

TERT Mutations Correlated With the Prognosis of Patients With Hepatitis B-Related Hepatocellular Carcinoma Underwent Curative Hepatectomy

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Research Article

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Abstract

Background To explore the value of *TERT* mutations in predicting the early recurrence and prognosis of hepatitis B-related hepatocellular carcinoma (HCC) patients underwent curative hepatectomy.

Methods A total of 81 patients with hepatitis B-related HCC were enrolled and all patients underwent curative hepatectomy. Associations were sought between *TERT* mutations and recurrence rate within 2 years after hepatectomy, time to progress (TTP) and overall survival (OS).

Results *TERT* mutations (HR: 2.985, 95%CI: 1.158-7.692, $p=0.024$) and Barcelona clinic liver (BCLC) stage B (HR: 3.326, 95%CI: 1.019-10.856, $p=0.046$) were independent risk factors for recurrence within 2 years after hepatectomy. Patients with a *TERT* mutation had poor TTP ($p=0.003$) and OS ($p=0.013$) than others. *TERT* mutations (HR: 2.245, 95%CI: 1.185-4.252, $p=0.013$) and BCLC stage B (HR: 2.132, 95%CI: 1.082-4.198, $p=0.029$) were independent risk factors for poor TTP after curative hepatectomy. A predictive model based on *TERT* mutations and BCLC stage had better ability to predict early recurrence after hepatectomy of HCC patients than any single factor (AUC: 0.688 vs. 0.639, 0.688 vs. 0.607, respectively). Patients with both *TERT* mutations and BCLC stage B had poorer TTP and OS than others ($p=0.001$, $p<0.001$, respectively).

Conclusion *TERT* mutations had ability to predict early recurrence and poor prognosis for hepatitis B-related HCC patients underwent curative hepatectomy.

1. Introduction

Hepatocellular carcinoma (HCC) ranks sixth in terms of incidence among cancers and fourth in terms of cancer related death [1]. Risk factors of HCC included hepatitis B virus (HBV) or hepatitis C virus (HCV) infection, alcoholism and Aflatoxin intake [2, 3], in which HBV infection is the leading risk factor for HCC [3–5]. Although treatments for HCC are more diversified, curative hepatectomy is still the first choice if the state of patients allowed. However, the recurrence after curative treatment is common in HCC patients [6, 7], which caused a negative effect on survival. Hepatocarcinogenesis is a complex multiple process that involves many genetic alterations [8]. Mutations of tumor suppressor genes or oncogenes promote hepatocarcinogenesis and contribute to post-operative recurrence [9]. A study reported that gene mutations correlated with recurrence in a short term after live transplantation [10]. There is a need to find genes associated with prognosis of HCC patients underwent curative hepatectomy.

Key genes mutated in HCC tissues were identified by Next-generation sequencing (NGS). The most common mutated genes included telomerase reverse transcriptase (*TERT*), *TP53* and *CTNNB1* [11–14]. As a catalytic component of telomerase complex, *TERT* could active the telomerase, thereby causing the telomere lengths maintained and cell survival [15]. *TERT* is suppressed in most cells of adults [16]. Overexpression of *TERT* gene increases the activity of telomerase and then leads to telomere synthesis that avoids the senescence and apoptosis caused by telomere shortening [17, 18]. Several studies have uncovered that *TERT* mutations predicted poor prognosis in solid tumors such as breast cancer and lung

cancer [19]. But the correlation between *TERT* mutations and prognosis of HCC patients was still unclear. In this study, we detected *TERT* mutations of HCC tissues by NGS and explore the potential prognostic utility of *TERT* mutations in hepatitis B-related HCC patients underwent curative hepatectomy.

