Tumor Biol. (2014) 35:1863–1866
DOI 10.1007/s13277-013-1248-3

RESEARCH ARTICLE

Genetic variation in the *TP63* gene is associated with lung cancer risk in the Han population

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Received: 13 August 2013 / Accepted: 23 September 2013 / Published online: 4 October 2013

Abstract Lung cancer is one of the most common malignant tumors that seriously threaten human health. Current evidence suggests that heredity contributes to the progression of lung cancer. To investigate and validate potential genetic associations with the risk of lung cancer, we conducted a case–control study including 309 cases and 310 controls from Xi'an City, which is located in northwest China, and genotyped six SNPs in five genes, which are related to metabolic process. Overall, our results show that the SNP rs10937405 was associated with a decreased occurrence of lung cancer (OR=0.72; 95 % CI=0.56–0.92; p=0.009). In the genetic models analysis, we found that genotype "CT" of rs10937405 in TP63 was associated with a decreased lung cancer risk (OR=0.71; 95 % CI, 0.51–0.99; p=0.031); the genotype "TT" of rs10937405 showed a decreased lung cancer risk in the co-dominant model (OR=0.53; 95 % CI, 0.30–0.95; p=0.031). The genotype "CT-TT" of rs10937405 also showed a decreased lung cancer risk in the dominant model (OR=0.67; 95 % CI, 0.49–0.92; p=0.014) and the log-additive model (OR=0.72; 95 % CI, 0.56–0.92; p=0.0085). The genotype "CC-CT" of rs10937405 confers a higher risk of lung cancer for males than females. Our results, combined with those from previous studies, suggest that genetic variation in *TP63* may influence lung cancer susceptibility in the Han population.

Keywords Lung cancer · Single-nucleotide polymorphism · *TP63* · Case–control studies

Introduction

Lung cancer is the most common malignancy worldwide [1]. In China, lung cancer is a major health problem and has been reported with high mortality rate for both men and women [2, 3]. Epidemiological studies have demonstrated that the high proportion of smokers in the general population and the polluted environment in cities with the process of urbanization and industrialization are primary etiologic factors for lung cancer occurrence. However, genetic factors may play an important role in determining susceptibility to lung cancer [4]. To investigate how genetic factors contribute to lung cancer susceptibility in the Han population, we conducted a case–control study and selected six tSNPs from five genes, which have previously been reported to be associated with lung cancer susceptibility in genome-wide association studies [5–7].

Materials and methods

Study participants

We recruited a total of 309 patients at the Affiliated Hospital of Tibet University for Nationalities from October 2011 to

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Table 1 Basic characteristics of the subjects

	Case (n=309)	Control (n=310)
Age (years)	58.2±10.2	50.4±8.1
Sex		
Male	235 (76.1 %)	197 (63.5 %)
Female	74 (23.9 %)	113 (36.5 %)
Histology		
SCLC	48 (15.5 %)	
LAC	86 (27.8 %)	
LSCC	90 (29.1 %)	
LASC	11 (3.6 %)	
Others	74 (23.9 %)	

SCLC small cell lung cancer, *LAC* lung adenocarcinoma, *LSCC* lung squamous cell carcinoma, *LASC* lung adenosquamous carcinoma

September 2012 (Xi'an City, China). All patients were newly diagnosed and histologically identified with lung cancer. None of them had a previous history of other cancers, chemotherapy, or radiotherapy. They were chosen without age, gender, or disease stage restrictions. We also selected 310 healthy unrelated individuals during the same time period from the medical examination center of the Affiliated Hospital of Tibet University for Nationalities based on standard recruitment and exclusion criteria. We ensured that they have no chronic or severe endocrinological, metabolic, and nutritional diseases. All study participants were Han Chinese living in Xi'an City or nearby.

Clinical data and demographic information

We use a standardized epidemiological questionnaire including residential region, age, gender, smoking status, alcohol use, ethnicity, education status, and family history of cancer to collect personal data in an in-person interview. We informed all participants of the purpose and experimental procedures of the study and obtained signed informed consent from each participant. The Human Research Committee of the Affiliated Hospital of Tibet University for Nationalities for Approval of Research Involving Human Subjects approved the use of human tissue in this study.

