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Risk of Lung Cancer and Genetic Variations of *TERT* and *CLPTM1L* Genes: A Case-Control Study in an Iranian Population

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ABSTRACT Genetic variants in chromosome 5p15.33 locus which is comprised of telomerase reverse transcriptase (*TERT*) and cleft lip and palate trans-membrane 1 like (*CLPTM1L*) genes were correlated with the susceptibility to lung cancer in several populations. The current study aimed to examine the frequency and also the association between two significant SNPs of *TERT*-*CLPTM1L* region and lung cancer in an Iranian population. The researchers carried out a case-control study, including 266 lung cancer patients and 250 cancer-free healthy controls matched for age, sex, and smoking status to test associations between *TERT* rs2736098 and *CLPTM1L* rs401681 polymorphisms, and lung cancer incidence in an Iranian population. The results revealed that the *TERT* rs2736098 T allele carriers were positively associated with lung cancer, especially lung adenocarcinoma. In contrast, T allele carriers of *CLPTM1L* were inversely associated with lung cancer and adenocarcinoma.

INTRODUCTION

Lung cancer, one of the most common types of cancer worldwide, has been pathologically classified into the two major subtypes, small cell lung cancer (SCLC) and non-small cell lung cancer (NSCLC). The NSCLC has also two major subgroups, adenocarcinoma and squamous cell carcinoma (Liao et al. 2018; Zhao et al. 2013). Although almost half of known lung cancer cases occur in developing countries, the incidence rate of this cancer is notably low in Iran. However, lung cancer is among the five top deadliest cancers in Iran in both men and women. In Iran, potential risk factors of lung cancer include smoking, occupational exposures to inorganic dusts, chemical compounds, and heavy metals (Hosseini et al. 2009). To date, several association studies and meta-analyses have shown significant association between genetic polymorphisms and the pathogenesis and susceptibility of lung cancer (Liu et al. 2018; Sepesi et al. 2018).

The chromosome 5p15.33 locus has two genes, telomerase reverse transcriptase (*TERT*) and cleft lip and palate transmembrane1-like (*CLPTM1L*), and its polymorphisms have been associated with lung cancer risk in different populations (McKay et al. 2008; Wang et al. 2008; Yoon et al. 2010; Chen et al. 2012; Li et al. 2013; Myneni et al. 2013; Zhang et al. 2014). *TERT*, as a component of telomerase, functions to maintain telomere length by its reverse transcription activity (Lantuéjoul et al. 2007). Telomeres protect chromosomes from degradation, fusion and rearrangement and allow chromosomal stability and cellular immortality (Rodier et al. 2005). Therefore, *TERT* is an important component of cellular immortality and carcinogenesis in various cancers, including lung cancer (Hanahan and Weinberg 2000). Likewise, dysregulation of *TERT* mRNA and protein has been reported in lung cancer (Wang et al. 2002). *CLPTM1L* gene predictably produces transmembrane protein with an almost unknown functionality and expresses in various normal or malignant tissues (Li et al. 2013). Several studies have reported associated polymorphisms of *CLPTM1L* among cancer patients, including lung cancer patients (Wang et al. 2008; Zhao et al. 2014). Moreover, James et al. (2012) suggested an anti-apoptotic function for *CLPTM1L* since it protects lung tumor cells from genotoxic stress induced apoptosis.

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Within *TERT-CLPTM1L* region, *TERT* rs2736098 polymorphism has been identified to be associated with genetic variant in lung cancer. Furthermore, in this region, rs401681 SNP with high linkage disequilibrium (LD) links to the regulatory sequence of *TERT*, the promoter region, and the whole coding region of the *CLPTM1L* gene (Li et al. 2013). To the researchers' knowledge, no association study has been conducted to evaluate the importance of these polymorphisms in the Iranian population.

Objective

In the present investigation, the researchers aimed to test the association between lung cancer susceptibility and genetic polymorphisms of *TERT* rs2736098 and *CLPTM1L* rs401681 in a case-control study, including whole blood sample of 266 lung cancer patients and 250 cancer-free controls in an Iranian population.

