G/A	0.607	0.345	0.450	0.64	0.50-0.82	0.000
rs1900442	VWA8	13q14.11	C/T	0.169	0.465	0.405	1.28	1.01-1.62	0.041
rs17175227	RPL7AP6-SLC8A3	14q24.2	A/G	0.106	0.037	0.039	0.94	0.51-1.74	0.842
rs8005962	GLRX5-TCL6	14q32.13	T/C	0.456	0.199	0.219	0.88	0.66-1.18	0.410
rs6575836	MEG9-DIO3OS	14q32.31	G/A	0.531	0.375	0.422	0.82	0.65-1.05	0.114
rs40363	NAA60	16p13.3	A/G	1.000	0.182	0.187	0.97	0.72-1.31	0.842
rs712039	DUSP14	17q12	C/T	1.000	0.507	0.433	1.34	1.06-1.70	0.014
rs1434579	ZNF229	19q13.31	A/G	0.248	0.191	0.273	0.63	0.47-0.84	0.002
rs2837857	DSCAM	21q22.2	T/C	0.682	0.180	0.248	0.66	0.49-0.89	0.006
rs3218255	IL2RB	22q12.3	T/C	0.227	0.094	0.091	1.04	0.69-1.55	0.859

Notes: [a]Minor allele; *p value ≤ 0.05 indicates statistical significance; Abbreviations: HWE, Hardy-Weinberg Equilibrium; MAF, minor allele frequency; SNP, single nucleotide polymorphism; ORs, odds ratios; CI, confidence interval.

0.014), and log-additive model (OR = 1.50; 95% CI = 1.11-2.02, *P* = 0.0091). The rs6676375 was associated with an increased risk in dominant (OR = 1.49; 95% CI = 1.01-2.20, *P* = 0.043) and log-additive model (OR = 1.52; 95% CI = 1.05-2.21, *P* = 0.026). We found the minor allele "C" of rs712039 was also increased the pulmonary tuberculosis risk in co-dominant (OR = 1.80; 95% CI = 1.13-2.84, *P* = 0.023) and recessive model (OR = 1.73; 95% CI = 1.17-2.55, *P* = 0.0063). The rs17217757 was also increase pulmonary tuberculosis risk in co-dominant (OR = 1.87; 95% CI = 1.28-2.73, *P* = 0.0041) and dominant model (OR = 1.85; 95% CI = 1.29-2.66, *P* = 9.00E-04) (**Table 3**).

In contrast, we found the minor allele "G" of rs2505675 was associated with decreased risk of pulmonary tuberculosis, based on results from the co-dominant model (OR = 0.52; 95% CI = 0.30-0.91, *P* = 0.05) and recessive model (OR = 0.53; 95% CI = 0.32-0.90, *P* = 0.015). Additionally, we found the rs586716 (OR = 0.65; 95% CI = 0.43-0.98, *P* = 0.081) and rs6538140 (OR = 0.47; 95% CI = 0.28-0.77, *P* = 7.00E-04) were significantly associated with decreased pulmonary tuberculosis risk under the co-dominant model. The "A/A" genotype of rs1434579 was associated with decrease the risk in co-dominant (OR = 0.44; 95% CI = 0.21-0.90, *P* = 0.0078) and dominant model (OR = 0.60; 95% CI = 0.42-0.84, *P* = 0.0031). The rs2837857 was also decrease the risk of pulmonary tuberculosis, based on the results from the co-dominant (OR = 0.36; 95% CI = 0.14-0.90, *P* = 0.019, dominant model (OR = 0.66; 95% CI = 0.46-0.93, *P* = 0.017) (**Table 3**).

Discussion

Pulmonary tuberculosis continues to be a serious global health problem. Previous studies have demonstrated contribution of host genetic factors in determining of susceptibility PTB [18, 19]. Thus, to date, little is known about the underlying genetic determinants or mechanisms contributing to differences in susceptibility to TB at the population level. This genetic association study was sought to identify novel PTB risk-related variants, 20 SNPs were not randomly chosen, but instead obtained from previous studies. Four tested SNPs rs7821565, rs6676375, rs712039, and rs17217757 were associated with an increased risk of PTB.

