Original paper

Investigation of clinicopathological and prognostic association of *TERT* promoter mutation in non-small cell lung cancer in a Turkish population

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Non-small cell lung cancer (NSCLC) is characterized by a complex and heterogeneous molecular basis. Telomerase reverse transcriptase (*TERT*) gene promoter mutations have been implicated in various cancer types. We aimed to investigate the status of *TERT* promoter region mutations in NSCLCs and determine associations of clinicopathological connections, driver mutations, programmed death-ligand 1 (PD-L1) expression, and overall survival (OS) in the Turkish population.
The study included 186 patients diagnosed with NSCLC at a tertiary care center pathology department between 2017 and 2022. *TERT* promoter mutations were present in 2.7% and associated with old age ($p = 0.015$).
The levels of PD-L1 expression were higher in *TERT* mutants ($p = 0.016$). *TERT* mutants had shorter median OS than wild types ($p = 0.006$) and *TERT* mutation was an independent risk factor ($p = 0.004$). *TERT* and EGFR mutations may co-occur and be associated with shorter median OS in patients who continue to receive EGFR treatment ($p < 0.001$).
TERT promoter mutations were associated with high PD-L1 expression and adverse prognosis in NSCLC. In addition, they may play a major role in patients' poor clinical outcomes during EGFR therapy. In conclusion, *TERT* may be a significant parameter for future follow-up and treatment selection of NSCLC.

Key words: clinicopathological features, non-small cell lung cancer, prognosis, *TERT* mutation.

Introduction

Lung cancer is a significant public health issue that has the highest mortality rate among all malignant tumors [1]. Some of the mechanisms related to the pathogenesis of lung cancer are not yet known [2]. Studies have shown that lung cancer is associated with tobacco smoke, environmental effects, family history, and genetic factors [3–5]. Based on its histological characteristics, lung cancer can be divided into two main subgroups: small-cell lung cancer and non-small cell lung cancer (NSCLC) [6]. Non-small cell lung cancer is the most common subtype of lung cancer, accounting for about 85% of cases [7]. Early-stage surgical treatment can provide the best prognosis for NSCLC patients [8]. However, most NSCLC patients are diagnosed at the locally advanced or metastatic stage and thus require comprehensive

treatment. Unfortunately, the prognosis for these patients is poor, with a short average overall survival (OS) [9]. Although new treatments, innovative surgical procedures, and novel clinical approaches have improved the survival of NSCLC patients to some extent in recent years, the 5-year survival rate remains weak, at 10–15% [10, 11].

Non-small cell lung cancer treatments have seen significant developments in recent years, with an increasing focus on treatment plans for the unique molecular profile of each patient's tumor cells [12, 13]. Various mutations that can be targeted for therapies have been identified by understanding the molecular characteristics of lung cancer [12]. Therapies targeting these mutations are more effective and less toxic than standard cancer treatments [14]. Therefore, it is crucial to investigate molecular markers that can improve prognostic outcomes and potentially lead to therapeutic strategies for NSCLC [15].

Telomeres are special structures located at both ends of a chromosome. They consist of repetitive nucleotide sequences (TTAGGG) and a telomere-associated protein complex called shelterin. Telomeres are nucleoprotein complexes that protect the ends of eukaryotic chromosomes from degradation or fusion with adjacent chromosomes. Therefore, they are essential for maintaining genomic integrity and ensuring stability [16]. Telomerase reverse transcriptase (*TERT*) is a ribonucleoprotein polymerase that protects the ends of telomeres. Mutations in the promoter region of the *TERT* gene can lead to increased telomerase activity. This, in turn, helps maintain telomere length and genomic stability. When cancer cells have these activation mutations, they can avoid cellular aging or programmed cell death and continue to divide uncontrollably [17]. In most cells, telomeres shorten after each division. When they reach a critical point, cells stop dividing and aging begins. This mechanism is considered to be a powerful tumor suppressor mechanism in humans [18, 19]. Unfortunately, most tumor cells become immortal by activating telomerase, which is responsible for adding nucleotides to the ends of telomeres. This action prolongs the length of telomeres and cell proliferative potential, preventing aging [20].

