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ORIGINAL ARTICLE

Tumor mutation burden-assisted risk stratification for papillary thyroid cancer

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Abstract

Purpose Although papillary thyroid cancer (PTC) has a low mortality rate, the rate of recurrence remains relatively high. This study aims to develop a molecular signature to predict the recurrence of PTC.

Methods A total of 333 PTC patients' data from The Cancer Genome Atlas (TCGA) were included. We calculated tumor mutation burden (TMB) and analyzed the mutation status of BRAF and TERT promoter.

Results Tumor recurrence occurred in 17 of 263 cases in TMB-L patients versus 14 of 70 cases in TMB-H patients (hazard ratio [HR], 3.55; 95% confidence interval [CI], 1.75–7.21; $P < 0.001$). The HR for recurrence in TMB-H patients remained significant after adjustment for classical clinicopathologic factors (patient age, gender, extrathyroidal extension and lymph node metastasis). These clinical factors had no effect on recurrence rate in TMB-L patients, but had a strong adverse effect on the prognosis of TMB-H patients. Compared with TMB-L patients lacking mutation, the HR (95% CI) of recurrence for TMB-H patients with coexisting BRAF V600E and/or TERT C228/250 T mutations was 6.68 (2.41–18.57), which remained significant after adjustment for clinicopathological factors. The mutation status of BRAF V600E and TERT C228/250 T had little effect on PTC recurrence in TMB-L patients. Either of the mutation was associated with high recurrence rate in TMB-H patients.

Conclusions The presence of BRAF V600E and/or TERT promoter mutations denotes a high risk of recurrence in TMB-H patients. This represents a powerful molecular prognostic genotype that can help predict patients with the highest risk of recurrence.

Keywords Thyroid cancer · Molecular analysis · Tumor mutation burden · Tumor recurrence

Background

Thyroid cancer is a common endocrine malignancy, and mostly consists of papillary thyroid cancer (PTC), which accounts for 80% to 85% of all the thyroid malignancies [1–3]. PTC includes variant types, such as conventional PTC (CPTC), follicular-variant PTC, and tall-cell PTC, among

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which CPTC is the most common. Although PTC is generally highly curable, approximately 10% of the patients experience tumor progression with aggressive tumor behaviors, high disease recurrence and mortality rates [4–6]. The PTC dynamics complicate clinical risk stratification and decision making in its management. Whereas several clinical features such as patient age, size of the primary tumor, histology, gross extrathyroidal extension (EXT), completeness of resection, involvement of the cervical lymph nodes, or distant metastasis might enable initial risk stratification [7], molecular-based management would enhance the accuracy of the patient stratification [8]. Regrettably, to date, data on the molecular mechanisms for PTCs recurrence remain scant.

The BRAF V600E mutation is the most frequent genetic change in PTC, with a prevalence between 30% and 80% [9–11]. Previous studies have demonstrated an association between BRAF V600E and PTC recurrence [12–14], as well as PTC-specific mortality [15, 16]. However, the high prevalence of BRAF V600E mutation does not encourage recommendation for aggressive treatment against the BRAF V600E PTC. Recently, two exclusive mutations in the promoter region of TERT gene (C228T and C250T) have been reported in thyroid cancer [17–19]. The average rate of occurrence of these two mutations in PTC has been reported to be 4–10% [11, 18, 20]. Studies have consistently shown a strong association between the TERT promoter mutations and aggressive clinicopathologic outcomes of thyroid cancer, suggesting a prognostic role of the TERT promoter mutations [11, 19, 21–24]. Further evidence shows that a coexistence of the BRAF V600E mutation and the TERT promoter mutations is strongly associated with aggressive features and worse prognosis [21, 25–30]. However, the low frequency of either their coexistence or the TERT promoter mutations may invalidate its use as a prognostic marker.

Whereas increasing evidence indicates that gene mutations in PTC can have important prognostic value, there is limited data on the impact of tumor mutation burden (TMB), which quantifies mutations in a tumor. Here, we used a unique multicenter cohort of PTC patients in The Cancer Genome Atlas (TCGA) database [31] to comprehensively interrogate the impact of TMB on the clinical outcomes of PTC.