METHODOLOGY

Study Subjects

This association study included 266 lung cancer patients and 250 healthy subjects (free of cancer and any other serious chronic disease). All participants were unrelated Iranian. Lung cancer patients were pathologically diagnosed and recruited between December 2012 and December 2015 at Imam Reza hospital, Tabriz, Iran. In the present study, patients with large-cell, mixed cell carcinomas and undifferentiated carcinomas were excluded. There were no gender, age, and smoking status restrictions. All samples were collected from the same geographic region and the same period of time. The inclusion criteria for cancer-free samples were gender and age matched to the cases. This study was approved by the Ethics Committee of the Faculty of Science, Yazd University, Yazd, Iran and written informed consent was obtained from the all subjects. 5 ml peripheral blood sample and relative clinicopathological information were collected.

TERT rs2736098 and *CLPTM1L* rs401681 Genotyping Analysis

The salting out method was used to isolate genomic DNA of peripheral blood lymphocytes. DNA purity and concentration were qualified by Nano Drop 1000 spectrophotometer (Thermo Scientific Inc., Wilmington, DE, USA). Notably, the *TERT* rs2736098 and *CLPTM1L* rs401681 variants were blindly genotyped to avoid any biased genotyping.

DNA samples were genotyped for *TERT* rs2736098 C>T using following primers: F-5' TGA CCG TGG TTT CTG TGT GG3' , R-5' TGT CGC CTG AGG AGT AGA GG3' . PCR was performed in a final volume of a 25 µl reaction mixture containing 50 ng of template DNA, 1X PCR buffer, 2.0 mM of MgCl2, 200 µM of dNTPs, 10 pM of each primer and 1 unit of Taq DNA polymerase. Standard cycling was performed in a Thermal cycler (Peqlab Primus 25, PEQLAB Biotechnology GmbH) by heating to 95 °C" for 2 min followed by 35" cycles of 94 °C for 30'', 60 °C for 50'', 72 °C for 40'', and finally 72 °C for 5 min. The PCR products were electrophoresed by two percent agarose gel electrophoresis in 1×Tris-Borate-EDTA buffer at 100V and stained with ethidium bromide for visualization. Detecting allelic variations was conducted by digesting PCR products with the restriction enzyme *ApaI* (#ER1411, Thermo Scientific Inc.). The *ApaI* restriction enzyme was selected in order to not provide cut in 215 bp PCR product containing C allele (band is 215 bp), while yielded two bands of 118 bp, and 97 bp due to existence of T allele in the fragment. To verify the obtained genotypes, ten randomly selected samples with different genotypes were directly sequenced using Macrogen sequencing service (Macrogen Inc., Seoul, Korea) (Fig. 1A).

The second SNP of interest, *CLPTM1L* rs401681 C>T, was genotyped using the Polymerase Chain Reaction-Single-Strand Conformational Polymorphism (PCR-SSCP) analysis followed by sequencing. The PCR was carried out in the same reaction mixture explained above and using of following primers: F-5' GCC AGA AAG CTG CTT CAC AC3' , R-5' AAA GGC ATA GAC CCC TGC AG3' . The amplicon size is 193 bp. The PCR program was 95 °C for 2 minutes followed by 35 cycles of 94 °C for 30" denaturation, 60.5 °C for 30" annealing temperature and 72 °C for 30'' elongation; followed by 72 °C for 5 minutes. For the SSCP assay, PCR products were mixed with loading buffer and heat-denatured at 95 °C for 10 min, chilled on ice for 10 min and then loaded onto ten percent polyacrylamide gels (acrylamide/bisacrylamide, 37.5:1). The SSCP