SNP selection and genotyping

Six SNPs in the five metabolic process genes selected were associated with lung cancer, with minor allele frequencies (MAF) >5 % in the HapMap Chinese Han Beijing (CHB) population. DNA was extracted from whole-blood samples using the GoldMag-Mini Whole Blood Genomic DNA Purification Kit (GoldMag Co. Ltd. Xi'an City, China). DNA concentrations were measured with the NanoDrop 2000 (Thermo Scientific, Waltham, MA, USA). Multiplexed SNP MassEXTENDED assay was designed by Sequenom MassARRAY Assay Design 3.0 Software (Sequenom Co. Ltd., San Diego, CA, USA) [8]. SNP genotyping with a standard protocol was performed using Sequenom MassARRAY RS1000 (Sequenom Inc., San Diego, CA, USA) [8]. Sequenom Typer 4.0 Software (Sequenom Inc., San Diego, CA, USA) was used to analyze the data [8, 9].

Statistical analysis

Statistical processing of our data was performed by SPSS 16.0 software (SPSS Chicago, IL, USA) and Microsoft Excel software. The p values presented in this study are all two-sided, and p=0.05 was used as the threshold of statistical significance. The validation of each SNP frequency in control group was tested for departure from Hardy-Weinberg equilibrium (HWE). A χ^2 test was used to compare the allelic frequencies of case and control groups [10].

The genetic association between genotype and lung cancer risk was tested under different genetic models (codominant,

Table 2 Basic information of candidate SNPs in this study

SNP #	Gene(s)	Band	Alleles	MAF		HWE p	ORs	95 % CI		p adj
				Case	Control					
rs4488809	TP63	3q28	C/T	0.50/0.50	0.45/0.55	0.389	1.22	0.98	1.53	0.077
rs10937405	TP63	3q28	C/T	0.26/0.74	0.33/0.67	1.000	0.72	0.56	0.92	0.009*
rs7626795	IL1RAP	3q28	G/A	0.80/0.20	0.81/0.19	0.556	1.04	0.79	1.38	0.773
rs753955	MIPEP	13q12.12	C/T	0.66/0.34	0.66/0.34	0.999	0.98	0.78	1.24	0.872
rs748404	TGM5	15q15.2	C/T	0.94/0.06	0.92/0.08	0.836	0.83	0.53	1.29	0.413
rs36600	MTMR3	22q12.2	C/T	0.08/0.92	0.10/0.90	5.59E-05	0.84	0.55	1.28	0.421

SNP single-nucleotide polymorphism, *OR* odds ratio, *95 % CI* 95 % confidence interval, *HWE* Hardy–Weinberg equilibrium, *adj* adjusted

* $p \leq 0.05$ indicates statistical significance

Table 3 Relationship between rs10937405 and lung cancer risk

Model	Genotype	Control	Case	OR (95 % CI)	*p* value	AIC	BIC
Codominant	C/C	138 (44.5 %)	168 (54.4 %)	1.00	0.031*	857.2	870.5
	C/T	138 (44.5 %)	119 (38.5 %)	0.71 (0.51–0.99)			
	T/T	34 (11 %)	22 (7.1 %)	0.53 (0.30–0.95)			
Dominant	C/C	138 (44.5 %)	168 (54.4 %)	1.00	0.014*	856.1	865
	C/T-T/T	172 (55.5 %)	141 (45.6 %)	0.67 (0.49–0.92)			
Recessive	C/C-C/T	276 (89 %)	287 (92.9 %)	1.00	0.094	859.3	868.2
	T/T	34 (11 %)	22 (7.1 %)	0.62 (0.36–1.09)			
Overdominant	C/C-T/T	172 (55.5 %)	190 (61.5 %)	1.00	0.13	859.8	868.7
	C/T	138 (44.5 %)	119 (38.5 %)	0.78 (0.57–1.08)			
Log-additive	–	–	–	0.72 (0.56–0.92)	0.0085*	855.2	864.1

OR odds ratio, *CI* confidence interval, *AIC* Akaike's Information criterion, *BIC* Bayesian Information criterion

*p value≤0.05 indicates statistical significance

dominant, recessive, overdominant, and log-additive) with SNPStats, a website software from http://bioinfo.iconcologia.net/snpstats/start.htm [11]. Testing of odds ratios (ORs) and 95 % confidence intervals (CIs) was performed using unconditional logistic regression analysis with adjustment for gender and age [12]. Akaike's Information Criterion (AIC) and Bayesian Information Criterion (BIC) were applied to estimate the best-fit model for each SNP. Furthermore, association between genotype and lung cancer risk in gender-specific populations under each model was performed using SNPStats software [11].