Methods

Mutation and clinical information of the PTC patients in the TCGA database

Whole-exon mutation data were obtained from the TCGA Genome Data Analysis Center (GDAC) firehose website GDAC (http://firebrowse.org/) and then extracted data on the BRAF V600E mutation and TMB. Data on the TERT promoter mutation was extracted from the TCGA thyroid cancer mark paper [31], with data generated from both the Sanger sequencing and whole genome sequencing. A total of 333 patients with available data on both exon mutations and TERT promoter mutations were included in our analyses. Using the available high risk clinicopathological features (patient age at diagnosis, gender, extrathyroidal extension and lymph node metastasis), we calculated the HR for the risk of recurrence. In this study, recurrence was defined as a strong suspicion of recurrence on imaging and/or histological proof of recurrence, and recurrence-free survival (RFS) was equivalent to disease-free survival. The clinical data for these patients were extracted directly from the TCGA Data Portal (https://tcga-data.nci.nih.gov/tcga/).

Statistical analyses

Comparisons of categorical variables were performed with either Pearson's chi-squared test or Fisher's exact test in cases with a small number. The independent t and Wilcoxon-Mann-Whitney tests were used for normally and non-normally distributed continuous variables, respectively. Survival curves were plotted with the Kaplan-Meier method with log-rank statistical analyses. In addition, Cox proportional hazard regression was used to assess the HR for the risk of recurrence. A two-sided P-value < 0.05 was considered statistically significant. The analyses and data presentation were carried out using IBM SPSS software (25.0) and GraphPad Prism (8.0.1).

Results

TMB and clinicopathologic outcomes in PTC

A total of 333 PTC patients with available data on the exon mutations as well as BRAF and TERT mutations were analyzed and then TMB was calculated. The genetic status for the BRAF V600E and the TERT promoter mutations were also analyzed. Clinical trials have mostly deferred study-specific cut-points using median TMB or dividing patients in tertile or quartile according to measured TMB [32–38]. The TMB median was 0.21/Mb (range, 0–1.26/Mb), while the upper tertile or quartile cutoff was 0.26/Mb or 0.32/Mb, respectively. With the increase of cut-off value, TMB can better predict the recurrence of patients (Fig. 1). Herein, the TMB-H genotype was defined as samples with TMB value higher than the upper quartile, while TMB-L was defined as samples with TMB value lower than the upper quartile. Overall, TMB-H was significantly associated with some of the high risk clinicopathologic characteristics, such as older patient age, EXT and stage III/IV (Table 1). Tumor recurrence was 6.5% (17 of 263) in the TMB-L patients versus 20.0% (14 of 70) in the TMB-H patients (HR, 3.55; 95% CI, 1.75–7.21; $P < 0.001$;

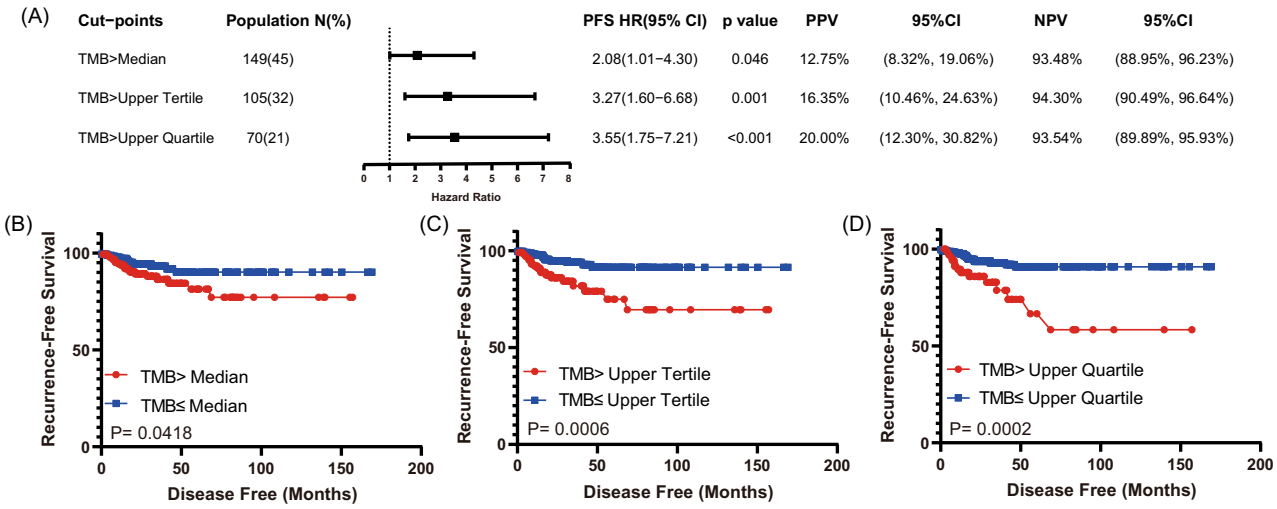

Fig. 1 The effects of TMB on PTC recurrence. A Effects of different TMB cut-off values on PTC recurrence. **B–D** Effects of the median TMB (**B**), the upper tertile TMB (**C**) and the upper quartile TMB (**D**) on tumor recurrence-free survival in PTC