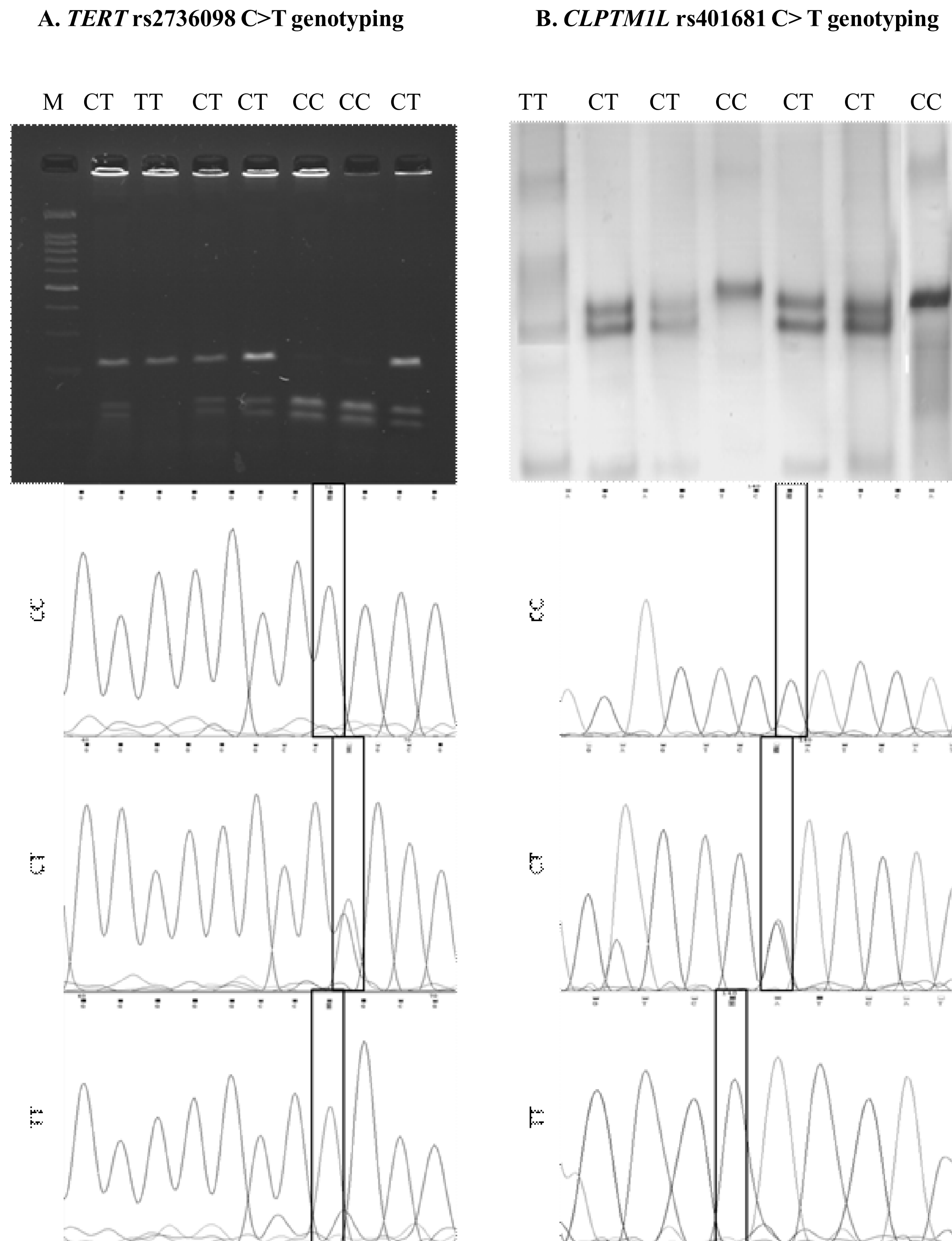

Fig. 1. Genotyping results of *TERT* rs2736098 C>T and *CLPTM1L* rs401681 C>T polymorphisms. A) rs2736098 was genotyped by PCR-RFLP and then the genotypes were validated by direct sequencing; B) rs401681 was genotyped using SSCP patterns and direct sequencing. M: DNA marker

amplicons were run at 110 V, 25 °C, for 14 h in 0.5X TBE buffer. Gels were silver-stained according to the means described by Byun et al. (2009). Three different SSCP patterns of the *CLPTM1L* region were directly sequenced using Macrogen sequencing service (Macrogen Inc., Seoul, Korea) (Fig.1B). Existence of all sequence variations was well defined by performing two separate PCR reactions and subsequent DNA sequencing. The BLAST procedure was carried out by NCBI GeneBank databases (http://blast.ncbi.nlm.nih.gov/) to find the polymorphism locus.

Statistical Analysis

Categorical variables among patients and controls compared using two sided chi-square test and student's t-test. After adjustment for gender, age, and smoking status, genotypic and allelic association tests were evaluated by calculation of the odds ratios (ORs) and ninety-five percent confidence intervals (CIs) using a logistic regression model. The two sided *P* value of <0.05 was considered to be statistically significant in this study. All statistical tests were carried out in Statistical Product and Service Solutions software (v.16.0; SPSS Institute Cary, Chicago, IL).