Table 3. Associations between selected SNPs and pulmonary TB risk

SNP	Model	Genotype	Controls	Cases	OR (95% CI)[a]	*P*-value	AIC	BIC
rs6676375	Co-dominant	T/T	305 (79.6%)	157 (72.3%)	1	0.056	785.5	798.7
		C/T	77 (20.1%)	57 (26.3%)	1.44 (0.97-2.13)			
		C/C	1 (0.3%)	3 (1.4%)	5.83 (0.60-56.49)			
	Dominant	T/T	305 (79.6%)	157 (72.3%)	1	0.043*	785.2	794
		C/T-C/C	78 (20.4%)	60 (27.6%)	1.49 (1.01-2.20)			
	Recessive	T/T-C/T	382 (99.7%)	214 (98.6%)	1	0.11	786.7	795.5
		C/C	1 (0.3%)	3 (1.4%)	5.36 (0.55-51.80)			
	Log-additive	---	---	---	1.52 (1.05-2.21)	0.026	784.3	793.1
rs2505675	Co-dominant	A/A	149 (38.9%)	94 (43.3%)	1	0.05	785.2	798.4
		G/A	170 (44.4%)	102 (47%)	0.95 (0.67-1.36)			
		G/G	64 (16.7%)	21 (9.7%)	0.52 (0.30-0.91)			
	Dominant	A/A	149 (38.9%)	94 (43.3%)	1	0.29	788.1	796.9
		G/A-G/G	234 (61.1%)	123 (56.7%)	0.83 (0.59-1.17)			
	Recessive	A/A-G/A	319 (83.3%)	196 (90.3%)	1	0.015	783.3	792.1
		G/G	64 (16.7%)	21 (9.7%)	0.53 (0.32-0.90)			
	Log-additive	---	---	---	0.78 (0.61-1.00)	0.05	785.4	794.2
rs7821565	Co-dominant	T/T	275 (72%)	137 (63.7%)	1	0.018	778.2	791.4
		C/T	99 (25.9%)	65 (30.2%)	1.32 (0.91-1.92)			
		C/C	8 (2.1%)	13 (6%)	3.26 (1.32-8.06)			
	Dominant	T/T	275 (72%)	137 (63.7%)	1	0.037	779.9	788.7
		C/T-C/C	107 (28%)	78 (36.3%)	1.46 (1.02-2.09)			
	Recessive	T/T-C/T	374 (97.9%)	202 (94%)	1	0.014	778.3	787
		C/C	8 (2.1%)	13 (6%)	3.01 (1.23-7.38)			
	Log-additive	---	---	---	1.50 (1.11-2.02)	0.0091	777.5	786.3
rs17217757	Co-dominant	G/G	292 (76.2%)	137 (63.4%)	1	0.0041	778.2	791.4
		G/C	81 (21.1%)	71 (32.9%)	1.87 (1.28-2.73)			
		C/C	10 (2.6%)	8 (3.7%)	1.71 (0.66-4.42)			
	Dominant	G/G	292 (76.2%)	137 (63.4%)	1	9.00E-04	776.3	785
		G/C-C/C	91 (23.8%)	79 (36.6%)	1.85 (1.29-2.66)			
	Recessive	G/G-G/C	373 (97.4%)	208 (96.3%)	1	0.46	786.7	795.4
		C/C	10 (2.6%)	8 (3.7%)	1.43 (0.56-3.69)			
	Log-additive	---	---	---	1.63 (1.20-2.23)	0.0021	777.7	786.5
rs586716	Co-dominant	A/A	274 (71.5%)	173 (79.7%)	1	0.081	786.2	799.4
		G/A	98 (25.6%)	40 (18.4%)	0.65 (0.43-0.98)			
		G/G	11 (2.9%)	4 (1.8%)	0.58 (0.18-1.84)			
	Dominant	A/A	274 (71.5%)	173 (79.7%)	1	0.025	784.2	793
		G/A-G/G	109 (28.5%)	44 (20.3%)	0.64 (0.43-0.95)			
	Recessive	A/A-G/A	372 (97.1%)	213 (98.2%)	1	0.43	788.6	797.4
		G/G	11 (2.9%)	4 (1.8%)	0.64 (0.20-2.02)			
	Log-additive	---	---	---	0.68 (0.48-0.97)	0.028	784.4	793.2
rs6538140	Co-dominant	A/A	113 (29.5%)	97 (44.9%)	1	7.00E-04	774.7	787.9
		G/A	195 (50.9%)	89 (41.2%)	0.53 (0.37-0.77)			
		G/G	75 (19.6%)	30 (13.9%)	0.47 (0.28-0.77)			
	Dominant	A/A	113 (29.5%)	97 (44.9%)	1	2.00E-04	773	781.8
		G/A-G/G	270 (70.5%)	119 (55.1%)	0.51 (0.36-0.73)			
	Recessive	A/A-G/A	308 (80.4%)	186 (86.1%)	1	0.074	784	792.8
		G/G	75 (19.6%)	30 (13.9%)	0.66 (0.42-1.05)			
	Log-additive	---	---	---	0.65 (0.51-0.83)	4.00E-04	774.6	783.4
rs712039	Co-dominant	T/T	123 (32.1%)	59 (27.2%)	1	0.023	783.7	796.9
		T/C	188 (49.1%)	96 (44.2%)	1.06 (0.72-1.58)			
		C/C	72 (18.8%)	62 (28.6%)	1.80 (1.13-2.84)			