Table 1 Relationship of TMB-H with clinicopathologic outcomes

Clinicopathologic Outcomes	TMB-H ($n = 70$) No. (%)	TMB-L ($n = 263$) No. (%)	*P*
Age at diagnosis, years, Means ± SD	58.4 ± 13.8	43.3 ± 13.3	0.000
Sex, male	27 (38.6)	64 (24.3)	0.018
Extrathyroidal Extension	28 (40.0)	70 (26.6)	0.029
Lymph node metastasis	32 (45.7)	125 (47.5)	0.787
Disease stage, No. of Missing	0	1 (0.4)	
I	16 (22.9)	177 (67.3)	0.000
II	13 (18.6)	20 (7.6)	0.007
III	27 (38.6)	44 (16.7)	0.000
IV	14 (20.0)	21 (8.0)	0.004
III + IV	41 (58.6)	65 (24.7)	0.000
Tumor recurrence	14 (20.0)	17 (6.5)	0.001
Total follow-up, months, Median (IQR)	24 (14–41)	31 (16–52)	0.105

IQR Interquartile range, *SD* Standard deviations

Table S1). The HR of TMB-H for tumor recurrence was significantly high, even after adjustment for patient age and sex, or adjustment for patient age, sex, EXT and lymph node metastasis (LNM) (Table S1). However, the HR of BRAF V600E and/or TERT promoter mutation for tumor recurrence was not significant, even after adjustment for these clinical factors (Table S1).

Impact of TMB-H on RFS in Patients with PTC

Our analysis showed a significant association between TMB-H and decreased recurrence-free probability (Fig. 1D). Several clinical features were used in the clinical risk stratification for PTC recurrence. We compared the effects of TMB-H and the classical clinicopathologic factors on PTC recurrence (Fig. 2). Patient age, EXT and LNM enabled stratification of recurrence risk (Fig. 2A, C, E). However, in the TMB-L cohort, patients aged ≥55, a high-risk clinical factor, did not show elevated risk for recrudescence (Fig. 2B). Similarly, EXT (Fig. 2C) and LNM (Fig. 2D) were also not associated with elevated risk for recrudescence. Furthermore, compared with TMB-L patients, patients aged <55 years with TMB-H were associated with declined recurrence-free probability curve, and the curve declined further in patients aged ≥55 years who were TMB-H (Fig. 2B). Similarly, compared with patients negative for both TMB-H and EXT, presence of EXT alone did not result in a decline in the recurrence-free probability curve, while presence of TMB-H alone had a lower recurrence-free probability, and the curve declined further with the coexistence of both TMB-H and EXT (Fig. 2D). In comparison with patients negative for both TMB-H and LNM, those with either TMB-H or LNM had a lower recurrence-free probability, and the coexistence of both TMB-H and LNM further reduced their probability (Fig. 2F). The positive predictive values (PPVs) of TMB-H combined with clinical high-risk factors were much higher than that of clinical high-risk factors alone (Fig. 2G, Table S2).

Additive effects of coexisting TMB-H and BRAF V600E on PTC recurrence

In this study, BRAF V600E was found in 194 (58.3%) cases (Table S3), and associated with EXT, LNM and stage III/IV. The tumor recurrence was 5.8% (8 of 139) in wild type BRAF (BRAF V600E–negative) patients versus 11.9% (23 of 194) in BRAF V600E patients (HR, 1.77; 95% CI, 0.79–3.97; $P = 0.164$; Table S1). Addition of the TMB