Results

We included a total of 619 participants, with 309 patients (235 males, 74 females; mean age at diagnosis 58 years, range 25–85 years) and 310 controls (197 males, 113 females; mean age at 50 years, range 29–75 years) for association analysis. Basic characteristics of the subjects were listed in Table 1. Six SNPs in five metabolic process genes in lung cancer patients and the healthy controls were genotyped. Table 2 listed the MAF of cases and controls. Rs36600 was excluded from further analysis because it derived from HWE at 1 % p level. We compared the differences in frequency distributions of alleles between cases and controls using χ^2 test and found one significant SNP in the *TP63* gene, rs10937405, was associated with decreased lung cancer risk (OR=0.72; 95 % CI, 0.56–0.92; p=0.009).

The genetic association between rs10937405 and lung cancer risk was tested under different genetic models listed in Table 3. In the codominant model, the genotypes "CT" (OR=0.71; 95 % CI, 0.51–0.99; p=0.031) and "TT" (OR=0.53; 95 % CI, 0.30–0.95; p=0.031) of rs10937405 were associated with decreased lung cancer risk. The genotype "CC-CT" of rs10937405 was associated with decreased lung cancer risk in the dominant model (OR=0.67; 95 % CI, 0.49–0.92; p=0.014) and in the log-additive model (OR=0.72; 95 % CI, 0.56–0.92; p=0.0085). Table 4 shows that the genotype "CC-CT" of rs10937405 confers a higher risk of lung cancer for males than females (OR=2.04; 95 % CI, 1.41–2.96; p=0.033).

Discussion

In the current case–control study in the Han population, we genotyped six SNPs previously reported to be associated with lung cancer risk and identified that rs10937405 in the *TP63* gene has a strong association with reduced lung cancer risk.

The SNP rs10937405 maps to the *TP63* gene whose product is the tumor protein p63, an important component of the

Table 4 Association between sex and lung cancer risk with rs10937405 under recessive model (n=619)
Interaction p value, 0.033
OR odds ratio, *CI* confidence interval

	Female			Male		
Genotype	Control	Case	OR (95 % CI)	Control	Case	OR (95 % CI)
C/C-C/T	102	64	1.00	174	223	2.04 (1.41–2.96)
T/T	11	10	1.45 (0.58–3.61)	23	12	0.83 (0.39–1.79)

p53 family of genes. The p53 pathway plays a critical role in cell-cycle regulation by functioning as a tumor suppressor in numerous cancers. In addition, p53 mutations occur in approximately two-thirds of all human tumors [13, 14]. Furthermore, p63 has been found to play an important role in cancer development and progression through its interaction with mutant p53 [15]. An isoform of *TP63* has been proposed to have oncogenic properties based on its dominant negative effects on p53, and TP63 genomic gains have been identified as potential indicators of pre-invasive lung lesions and early lung cancer diagnosis [16]. This SNP was previously reported in a GWAS conducted in Japan and South Korea [17]. In addition, it has been confirmed that TP63 is associated with lung adenocarcinoma in the UK population [5]. The two researches both confirmed that rs10937405 increased the risk of lung adenocarcinoma. However, in our study, we did not conduct sub-grouped for the small samples and found the loci may decrease the risk of lung cancer.

Other genes were not found associations in our study. Maybe, the sample size was a little small. We really need more samples to further validate the findings.

The genotype "CC-CT" of rs10937405 confers a higher risk of lung cancer for males than females. In fact, many tumors tend to occur in males. The real mechanism remains unclear. Perhaps the activity of gene product has difference in males and females.

In conclusion, our study described the association between rs10937405 in the *TP63* gene and lung cancer risk in the Han population. Our findings, combined with previous studies, suggest a potential genetic susceptibility in *TP63* for lung cancer progression.

Acknowledgments This work is supported by the National Natural Science Foundation 31260252 and Major Training Program of Tibet University for Nationalities (no.12KJZRYMY10). We are grateful to all the patients and individuals in the study who made this work possible. We would also like to thank the clinicians and hospital staff who contributed to data collection for this study.

Conflict of interest None.

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