RESULTS

The clinicopathological characteristics and risk factors among 266 lung cancer cases and 250 cancer-free controls, all Iranian, are summarized in Table 1. The distributions of sex, age, and smoking status among patients and cancer-free controls indicated no statistically significant differences (*P* value > 0.05). Cases included 166 adenocarcinomas, 53 squamous cell carcinomas, and 47 small-cell carcinomas.

Table 1: Baseline characteristics of lung cancer patients and healthy participants

Variables	*Cases (n=266)*	*Controls (n=250)*	*P-value*
Gender			
Male	188	182	0.592[a]
Female	78	1 68	
Mean age (±SD)	62.3±11.2	61.1±10.8	0.251[b]
Smoking Status			
Smoker	101	99	0.704[a]
Never smoker	165	151	
Histological Type			
Adenocarcinoma	166		
Squamous cell carcinoma	53		
Small-cell carcinoma	47		

Note: [a]: Derived from Pearson's chi-square test
[b]: Derived from Student's t-test

Both of SNPs are consistent with Hardy-Weinberg equilibrium in case and the control cohorts (*P*>0.05; data not shown). The distributions and association tests of chromosome 5p15.33 locus comprised of the *TERT* rs2736098 C>T and *CLPTM1L* rs401681 C>T SNPs are presented in Table 2. The T allele of *TERT* rs2736098 (42% in cases and 31% in controls) and of *CLPTM1L* rs401681 (25% in cases and 36.1% in controls) were the minor alleles in this study population.

The regression model adjusted for age, gender, and smoking status revealed that the subjects with *TERT* rs2736098 TT or CT genotypes had a significantly increased risk for lung cancer compared to CC genotype carriers (CT vs CC, adjusted OR=1.58, 95% CI: 1.09-2.30, *P*=0.0167; TT vs CC, adjusted OR=2.64, 95% CI: 1.51-4.64, *P*= 0.0007). Likewise, the dominant model (CT+TT genotype) found a significant association (CT+TT vs CC, adjusted OR=1.78, 95% CI: 1.24-2.53, *P*= 0.0015). In addition, based on allelic association test results (Table 2), the rs2736098 T allele was significantly associated with lung cancer risk (T vs C, adjusted OR=1.62, 95% CI: 1.25-2.09, *P*= 0.0002). When classified by histological type, there was found a significant association between rs2736098 SNP and adenocarcinoma (CT vs CC, adjusted OR=1.82, 95% CI: 1.18-2.83, *P*=0.0069; TT vs CC, adjusted OR=3.37, 95% CI: 1.82-6.26, *P*<0.0001; CT+TT vs CC, adjusted OR=2.11, 95% CI: 1.39-3.19, *P*= 0.0004; T vs C, adjusted OR=1.83, 95% CI: 1.38-2.44, *P*<0.0001) (Table 3).

On the other hand, the *CLPTM1L* rs401681 T allele carriers were inversely associated with lung cancer susceptibility (CT vs CC, adjusted OR=0.58, 95% CI: 0.40-0.85, *P*=0.0044; TT vs CC, adjusted OR=0.35, 95% CI: 0.18-0.66, *P*=0.0012; CT+TT vs CC, adjusted OR=0.53, 95% CI: 0.38-0.76, *P*=0.0004; T vs C, adjusted OR=0.59, 95% CI: 0.45-0.77, *P*=0.0001) (Table 2). Moreover, *CLPTM1L* rs401681 genotype distributions in lung cancer histology types showed a protective association with adenocarcinoma (CT vs CC, adjusted OR=0.65, 95% CI: 0.43-0.98, *P*=0.0405; TT vs CC, adjusted OR=0.42, 95% CI:

Table 2: *TERT* rs2736098, *CLPTM1L* rs401681 polymorphisms and risk of lung cancer