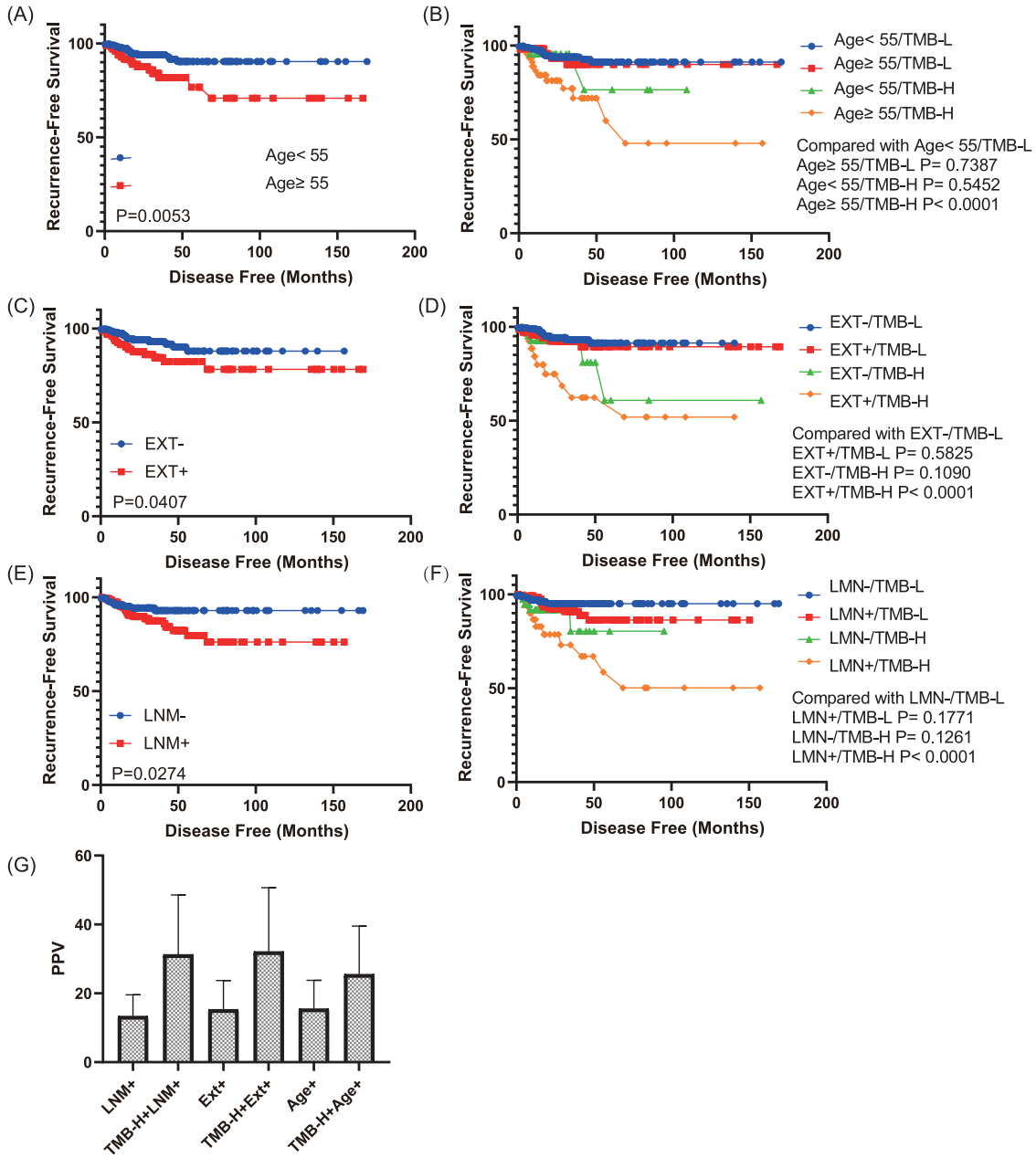

Fig. 2 Kaplan-Meier survival curves of interaction of TMB-H with clinicopathologic risk factors in affecting recurrence-free probability. A Patients age ≥55 years, **B** patients age ≥55 years and TMB-H, **C** EXT, **D** EXT and TMB-H, **E** LNM, **F** LNM and TMB-H. In each panel, P values were from log-rank tests. (**B**) (**D**) and (**F**) were adjusted for multiple comparisons, comparing each stratum with patients negative for both TMB-H and indicated clinicopathologic factor. **G** The PPVs of PTC recurrence

status to the analysis resulted into a better stratification of prognosis. In comparison with the group with wild type BRAF and TMB-L, BRAF V600E alone (BRAF V600E & TMB-L) was significantly associated with EXT and LNM, while TMB-H alone (BRAF V600E negative & TMB-H) was associated with older patients and less patients with stage I PTC (Table 2). In contrast, the coexistence of BRAF V600E and TMB-H was strongly associated with virtually all the classical high-risk characteristics, including older patient age, EXT, LNM and stage III/IV PTC. Patients harboring BRAF V600E and TMB-H had the highest recurrence rate at 28.2% (11 of 39), while TMB-L patients lacking the BRAF V600E mutation had a recurrence rate of 4.6% (5 of 108) (Table S4). The HR of coexisting BRAF V600E and TMB-H for tumor recurrence was 5.91 (95% CI, 2.05 to 17.03). The HR remained significant at 4.54 (95% CI, 1.46 to 14.15) after the first adjustment for patient age at diagnosis and sex, and 3.37 (95% CI, 1.03 to 10.96) after the additional adjustment for tumor behaviors, including EXT and LNM.