Mutations in the *TERT* gene were first observed in melanomas and later identified in multiple common types of cancer, including hepatocellular, bladder, glioblastoma, and thyroid cancer. These studies indicated that the *TERT* promoter mutations may have prognostic, predictive, and therapeutic effects on certain types of cancer [21–24]. However, there is limited research on the correlation between lung cancer and *TERT* mutations. While some studies were not able to detect the presence of *TERT* mutations in NSCLCs, others reported the existence of *TERT* mutations at a low frequency [25]. Additionally, there are conflicting results in the literature regarding the effect of *TERT* mutations on prognosis in NSCLC [25–28]. Furthermore, the low frequency of *TERT* mutations has made it challenging to determine the exact prevalence of these mutations in lung cancer patients [26].

This study aimed to investigate the presence of *TERT* promoter region mutations in NSCLC patients and reveal associations of clinicopathological connections, driver mutations, programmed death-ligand 1 (PD-L1) expression, and OS. For this purpose, *TERT* mutations were screened by a polymerase chain reaction (PCR)-based direct sequencing method in patients with NSCLCs. The effect of *TERT* mutations on prognosis and their potential relationship with demographic, clinicopathological, and molecular markers were examined.

Material and methods

Patient collection and study design

Patients with NSCLC were collected from the archival samples of the Department of Pathology, at University Hospital. The study was approved by the medical ethics committee of the University Hospital Institute and was conducted according to the principles stated in the Declaration of Helsinki. All patients' clinicopathological and follow-up data were obtained from institutional pathology and clinical records. The follow-up time was defined as the time from initial diagnosis to death or the date of last follow-up for living patients. As a result, 186 NSCLC patients with sufficient tumor volume and complete follow-up information were included in the study. The patients were informed about the study, and written consent was obtained. To detect mutations in the promoter region of the *TERT* gene, formalin-fixed and paraffin-embedded (FFPE) representative tumor sample DNAs were extracted and analyzed using PCR-based direct sequencing.

Mutation analysis

TERT mutations are usually investigated using sequencing technologies or real-time PCR methods [29]. Polymerase chain reaction based direct sequencing shows 100% concordance with next-generation sequencing for the detection of TERT promoter mutations [30]. In this study, mutations in the promoter region of the *TERT* gene (chr5, 1,295,228 C > T and 1,295,250 C > T) were analyzed by a PCR-based direct sequencing method with analytical sensitivity of 25% [31, 32]. Firstly, to increase the tumor-cell ratio for DNA isolation, we manually microdissected tumor targets with over 75% viable tumors from 5-μm thick unstained histologic sections.

Then, DNA isolation was performed from FFPE tumor samples of the patients using the QIAamp DNA FFPE Tissue Kit (QIAGEN, 56404). The promoter region of the *TERT* gene was amplified by PCR using forward (5'CAGCGCTGCCTGAAACTC3') and reverse (5'GTCCTGCCCCTTCACCTT3') primers (QIAGEN, 203205). Polymerase chain reaction amplifications were as follows: 15 min at 95°C for initial denaturation; 42 cycles of 95°C for 30 s, 55°C for 30 s, and 72°C for 45 s, with a final extension at 72°C for 10 min. Polymerase chain reaction products were checked by 2% agarose gel electrophoresis. Polymerase chain reaction products were then purified using the PCR Purification Kit (QIAGEN, 28104). Purified PCR products were sequenced with an ABI-3730 48-capillary DNA analyzer (Applied Biosystems) using a BigDye Terminator v3.1 Cycle Sequencing kit (Applied Biosystems; 4337455) according to the manufacturer's instructions. The sequencing results were analyzed using SeqScape Software v3.0 (Applied Biosystems).