Polymorphism	*Cases (%)*	*Controls (%)*	*Adjusted OR (95% CI)*[a]	*P value*
TERT rs2736098				
Genotypic Association				
CC	90 (33.8%)	119 (47.6%)	1	
CT	128 (48.1%)	107 (42.8%)	1.58 (1.09-2.30)	0.0167
TT	48 (18.1%)	24 (9.6%)	2.64 (1.51-4.64)	0.0007
CT+TT	176 (66.2%)	131 (52.4%)	1.78 (1.24-2.53)	0.0015
Allelic Association				
C allele	308 (58%)	345 (69%)	1	
T allele	224 (42%)	155 (31%)	1.62 (1.25-2.09)	0.0002
CLPTM1L rs401681				
Genotypic Association				
CC	150 (56.2%)	102 (40.9%)	1	
CT	99 (37.5%)	115 (46.0%)	0.58 (0.40-0.85)	0.0044
TT	17 (6.2%)	33 (12.9%)	0.35 (0.18-0.66)	0.0012
CT+TT	116 (43.8%)	148 (59.1%)	0.53 (0.38-0.76)	0.0004
Allelic Association				
C allele	399 (75%)	319 (63.8%)	1	
T allele	133 (25%)	181 (36.2%)	0.59 (0.45-0.77)	0.0001

Note: [a]: Adjusted by age, sex, and smoking status

0.20-0.86, *P*=0.0151; CT+TT vs CC, adjusted OR=0.60, 95% CI: 0.40-0.89, *P*=0.0102; T vs C, adjusted OR=0.64, 95% CI: 0.48-0.87, *P*=0.0046) (Table 3). However, no significant association was found between each SNP and squamous cell, and small-cell carcinomas.

DISCUSSION

In this case-control study, the association between genetic polymorphisms at chromosome 5p15.33 locus, *TERT* rs2736098 C>T and *CLPTM1L* rs401681 C>T, and lung cancer with a cohort of 266 cases and 250 cancer-free healthy controls were conducted for the first time in an Iranian population. The results suggested that T allele carriers of *TERT* rs2736098 and *CLPTM1L* rs401681 were positively and inversely correlated with lung cancer susceptibility, respectively. Furthermore, the similar associations were observed in lung adenocarcinoma subgroup. However, there is no association between *TERT* rs2736098 and *CLPTM1L* rs401681, and squamous cell/small-cell carcinomas. Hence, our study suggested that the *CLPTM1L-TERT* variants may have different roles based on pathological subtypes of lung cancer.

TERT, also known as *TP2*, is a key component of telomerase ribonucleoprotein complex and maintains telomere length (Lantuéjoul et al. 2007) which has a crucial role in chromosomal integrity and stability; hence, it participates in carcinogenesis process of several cancer types (Feldser et al. 2003; Rodier et al. 2005). It has been shown that *TERT* is up-regulated in several cancer types, including lung cancer (Bagheri et al. 2006). Several studies focused on *TERT* rs2736098 polymorphism and susceptibility to lung cancer; however, the functional significance and the molecular mechanism of the synonymous coding SNP rs2736098, located on the second exon of the gene *TERT*, were unknown (Li et al. 2013). Choi et al. (2009) found an association between TT genotype of rs2736098 and lung cancer risk in a Korean population. In Chinese populations, Li et al. (2013) and Wu et al. (2013) also showed higher risk of lung cancer for the rs2736098 TT genotype carriers, particularly lung adenocarcinoma. In agreement with the above researches, the current study found significant associations between rs2736098 polymorphism and lung cancer using various statistical tests (CT vs CC, adjusted OR=1.58, 95% CI: 1.09-2.30, *P*=0.0167; TT vs CC, adjusted OR=2.64, 95% CI: 1.51-4.64, *P*= 0.0007; CT+TT vs CC, adjusted OR=1.78, 95% CI: 1.24-2.53, *P*= 0.0015; T vs C, adjusted OR=1.62, 95% CI: 1.25-2.09, *P*= 0.0002). Similar association pattern was revealed in the lung adenocarcinoma subgroup (CT vs CC, adjusted OR=1.82, 95% CI: 1.18-2.83, *P*=0.0069; TT vs CC, adjusted OR=3.37, 95% CI: 1.82-6.26, *P*<0.0001; CT+TT vs CC, adjusted OR=2.11, 95% CI: 1.39-3.19, *P*= 0.0004; T vs C, adjusted OR=1.83, 95% CI: 1.38-2.44, *P*<0.0001). Notably,

Table 3: The association of the rs2736098 and rs401681 polymorphisms in pathological subgroups