Table 2 Impact of BRAF V600E or TMB-H or their coexistence on clinicopathologic outcomes of PTC

Clinicopathologic Outcomes	Wild-Type BRAF & TMB-L ($n = 108$) No. (%)	BRAF V600E & TMB-L ($n = 155$) No. (%)	*P*	Wild-Type BRAF & TMB-H ($n = 31$) No. (%)	*P*	BRAF V600E & TMB-H ($n = 39$) No. (%)	*P*
Age at diagnosis, years	43.2 ± 14.6	43.4 ± 12.4	0.900	54.1 ± 13.2	0.000	61.7 ± 13.2	0.000
Sex, male	25 (23.1)	39 (25.2)	0.708	13 (41.9)	0.039	14 (35.9)	0.122
Extrathyroidal extension	16 (14.8)	54 (34.8)	0.000	5 (16.1)	0.917	23 (59.0)	0.000
Lymph node metastasis	43 (39.8)	82 (52.9)	0.037	7 (22.6)	0.078	25 (64.1)	0.009
Disease stage, No. of Missing	1 (0.9)	0		0		0	
I	78 (72.2)	99 (63.9)	0.125	9 (29.0)	0.000	7 (17.9)	0.000
II	6 (5.6)	14 (9.0)	0.305	12 (38.7)	0.000	1 (2.6)	0.746
III	16 (14.8)	28 (18.1)	0.508	6 (19.4)	0.756	21 (53.8)	0.000
IV	7 (6.5)	14 (9.0)	0.466	4 (12.9)	0.438	10 (25.6)	0.004
III + IV	23 (21.3)	42 (27.1)	0.302	10 (32.3)	0.216	31 (79.5)	0.000
Tumor recurrence	5 (4.6)	12 (7.7)	0.313	3 (9.7)	0.531	11 (28.2)	0.000
Total follow-up, months, Median (IQR)	28 (14–45)	35 (18–56)	0.019	22 (14–30)	0.089	26 (14–47)	0.406

IQR interquartile range, *SD* Standard deviations. *P*-values are from the comparison of the indicated genetic group in the column immediately left of the *P*-value column with the "Wild-Type BRAF & TMB-L" group

Additive effects of coexisting TMB-H and TERT C228/250T on PTC recurrence

The data showed that the mutations in the TERT promoter region had an overall prevalence of 9.0% (30/333), and were significantly associated with older patients, EXT and stage III/IV (Table S3). There was a tumor recurrence rate of 7.9% (24 of 303) in the wild type TERT (TERT C228/250T–negative) cases versus 23.33% (7 of 30) in the TERT C228/250 T cases (HR, 3.17; 95% CI, 1.36–7.36; $P = 0.007$; Table S1). A significant association between TERT C228/250 T and TMB-H was observed (Table S5). Specifically, there was a 2.28% (6 of 263) TERT C228/250 T mutation rate in the TMB-L cases compared to 34.29% (24 of 70) in the TMB-H cases. Conversely, the TMB-H cases were found in 46 of 303 (15.18%) wild type TERT cases versus 24 of 30 (80.00%) C228/250 T cases (odds ratio [OR], 22.35; 95% CI, 8.66–57.67; $P < 0.0001$). Coexistence of TMB-H and TERT mutations was found in 24 of 333 (7.2%) PTC cases (Table 3). In addition, compared with the group negative for both TERT C228/250 T and TMB-H, TERT C228/250 T alone (TERT C228/250 T & TMB-L) was significantly associated with older patients, while TMB-H alone (TERT C228/250 T negative & TMB-H) was significantly associated with both older patients and stage III/IV. In contrast, the coexistence of BRAF V600E and TMB-H was strongly associated with older patients, EXT and stage III/IV (Table 3). Patients harboring both the TERT C228/250 T and TMB-H had the highest recurrence rate of 29.2% (7 of 24), against 6.6% (17 of 257) in patients lacking neither the mutation nor TMB-H (Table S6). The impacts of TERT C228/250 T, TMB-H, and their coexistence on PTC recurrence were shown in Table S6. The HR for the co-occurrence of TERT C228/250 T and TMB-H in tumor recurrence was 5.11 (95% CI, 2.11–12.33, $P < 0.001$). The HR was 3.70 (95% CI, 1.32–10.33, $P = 0.013$) after the first adjustment for patient age at diagnosis and sex, and 2.82 (95% CI, 0.97–8.17, $P = 0.056$) after the additional adjustment for tumor behaviors, including EXT and LNM.