Statistical analysis

The mutations in the promoter region of the *TERT* gene were compared with age, gender, smoking, tumor grade, histological subtypes, PD-L1 expression, and driver oncogene mutations (*EGFR*, *ALK*, *ROS1*, *KRAS*, *BRAF V600E*) and OS. Statistical analysis and graphics were performed using IBM SPSS Statistics version 21.0 (IBM Corp., Armonk, NY). Correlations between categorical variables were analyzed using Fisher's exact test or the χ^2 test where appropriate. Continuous variables were given as mean ± standard deviation and analyzed using the Mann-Whitney *U* test. Overall survival was defined as the time from initial diagnosis to death or the date of last follow-up for surviving patients. We used the Kaplan-Meier method to estimate survival curves and compared the distribution of survival using the log-rank test. To further analyze the variable's prognostic effects and build a multivariate prediction model, Cox regression was applied. A multivariate prediction model was built according to univariate regression analysis results to test the independent effect of variables on survival time. The multivariate prediction model was included with statistically significant univariate analysis results. The confidence interval for statistical significance was determined as 95%, and $p < 0.05$ was considered statistically significant in all comparisons.

Results

Patient characteristics

Of the overall cohort, 48 were female (25.8%) and 138 were male (74.2%); the mean age was 62.87. According to the smoking status, 128 were smokers (68.8%) and 58 were non-smokers (31.2%). One hundred forty-nine were metastatic (80.1%), and 37 had no metastasis (19.9%). Patients were classified into two groups according to their tumor stage: I/II and III/IV. There was a significant predominance of advanced stage disease, with 22 patients in stage I/II (11.8%) and 164 in stage III/IV (88.2%). According to histopathological subtypes, 174 were adenocarcinoma (AC) (93.5%) and 12 were squamous cell carcinoma (SqCC) (6.5%). Thirty-seven patients were *EGFR* mutation-positive (19.9%) and 149 were negative (80.1%). Forty-four patients were *KRAS* mutation-positive (23.7%), and 142 were negative (76.3%). Three were *BRAF V600E* mutation positive (1.6%), while 183 patients were negative (98.4%). The results for *ALK* and *ROS1* were available for 184 cases. Ten were *ALK* rearrangement positive (5.4%), and 174 (94.6%) were negative. Four were *ROS1* rearrangement positive (2.2%), and 180 were negative (97.8%). The clinicopathologic features of the cohort are summarized in Table 1.

TERT mutation profile

TERT promoter region mutations were examined by PCR-based direct sequencing analysis. *TERT* promoter region mutations were present in 5 (2.7%) of 186 patients. We identified two different *TERT* promoter region mutations (Fig. 1). These mutations were C228T (3 of 5; 60%) and C250T (2 of 5; 40%).

Association of clinicopathological characteristics with TERT promoter mutations

The distribution of *TERT* mutant and *TERT* wild-type tumors for variables are summarized in Table 1. A statistically significant age difference was found between patients with *TERT* promoter region mutation-positive NSCLCs (median age 62.59 years) and those with *TERT*-negative NSCLCs (median age 73 years) ($p = 0.015$) (Table 1). *TERT* promoter region mutation-positive NSCLCs were observed to have statistically significantly higher PD-L1 levels (median: 55%, mean: 56%) than negatives (median: 3%, mean: 22.24%) ($p = 0.016$) (Fig. 2). In the overall cohort, there was no statistically significant association between *TERT* mutation status and gender, smoking, metastasis, tumor stage, histopathological subgroups, and status of *EGFR*, *KRAS*, *BRAF V600E*, *ALK*, and *ROS1* (Table 1).