Polymorphism	*Cases (%)*	*Controls (%)*	*Adjusted OR (95% CI)[a]*	*P value*
TERT rs2736098				
Adenocarcinoma				
Genotypic Association				
CC	50 (30.2%)	119 (47.6%)	1	
CT	82 (49.5%)	107 (42.8%)	1.82 (1.18-2.83)	0.0069
TT	34 (20.3%)	24 (9.6%)	3.37 (1.82-6.26)	<0.0001
CT+TT	116 (69.8%)	131 (52.4%)	2.11 (1.39-3.19)	0.0004
Allelic Association				
C allele	182 (55%)	345 (69%)	1	
T allele	150 (45%)	155 (31%)	1.83 (1.38-2.44)	<0.0001
Squamous Cell Carcinoma				
Genotypic Association				
CC	23 (43%)	119 (47.6%)	1	
CT	24 (45%)	107 (42.8%)	1.16 (0.69-2.18)	0.6424
TT	6 (12%)	24 (9.6%)	1.29 (0.48-3.51)	0.6132
CT+TT	30 (57%)	131 (52.4%)	1.18 (0.65-2.15)	0.5775
Allelic Association				
C allele	70 (66%)	345 (69%)	1	
T allele	36 (34%)	155 (31%)	1.14 (0.73-1.78)	0.551
Small-cell Carcinoma				
Genotypic Association				
CC	17 (36%)	119 (47.6%)	1	
CT	22 (48%)	107 (42.8%)	1.44 (0.73-2.85)	0.2956
TT	8 (16%)	24 (9.6%)	2.33 (0.90-6.02)	0.0738
CT+TT	30 (64%)	131 (52.4%)	1.60 (0.84-3.05)	0.149
Allelic Association				
C allele	56 (59.6%)	345 (69%)	1	
T allele	38 (40.4%)	155 (31%)	1.51 (0.96-2.38)	0.0734
CLPTM1L rs401681				
Adenocarcinoma				
Genotypic Association				
CC	89 (53.6%)	102 (40.9%)	1	
CT	65 (39.2%)	115 (46.0%)	0.65 (0.43-0.98)	0.0405
TT	12 (7.2%)	33 (12.9%)	0.42 (0.20-0.86)	0.0151
CT+TT	77 (46.4%)	148 (59.1%)	0.60 (0.40-0.89)	0.0102
Allelic Association				
C allele	243 (73.1%)	319 (63.8%)	1	
T allele	89 (26.9%)	181 (36.2%)	0.64 (0.48-0.87)	0.0046
Squamous Cell Carcinoma				
Genotypic Association				
CC	23 (42.2%)	102 (40.9%)	1	
CT	24 (45.5%)	115 (46.0%)	0.93 (0.49-1.74)	0.8099
TT	6 (12.2%)	33 (12.9%)	0.81 (0.30-2.15)	0.6665
CT+TT	30 (57.8%)	148 (59.1%)	0.90 (0.49-1.64)	0.7273
Allelic Association				
C allele	70 (66.0%)	319 (63.8%)	1	
T allele	36 (34%)	181 (36.2%)	0.91 (0.58-1.41)	0.6626
Small-cell Carcinoma				
Genotypic Association				
CC	16 (34.8%)	102 (40.9%)	1	
CT	23 (48.3%)	115 (46.0%)	1.27 (0.64-2.55)	0.4904
TT	8 (16.8%)	33 (12.9%)	1.55 (0.61-3.94)	0.359
CT+TT	31 (65.2%)	148 (59.1%)	1.33 (0.69-2.57)	0.385
Allelic Association				
C allele	55 (58.5%)	319 (63.8%)	1	
T allele	39 (41.5%)	181 (36.2%)	1.25 (0.80-1.96)	0.3305

Note: [a]: Adjusted by age, sex, and smoking status

although previous studies have shown associations between *TERT* rs2736098 SNP and lung small cell carcinoma (Zhao et al. 2014), and squamous cell carcinoma (Zhang et al. 2014); herein, no significant association was observed in these lung cancer subtypes.