Effect of TMB-H and BRAF V600E or/and TERT C228/250T on RFS of Patients with PTC

Here, we demonstrate that the TMB status had different effects on the RFS of patients with different molecular subtypes (wild type BRAF/TERT, BRAF V600E only, TERT C228/250 T only, or both BRAF V600E and TERT C228/250 T) as shown in Fig. 3(A–D). In patients without BRAF V600E or TERT promotor mutations, TMB status had no effect on RFS (Fig. 3A). On the other hand, TMB-H was significantly associated with higher recurrence rate in patients with only BRAF V600E mutation ($P = 0.0006$) (Fig. 3B). Similarly, in patients harboring only the TERT C228/250 T mutation or both the TERT C228/250 T and

Table 3 Impact of TERT C228/250T or TMB-H or their coexistence on clinicopathologic outcomes of PTC

Clinicopathologic Outcomes	Wild-Type TERT & TMB-L ($n = 257$) No. (%)	TERT C228/250T & TMB-L ($n = 6$) No. (%)	*P*	Wild-Type TERT & TMB-H ($n = 46$) No. (%)	*P*	TERT C228/250T & TMB-H ($n = 24$) No. (%)	*P*
Age at diagnosis, years	43.0 ± 13.3	54.7 ± 9.3	0.035	56.2 ± 12.4	0.000	62.3 ± 15.2	0.000
Sex, male	64 (24.9)	0	0.355	18 (39.1)	0.045	9 (37.5)	0.178
Extrathyroidal extension	68 (26.5)	2 (33.3)	0.928	16 (34.8)	0.245	12 (50)	0.015
Lymph node metastasis	122 (47.5)	3 (50)	0.771	17 (37.0)	0.188	15 (62.5)	0.159
Disease stage, No. of Missing	1 (0.4)	0		0		0	
I	175 (68.1)	2 (33.3)	0.171	12 (26.1)	0.000	4 (16.7)	0.000
II	18 (7.0)	2 (33.3)	0.105	12 (26.1)	0.000	1 (4.2)	0.913
III	43 (16.7)	1 (16.7)	0.586	19 (41.3)	0.000	8 (33.3)	0.084
IV	20 (7.8)	1 (16.7)	0.977	3 (6.5)	0.998	11 (45.8)	0.000
III + IV	63 (24.5)	2 (33.3)	0.991	22 (47.8)	0.001	19 (79.2)	0.000
Tumor recurrence	17 (6.6)	0	0.851	7 (15.2)	0.090	7 (29.2)	0.001
Total follow-up, months, Median (IQR)	31 (16–51)	45 (22–57)	0.961	24 (14–39)	0.174	24 (14–42)	0.321

IQR Interquartile range, *SD* Standard deviations. *P*-values are from the comparison of the indicated genetic group in the column immediately left of the *P*-value column with the "Wild-Type TERT & TMB-L" group

BRAF V600E mutations, TMB-H was associated with poor RFS, despite lack of statistical significance associated with the sample size (Fig. 3C, D). We then explored the effect of the different mutations on the RFS of patients with the same TMB status (Fig. 3E, F). For the TMB-L patients, the RFS curves were modest, regardless of whether they carry mutations or not (Fig. 3E). While, analyses of four groups divided from the TMB-H PTC cohort showed that the RFS curve for patients harboring either mutation was associated with a sharp decline (Fig. 3F), and the curve for TMB-H alone stayed flat. This data showed a strong additive effect in patients' RFS in the co-existence of both TMB-H and BRAF/TERT mutations (Fig. 3G).