Prognostic associations

The median OS for the entire cohort was 23 months (95% confidence interval [CI]: 20.699–25.301 months). There was a statistically significant difference between OS and *TERT* mutation status in the entire cohort ($p = 0.006$) (Fig. 3A). *TERT*

Table I. Clinicopathologic features and TERT promoter region mutations

Parameters	*N*	*TERT* wild type	*TERT* mutant	*p*-value
Patients, *n* (%)	186 (100)	181 (97.3)	5 (2.7)	
Age				
Mean ±SD	62.87 ±9.56	62.59 ±9.48	73 ±7.1	0.015
Gender, *n* (%)				
Male	138 (74.2)	135 (74.6)	3 (60)	0.605
Female	48 (25.8)	46 (25.4)	2 (40)	
Smoking, *n* (%)				
Non-smoker	58 (31.2)	56 (30.9)	2 (40)	0.648
Smoker	128 (68.8)	125 (69.1)	3 (60)	
Metastasis, *n* (%)				
Non-metastatic	37 (19.9)	35 (19.3)	2 (40)	0.259
Metastatic	149 (80.1)	146 (80.7)	3 (60)	
Tumor stage, *n* (%)				
I/II	22 (11.8)	21 (11.6)	1 (20)	0.471
III/IV	164 (88.2)	160 (88.4)	4 (80)	
PD-L1				
Mean ±SD	23.21 ±32.28	22.24 ±31.92	56 ±9.87	0.016
EGFR, *n* (%)				
Positive	37 (19.9)	34 (18.8)	3 (60)	0.055
Negative	149 (80.1)	147 (81.2)	2 (40)	
KRAS, *n* (%)				
Positive	44 (23.7)	44 (24.3)	0 (0)	0.594
Negative	142 (76.3)	137 (75.7)	5 (100)	
BRAF V600E, *n* (%)				
Positive	3 (1.6)	3 (1.7)	0 (0)	1
Negative	183 (98.4)	178 (98.3)	5 (100)	
ALK, *n* (%)				
Positive	10 (5.4)	10 (5.6)	0 (0)	1
Negative	174 (94.6)	169 (94.4)	5 (100)	
ROS1, *n* (%)				
Positive	4 (2.2)	4 (2.2)	0 (0)	1
Negative	180 (97.8)	175 (97.8)	5 (100)	
Histopathology, *n* (%)				
AC	174 (93.5)	170 (93.4)	4 (80)	0.286
SqCC	12 (6.5)	11 (6.1)	1 (20)	

AC – adenocarcinoma, PD-L1 – programmed death-ligand 1, SD – standard deviation, SqCC – squamous cell carcinoma, TERT – telomerase reverse transcriptase

mutation-positive NSCLC patients had significantly shorter median OS (median 12 months; 95% CI: 9.85–14.147 months) compared with *TERT* mutation-negatives (median 24 months; 95% CI: 21.33–26.66 months).

Three patients with *TERT* mutations were simultaneously harboring *EGFR* mutations. There was a significant difference in OS between patients with *EGFR* mutations with or without *TERT* mutations ($p < 0.001$) (Fig. 3B). The median OS for *TERT* wild-type + *EGFR* mutant patients was 39 months (95% CI: 23.762–54.238 months). The median OS for *TERT* mutant + *EGFR* mutant patients was 18 months (95% CI: 8.398–27.602 months).