	Dominant	T/T	123 (32.1%)	59 (27.2%)	1	0.21	787.6	796.4
		T/C-C/C	260 (67.9%)	158 (72.8%)	1.27 (0.88-1.83)			
	Recessive	T/T-T/C	311 (81.2%)	155 (71.4%)	1	0.0063	781.8	790.6
		C/C	72 (18.8%)	62 (28.6%)	1.73 (1.17-2.55)			
	Log-additive	---	---	---	1.33 (1.05-1.68)	0.016	783.5	792.3
rs1434579	Co-dominant	G/G	207 (54%)	144 (66.4%)	1	0.0078	781.5	794.7
		G/A	143 (37.3%)	63 (29%)	0.63 (0.44-0.91)			
		A/A	33 (8.6%)	10 (4.6%)	0.44 (0.21-0.91)			
	Dominant	G/G	207 (54%)	144 (66.4%)	1	0.0031	780.5	789.3
		G/A-A/A	176 (46%)	73 (33.6%)	0.60 (0.42-0.84)			
	Recessive	G/G-G/A	350 (91.4%)	207 (95.4%)	1	0.059	785.7	794.5
		A/A	33 (8.6%)	10 (4.6%)	0.51 (0.25-1.06)			
	Log-additive	---	---	---	0.65 (0.49-0.86)	0.0019	779.6	788.4
rs2837857	Co-dominant	C/C	218 (56.9%)	145 (66.8%)	1	0.019	783.3	796.5
		C/T	140 (36.5%)	66 (30.4%)	0.71 (0.49-1.02)			
		T/T	25 (6.5%)	6 (2.8%)	0.36 (0.14-0.90)			
	Dominant	C/C	218 (56.9%)	145 (66.8%)	1	0.017	783.5	792.3
		C/T-T/T	165 (43.1%)	72 (33.2%)	0.66 (0.46-0.93)			
	Recessive	C/C-C/T	358 (93.5%)	211 (97.2%)	1	0.036	784.9	793.6
		T/T	25 (6.5%)	6 (2.8%)	0.41 (0.16-1.01)			
	Log-additive	---	---	---	0.67 (0.49-0.89)	0.0059	781.7	790.5

Notes: *P-value ≤ 0.05 indicates statistical significance. Abbreviations: SNP, single nucleotide polymorphism; OR, odds ratio; CI, confidence interval; AIC, Akaike's Information Criterion; BIC, Bayesian Information Criterion.

Additionally, five SNPs rs2505675, rs586716, rs6538140, rs1434579, and rs2837857, respectively decreased the risk of PTB in genetic models analysis.

Rs712039 located on the DUSP14 gene. DUSP14 gene is a member of a large family of phosphatases, which was considered as a susceptibility gene to pulmonary TB [20]. Previous study has demonstrated the association between rs712039 and pulmonary TB [21]. In addition, we found rs712039 associated with increased risk of pulmonary TB in our study.

The SNP rs7821565 is located downstream of CYCSP22 and upstream of PXDNL gene region at 8q11.22. This SNP was not discussed in the original GWAS paper, probably because it was not significant at a genome-wide threshold, yet it shows one of the strongest genetic associations with pulmonary TB in our study. The mechanisms of functions of this polymorphism were not clearly.

Rs2837857 maps to DSCAM gene at chromosome band 21q22.2-q22.3. The DSCAM gene is a novel member of the immunoglobulin superfamily maps in a Down syndrome region and is involved in the development of the nervous system [22]. Rs2837857 was found to show a significant decreased risk of pulmonary TB in our present study. Since the study about rs2837857 and pulmonary TB was few, further investigation was needed to verify this.

Our study also found that rs2505675 and rs6538140 show a protctive effect of pulmonary TB in the genetic models anylasis. The two SNPs were not found to be associated with pulmonary TB risk in previous studies. In addition, the two SNPs were raraly studied in other studies. So the association between this two polymorphisms and pulmonary TB need to be further investigated in a large study with more samples.

Despite the current study possessing enough power, potential limitations of this study be considered. Firstly, the sample size of our study was relatively small. The statistical power may be limited because of the sample size. Secondly, this was a hospital-based study; therefore, selection bias may be unavoidable. Thirdly, the association between genetic polymorphism and clinicopathological type (TB or PTB) was not evaluated in this study. So we could not explore how genetic polymorphisms play a role in the Tibetan population or subtype. So larger well-designed studies combined with functional evaluations are needed to confirm the asso-

ciations and clarify the potentially biological mechanisms of these polymorphisms in pulmonary tuberculosis.

Conclusions

In conclusion, this association study investigated 20 SNPs from previous GWAS as genetic susceptibility factors for pulmonary TB in a Chinese Tibetan population. Our study provides the first reported data of a possible association between the SNPs rs2505675, rs6538140, rs5867716, rs586716, and rs6676375 and pulmonary TB. Our findings and those of previous studies suggest that polymorphisms of particular genes play a role in pulmonary TB development. However, these SNPs require further investigation before definitive conclusions can be drawn.

Acknowledgements

This work is supported by the Major science and technology research projects of Xizang (Tibet) Autonomous Region (2015). We are grateful to the patients and control subjects for their participation in this study. We also thank the clinicians and hospital staff who contributed to the sample and data collection for this study.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Tianbo Jin, Key Laboratory of High Altitude Environment and Genes Related to Diseases of Tibet Autonomous Region, School of Medicine, Xizang Minzu University, 6 East Wenhui Road, Xianyang 712082, Shaanxi, China. Tel: +86-29-33755000; Fax: +86-29-33755000; E-mail: jintianbo@gmail.com

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