Discussion

With the wide application of NGS, more and more attention has been paid to the role of TMB in cancer screening, surveillance, and therapy [39]. The definition of TMB is the total number of mutations per coding area of a tumor genome [40]. It was also called as tumor mutation load or tumor mutational burden [41, 42]. In this study, we evaluated the value of TMB in PTC. We first analyzed the relationship between TMB and clinicopathologic outcomes using the unique PTC cohort. TMB-H was found in 21.02% of the PTC cases. Analysis of the clinicopathological parameters showed a significant association between TMB-H and several high risk clinicopathologic characteristics, such as older age, EXT and higher cancer stage, which were correlated with tumor recurrence. We then explored the effects of TMB-H along with age, EXT or LNM on the risk of recurrence. Each of the high-risk clinicopathologic characteristics alone was not associated with tumor recurrence in patients with TMB-L, but their RFS curves declined sharply in TMB-H patients. Therefore, the risk of recurrence in PTC is TMB-H dependent; and, in the absence of TMB-H, patient age, EXT or LNM alone is not a significant risk factor. In other words, the utility of patient age, EXT or LNM as a prognostic risk factor depends on the TMB status. The data demonstrated that, age, EXT or LNM has a strong adverse effect on the prognosis of PTC patients with TMB-H. Thus, in patients with TMB-H, age, EXT and LNM are important factors in risk stratification and management of PTC.

Many studies have demonstrated that co-occurrence of BRAF V600E and TERT promoter mutations defines an aggressive subgroup of PTCs [21, 25–30]. The average rate of occurrence of these two mutations in PTC has been reported to be 4–10% [11, 18, 20]. The low prevalence of the co-occurrence of BRAF V600E and TERT promoter mutations can only predict the risk of recurrence in a small number of patients. Here we discuss the effect of TMB-H along with BRAF V600E and/or TERT C228/250 T mutations in the recurrence of PTC (Table S7 and Table S8). The