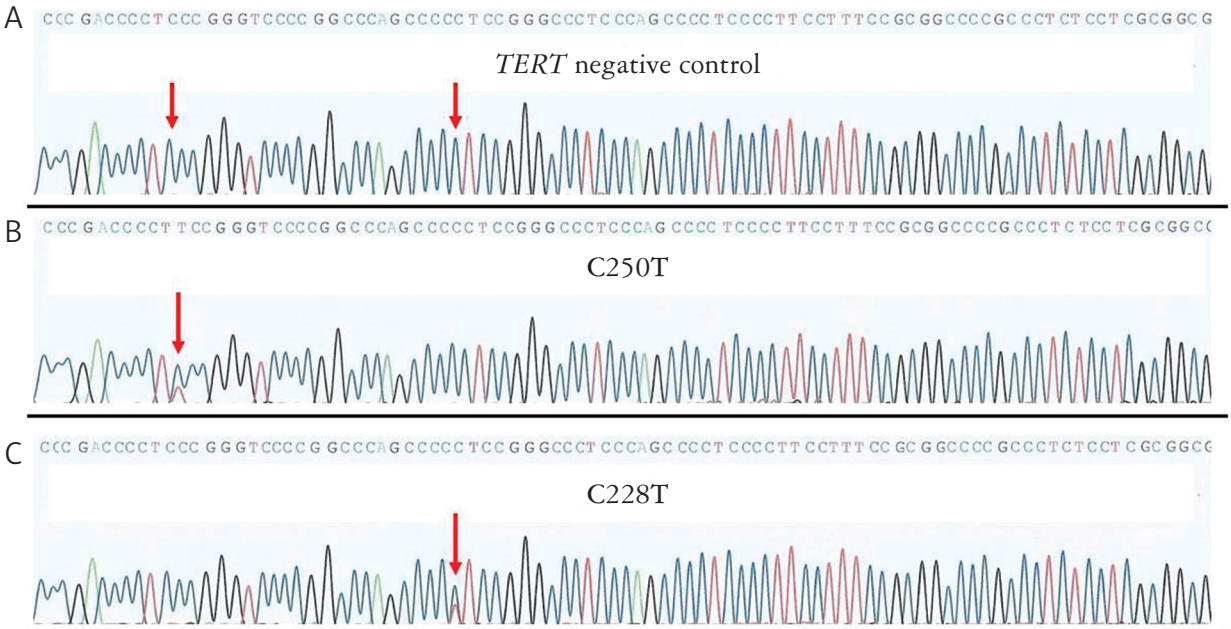
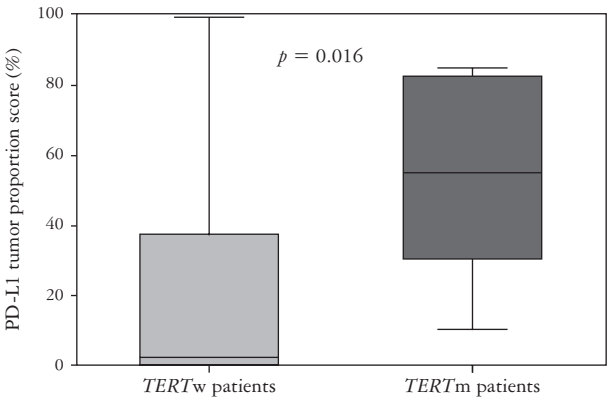


Fig. 1. Samples of sequencing electropherograms of mutated patients for the TERT gene. **A)** Corresponding regions of the IVS-000 polyclonal control DNA's reference sequence. **B)** Sample of sequencing electropherogram of C250T mutated patients. **C)** Sample of sequencing electropherogram of C228T mutated patients



PD-L1 – programmed death-ligand 1, TERT – telomerase reverse transcriptase

Fig. 2. Boxplots of TERT mutant and TERT wild-type patients' programmed death-ligand one levels

Predictors of overall survival

A multivariate prediction model was built according to univariate regression analysis results to test the independent effect of variables on survival time. The variables available for univariate regression were *TERT* mutated, *EGFR* mutated, *TERT* with *EGFR* mutation, PD-L1, age, gender, smoking, metastasis, stage, and histopathology. After performing univariate analysis, according to these results, the following variables were included in the multivariate model: *TERT* mutated, *EGFR* mutated, age, smoking, metastasis, and stage. Univariate and multivariate analysis showed that harboring a *TERT* mutation was significantly associated with OS and an independent risk factor (Table 2). The risk of death for patients harboring a *TERT* promoter region mutation was 4.236 times (95% CI: 1.578–11.377) higher than the reference category *TERT* wild-type group (Table 2).

The results for other variables showed that *EGFR*, age, smoking, metastasis, and stage were significant predictors for OS, while age, smoking, metastasis, and stage were independent risk factors. *TERT* with *EGFR* mutation, PD-L1, gender, and histopathology were not statistically significantly associated with OS (Table 2).