CLPTM1L gene, also called cisplatin resistance-related protein 9 (*CRR9*), encodes a predicted transmembrane protein which likely participates in cellular response to apoptosis, genotoxic stress, and resistance to cisplatin (Yamamoto et al. 2001; Myneni et al. 2013). Although *CLPTM1L* gene is expressed in various tissues, its role is mainly unknown. Polymorphisms localized to *CLPTM1L* are connected with several cancer types, including lung cancer (Wang et al. 2008; Zhao et al. 2014). To illustrate, Ke et al. (2013) and Wang et al. (2008) found the inverse association between the rs401681 T allele, located at intron 13 of *CLPTM1L*, and lung cancer risk in Chinese and Caucasian populations, respectively. Likewise, numerous studies indicated that the rs401681 C allele was associated with the lung cancer development in a Caucasian and an Asian population (McKay et al. 2008; Rafnar et al. 2009; Miki et al. 2010; Yoon et al. 2010; Hu et al. 2011; Pande et al. 2011; Yin et al. 2012; Li et al. 2013). Similarly, in the present study, genotypic and allelic association tests found inverse associations between rs401681 T allele and susceptibility to lung cancer (CT vs CC, adjusted OR=0.58, 95% CI: 0.40-0.85, *P*=0.0044; TT vs CC, adjusted OR=0.35, 95% CI: 0.18-0.66, *P*=0.0012; CT+TT vs CC, adjusted OR=0.53, 95% CI: 0.38-0.76, *P*=0.0004; T vs C, adjusted OR=0.59, 95% CI: 0.45-0.77, *P*=0.0001), and lung adenocarcinoma (CT vs CC, adjusted OR=0.65, 95% CI: 0.43-0.98, *P*=0.0405; TT vs CC, adjusted OR=0.42, 95% CI: 0.20-0.86, *P*=0.0151; CT+TT vs CC, adjusted OR=0.60, 95% CI: 0.40-0.89, *P*=0.0102; T vs C, adjusted OR=0.64, 95% CI: 0.48-0.87, *P*=0.0046). On the other hand, several studies did not find any significant association between the rs401681 polymorphism and lung cancer risk (Zienolddiny et al. 2009; Sun et al. 2013; Zhang et al. 2014; Zhao et al. 2014). In addition, a protective effect of rs401681 CT genotype on lung cancer risk and no association of TT genotype were reported in Asian populations (Bae et al. 2012; Chen et al. 2012; Myneni et al. 2013). Li et al. (2013) and Zhao et al. (2014) discussed that association studies of *CLPTM1L* rs401681 in lung cancer have had different results due to different allele frequencies in various ethnicities, sample sizes, genotyping methodologies, and sources of control group. As an illustration, according to the 1000 Genomes Project Phase 3, *CLPTM1L* rs401681 T allele frequency in African, American, East Asian, South Asian, and European is 0.60, 0.43, 0.32, 0.20, and 0.44, respectively. In this population study, the frequency of *CLPTM1L* T allele was 0.25 in cases and 0.361 in controls. Furthermore, genetic backgrounds may explain these conflicting associations in different populations (Zhang et al. 2014).

To put it in a nutshell, the *TERT* rs2736098 and *CLPTM1L* rs401681 polymorphisms contribute to the susceptibility to lung cancer in Iranian populations. The TERT rs2736098 T allele carriers increased the risk of lung cancer, particularly lung adenocarcinoma. Moreover, the T allele carriers of CLPTM1L rs401681 polymorphism were inversely associated with lung cancer and adenocarcinoma.

CONCLUSION

In summary, this association case-control study demonstrated that the *TERT* rs2736098 T allele was associated with the risk of lung cancer and lung adenocarcinoma incidence in an Iranian population. Moreover, the T allele in *CLPTM1L* rs401681 polymorphism may reduce the risk of lung cancer and adenocarcinoma.

Further studies focusing on the chromosome 5p15.33 polymorphisms are required in larger and in different populations because the researchers stratified analyses had small sample sizes and also frequency of genetic polymorphisms often are not similar in different ethnicities. Notably, future studies need to be conducted in conjunction with environmental risk factors since their importance in lung cancer susceptibility. In addition, the molecular function of the *TERT-CLPTM1L* genes and their SNPs in lung cancer need to be clarified in the further investigations. To date, many studies computationally have predicted the function of genes and polymorphisms in several phenotypes thus, using bioinformatics tools can shed light on the *TERT-CLPTM1L* region genes and its genetic variants.

ABBREVIATION LIST

95 percent confidence intervals (CIs); cisplatin resistance-related protein 9 (*CRR9*); cleft lip and palate trans-membrane 1 like (*CLPTM1L*);

linkage disequilibrium (LD); non-small cell lung cancer (NSCLC); odds ratios (ORs); PCR restriction fragment length polymorphism (PCR-RFLP); small cell lung cancer (SCLC); telomerase reverse transcriptase (*TERT*)

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