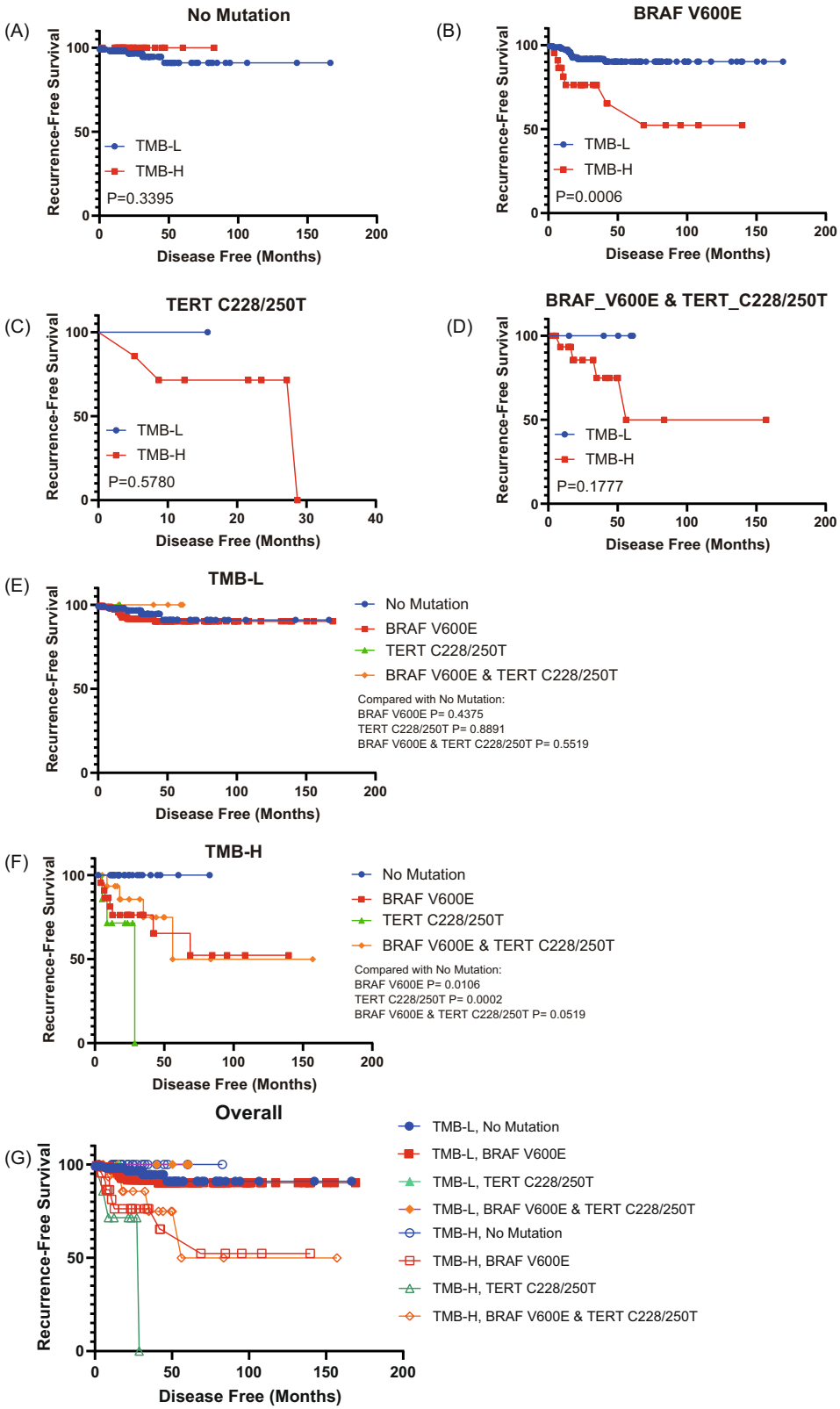

Fig. 3 Kaplan-Meier analyses of impacts of TMB-H or BRAF/TERT mutations or their coexistence on recurrence-free survival. **A** Effects of TMB-H on tumor recurrence-free survival in the wild type BRAF/TERT patients. **B** BRAF V600E mutation only patients. **C** TERT C228/250 T mutations only patients. **D** The coexisting of BRAF V600E and TERT C228/250 T patients. **E**, **F** Effects of different mutations on tumor recurrence-free survival in the TMB-L (**E**) and TMB-H patients (**F**). **G** Effects of the interaction of different mutations with TMB-H on tumor recurrence-free survival in the overall patients. In each panel, P-values were from log-rank tests and P-values in (**E**) and (**F**) were adjusted for multiple comparisons, comparing each stratum with patients negative for mutations

overall prevalence of such molecular alterations was 13.8% (46/333), with a PPV of 30.43% for recurrence, which was much higher than the other genotypes (Table S9). Our analysis on TMB in thyroid cancer demonstrated a significant association between TMB-H and TERT promoter mutations, but not between BRAF V600E and TMB-H. We

showed that, whereas either BRAF V600E or TERT C228/250 T mutations had only a modest effect, their coexistence with TMB-H yielded a robust effect and defined poor clinical outcomes of PTC.

According to previous reports, papillary thyroid microcarcinoma (PTMC), which is defined as a PTC measuring 1 cm or less in maximal diameter, was more likely to be developed by BRAF V600E mutation, and less frequently by TERT promoter mutations, compared to PTC [43]. For most poorly differentiated thyroid cancers (PDTCs) and anaplastic thyroid cancers (ATCs), which are thought to arise from preexisting PTCs based on their frequent co-occurrence in the same tumor specimen [44, 45], the mutations in the TERT promoter display a stepwise increase in frequency along the spectrum of disease progression (9% in PTCs, 40% in PDTCs, and 73% in ATCs) [17, 31, 46–49] and BRAF mutations were less prevalent in advanced tumors compared with PTCs [49, 50]. Therefore, as the malignant degree of the tumor progresses, the frequency of BRAF and TERT mutations changes. It is infeasible to use co-occurrence of BRAF V600E and TERT promoter mutations to predict the prognosis of the thyroid cancers. While, for the TMB value, there was no change among PTMCs and PTCs [43], but ATCs harbored a higher number of mutations than PDTCs and the mutation burden in PDTCs was increased compared with the PTCs [49]. So, the increase in TMB is associated with a higher degree of tumor malignancy, and is more likely to relapse and progress.

Though findings of the present study are inspiring, one limitation is lack of a validation set to add more supporting data. However, as cohorts with both genetic information and long-term follow-up are rare at the current stage, and a new prospective cohort takes time, we feel reasonable to report our findings before validation studies are available. We hope our results would raise more attention to the role of TMB in risk stratification for thyroid cancer and promote more research in this field.

Taken together, the analysis demonstrates the prognostic value of TMB in PTC, and it may also be applicable to PTMC, PDTC, and ATC. The role of TMB-H was robustly established following the analyses on its co-occurrence with BRAF V600E or TERT promoter mutations. Our analysis showed that incorporation of the TMB-H into the risk stratification system for PTC could increase the prognostic robustness of BRAF V600E and/or TERT promoter mutations. This genetic molecular prognostic system may help pinpoint the subgroup of PTC patients with the highest risk and optimize personalized precision treatment.

Data availability

The datasets analyzed during the current study are available in the TCGA Data Portal, [https://tcga-data.nci.nih.gov/tcga/].

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Compliance with ethical standards

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