Discussion

The development and spread of lung cancer are linked to factors such as tobacco smoke, environmental elements, and the abnormal regulation of oncogenes and tumor suppressor genes [2–5]. Nowadays, genomic-based biomarkers associated with NSCLC are gaining popularity. The molecular basis of lung cancer is becoming increasingly important in diagnosis, prognosis, and treatment selection [12]. In addition, treatment strategies guided by the molecular profiles of tumor cells have significantly improved the treatment of NSCLC [14].

Telomeres are a unique heterochromatic structure consisting of repetitive nucleotide sequences and a telomere-associated protein complex called shelterin located at the end of the chromosomes. It plays a crucial role in protecting the ends of chromosomes from fusion and distortion and ensuring genomic integrity [16]. The length and stability of telomeres determine cell lifespan and are closely related to cellular

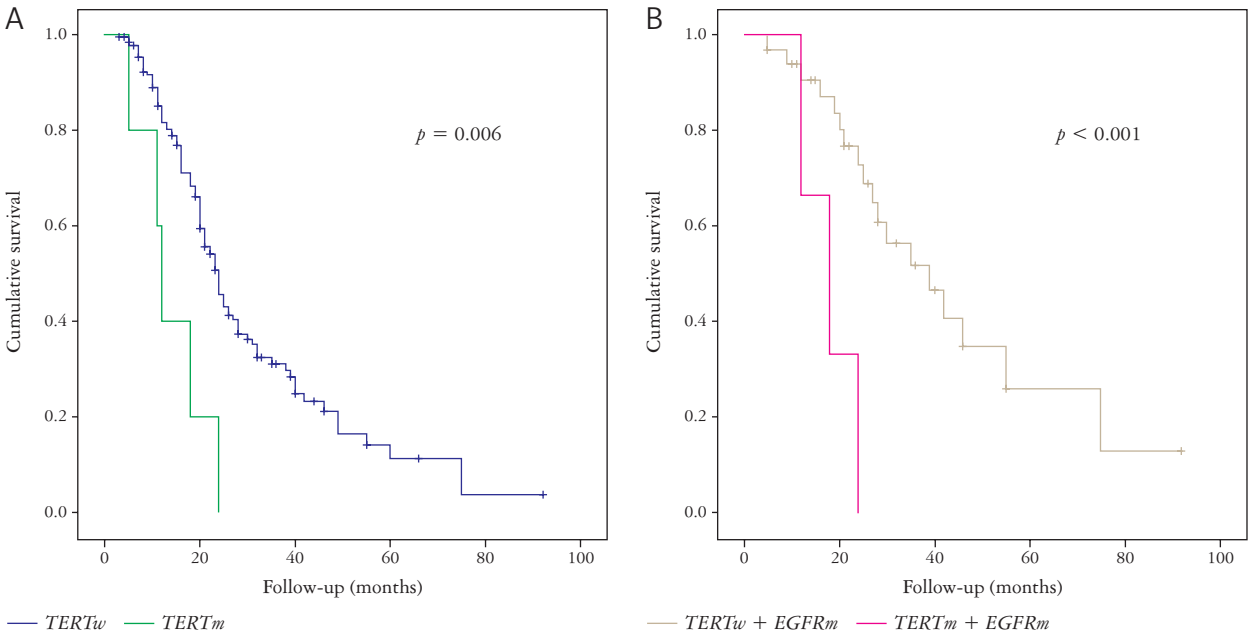


Fig. 3. A) Kaplan-Meier curves show the association between TERT promoter region mutation status (TERTm and TERTw) and overall survival (OS). **B)** OS status according to TERT promoter mutation status in EGFR mutants

aging and tumorigenesis [18, 19]. Telomerase is an essential enzyme for the survival of cells. The main factor that controls telomerase activity is *TERT*. Shortening telomeres or inhibiting telomerase in human cells is considered a natural evolutionary strategy against cancer [33]. In humans, inhibiting telomerase and preserving shorter telomeres is a crucial strategy to fight against tumors. Conversely, the loss of function in the *TERT* gene or reactivation of telomerase is a significant factor that leads to the malignant transformation of human cells [33, 34]. Most immortal tumor cells can evade aging by expressing telomerase, which extends telomere length and cell potential [20].

TERT promoter region mutations have been identified in various types of cancers, and they may also be associated with certain prognostic factors and survival rates [21–24]. It has not been studied before whether there is any association between *TERT* mutations and PD-L1 in NSCLC. A few studies have analyzed *TERT* promoter region mutations in NSCLC, but their relationship with driver mutations remains unclear. Moreover, there are limited studies on the effect of *TERT* promoter mutations on prognosis in NSCLC, and the results are inconsistent [25–28]. In our study, we investigated the presence of *TERT* promoter region mutations in NSCLC patients and evaluated its correlation with prognosis, clinicopathological factors, driver mutations, and PD-L1 expression.

Ma *et al*. identified *TERT* promoter region mutations in 2.57% of NSCLCs and found their presence to be associated with older age (≥ 60 years) [27]. However, they did not find a significant difference between OS or relapse-free survival between *TERT*-mutated and *TERT* wild-type patients. Yuan *et al*. reported that, in the NSCLC cohort part of their study, the frequency of *TERT* promoter region mutations was 5.8%, and significant differences were found only for age in the associations of clinicopathological features with *TERT* status [25]. Jung *et al*. reported a *TERT* promoter mutation rate of 2.2% and found a significant association with regional lymph node invasion [26]. They demonstrated poor prognoses in survival analyses for *TERT*-mutated NSCLCs. In our study, we found a low frequency of *TERT* promoter region mutations in NSCLCs, similar to previous reports (5 of 186; 2.7%); 3 of them had the C228T mutation, and 2 had the C250T mutation. To examine the relationship between *TERT* promoter region mutations and clinicopathological parameters, we compared age, gender, smoking status, metastasis, histopathology, and stage. In our study, patients harboring *TERT* promoter region mutations were older than *TERT* wild-type patients ($p = 0.015$). There were no statistically significant differences in terms of gender, smoking status, metastasis, histopathology, and stage.

The current study investigated the association of *TERT* promoter region mutations with PD-L1 and driver mutations (*EGFR*, *KRAS*, *BRAF V600E*, *ALK*, *ROS1*). The programmed death-ligand 1 expression levels of *TERT* mutant patients were significantly higher than wild types ($p = 0.021$). No statistically significant associations were detected between *EGFR*, *KRAS*, *BRAF V600E*, *ALK*, and *ROS1* and *TERT* promoter region mutations.

Ma *et al*. [27] reported on the effects of *TERT* promoter region mutations on survival, and they did not

Table II. Cox regression analysis in non-small cell lung cancer patients

Covariates	Univariable					Multivariable				
	B	SE	Exp (β)	95% CI Lower-upper	*p*-value	B	SE	Exp (β)	95% CI Lower-upper	*p*-value
TERT mutant	1.185	0.463	3.272	1.321–8.106	0.01	1.444	0.504	4.236	1.578–11.377	0.004
EGFR mutant	–0.658	0.248	0.518	0.318–0.843	0.008	–0.542	0.305	0.582	0.320–1.059	0.076
TERT and EGFR mutant	0.775	0.589	2.171	0.685–6.884	0.188					
PD–L1 (positive/negative)	–0.332	0.200	0.718	0.485–1.061	0.097					
Age	0.032	0.10	1.033	1.013–1.053	0.001	0.036	0.010	1.036	1.017–1.056	< 0.001
Gender	0.401	0.231	1.494	0.951–2.348	0.082					
Smoking	0.710	0.219	2.035	1.326–3.123	0.001	0.523	0.252	1.687	1.029–2.766	0.038
Metastasis	1.042	0.303	2.836	1.566–5.136	0.001	0.972	0.343	2.644	1.394–5.182	0.005
Stage (I –II/III –IV)	0.940	0.317	2.560	1.377–4.760	0.003	0.949	0.356	2.583	1.285–5.191	0.008
Histopathology	–0.518	0.517	0.596	0.216–1.639	0.316					

B – coefficient, CI – confidence interval, Exp (β) – hazard ratio, SE – standard error

find any significant difference in the survival time of patients with *TERT*-mutated and *TERT* wild-type patients. Zalewska-Ziob *et al*. [28] found no association between *TERT* expression and survival in NSCLCs. In contrast, Jung *et al*. reported that *TERT* promoter mutations have an unfavorable prognostic impact on NSCLCs [26]. Our results showed that *TERT* promoter region mutations had a significant adverse prognostic effect ($p = 0.006$). *TERT* mutation-positive NSCLCs had significantly shorter median OS (median 12 months; 95% CI: 9.85–14.147 months) compared with *TERT* wild types (median 24 months; 95% CI: 21.33–26.66 months). In further analysis of the predictors of the prognosis, *TERT* mutation harboring status was an independent risk factor for adverse OS in patients with NSCLCs ($p = 0.004$). The parameters age, metastasis, smoking, and stage had similar results in further analysis for prognosis predictors. We found that NSCLCs with *TERT* mutations had a 4.236 times (95% CI: 1.578–11.377) higher risk than *TERT* wild-type patienTo determine the actual predictive value of the *TERT* promoter region mutation, mutations identified as having prognostic significance in NSCLCs should also be analyzed. Due to the context of cancer, *TERT* mutations may co-occur with other risk-associated mutations in genes such as *BRAF, EGFR*, and *KRAS* [27, 28, 33–35]. Furthermore, the co-occurrence of *TERT* mutations with other risk-associated mutations has been utilized in identifying different types and predicting prognosis in thyroid tumors and gliomas. In the analysis of associations with driver mutations, three patients had co-occurrence of *TERT* and *EGFR* mutations, which was significantly associated with OS ($p < 0.001$). Compared to those only harboring an *EGFR* mutation, patients with both *EGFR* and *TERT* mutations had significantly shorter median survival times (39 months, 95% CI: 23.762–54.238 months, 18 months, 95% CI: 8.398–27.602 months, respectively). *EGFR* mutations are sensitive to targeted therapies and increase the survival results of NSCLCs [36]. Patients with *EGFR* mutations in our cohort were receiving or had received *EGFR*-targeted therapy. We believe that this information is crucial to take into consideration during the clinical follow-up of patients who continue *EGFR* treatment. These results suggested that *TERT* mutation may be a reason for the poor clinical outcome, contrary to the anticipated positive outcome during *EGFR* treatment in NSCLCs, and that it would be valuable to investigate *TERT* mutations in such patients.

Conclusions

Molecular biomarkers are increasingly important for cancer diagnosis, monitoring progress, and determining the treatment. It is crucial to investigate

molecular markers that may help improve prognostic processes and direct potential therapeutic strategies in NSCLCs [15]. The present study identified the presence of *TERT* promoter region mutations in NSCLCs, their prognostic and predictive importance, and details of clinicopathological connections. We found *TERT* promoter region mutation at low frequency in NSCLCs and an independent risk factor for adverse prognosis. Similarly, age, metastasis, smoking, and stage parameters were independent risk factors. We also observed that patients harboring *TERT* promoter region mutations were older. Our study is the first to investigate the association of *TERT* promoter region mutations and PD-L1 in NSCLC. In the present study, we identified that the *TERT* mutant patients had significantly higher PD-L1 expression levels. We found that *TERT* and *EGFR* mutations may co-occur and be associated with shorter median survival times in patients who continue to receive *EGFR* treatment. The results indicated that, contrary to the anticipated positive outcome during *EGFR* treatment in NSCLCs, *TERT* mutations may lead to a poor clinical outcome despite treatment with *EGFR*. These findings suggest that it would be advisable to investigate *TERT* mutations in such patients during the clinical follow-up. Also, this evidence is important for the clinical follow-up of prognosis in NSCLCs. Further larger studies are required to validate our findings due to the low frequency of *TERT* promoter region mutations.

Disclosures

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2. Assistance with the article: None.
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4. Conflicts of interest: None.

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