2. Method

2.1 Patients enrollment

From August 2015 to August 2018, patients underwent curative hepatectomy in the Affiliated hospital of Qingdao University were enrolled based on the following criterion: no macrovascular invasion; no extrahepatic metastasis; with positive serum hepatitis B surface antigen (HBsAg); the pathological diagnosis of tumor was HCC; the surgical margins of specimens were negative; no recurrence in the first 2 months after hepatectomy; abnormal preoperative serum alpha-fetoprotein (AFP) dropped to normal level in the first 2 months after hepatectomy; patients agreed with genetic testing of tumor tissues. All patients signed a written informed consent. The study was conducted in strict compliance with the Helsinki Declaration and was approved by the Ethics Committee (ethics approval number: QDFYKYL-20161212). Corresponding epidemiological information and clinicopathological characteristics were collected from the Affiliated Hospital of Qingdao University.

2.2 Identification of genetic alterations and TMB

Formalin-fixed, paraffin-embedded tissues were collected from patients for deep sequencing. The genes were captured and sequenced by genomic profile produced using the NGS-based YuanSu 450 gene panel. Genetic alterations were identified as follows: single nucleotide variants (SNVs) were identified by MuTect (v1.7); Insertion-deletions (InDels) were identified by using PINDEL (V0.2.5). The functional impact of genetic alterations was annotated by SnpEff3.0. Copy number variations (CNV) regions were identified by Control-FREEC (v9.7). Gene rearrangement/fusion were detected through an in-house developed pipeline. Tumor mutational burden (TMB) was measured by an algorithm developed in-house[20].

2.3 Follow up

All patients enrolled in this study were followed up regularly after surgery. During the first 3 months after liver resection, the patients were followed up once a month; during 3-24 months after liver resection, they were followed up every 3 months; and after 2 years, they were followed up every 6 months. The follow up examination included serum AFP, liver function, ultrasonic examination of liver and computed tomography (CT) of lung. Patients received the contrast enhanced CT of upper abdomen annually. When suspected signs of recurrence were found, contrast enhanced CT or magnetic resonance imaging (MRI) was performed to clarify the diagnosis. Time to progress (TTP) was confirmed by imaging examination. All patients were followed up until August 31 2020 or died[20].

2.4 Statistical analysis

All statistical analyses were performed using Statistical Package for the Social Science version 25 for windows (IBM Corp, Armonk, NY). Chi-square test or Fisher's exact test were used to analyze the association between different parameters and *TERT* mutations. Chi-square test or Fisher's exact test were also used to analyze the association between different parameters and recurrence rate with 2 years after hepatectomy, then variables with $p < 0.05$ were subjected to logistic binary regression. Kaplan-Meier method and log-rank test were used to compare the TTP and overall survival (OS) between different groups. Univariate Cox regression model was used to analyze the association between different parameters and TTP/OS. Variables with $p < 0.05$ in univariate analysis were subjected to multivariate Cox regression model. The area under the curve (AUC) of receiver operating characteristic curve (ROC) was calculated to evaluate the predictive value for recurrence within 2 years after hepatectomy of the predictive model, *TERT* mutations and Barcelona clinic liver (BCLC) stage. $P < 0.05$ was considered significant.

3. Results

3.1. Patients characteristics and gene alterations

In our study, 81 patients were enrolled, including 66 males and 15 females. The average age was 56 years old. All patients were HBsAg positive and were not infected with other hepatitis virus. The Child-Pugh grades of patients were A (n = 79) and B (n = 2). There were 20 patients with preoperative serum AFP level above 400 μ g/L before surgery. The median maximum diameter of tumor was 5.5cm (range: 1-19cm) and maximum diameter of tumor in 25 patients were larger than 5cm. There were 20 patients with multiple tumors. The BCLC stage of 10, 53 and 18 patients were 0, A and B respectively. All patients underwent curative hepatectomy, and specimens of 36 patients had microvascular invasion (MVI) in para-carcinoma tissue. The clinicopathological information of enrolled patients were summarized in Table 1.

Table 1
Clinicopathological factors of enrolled patients

| Factors | Number of patients |
|---|--------------------|
| Age (< 56/≥56) | 41/40 |
| Gender (male/female) | 66/15 |
| Hypertension (without/with) | 68/13 |
| Diabetes (without/with) | 73/8 |
| Family history of cancer (without/with) | 53/28 |
| History of alcoholism (without/with) | 56/25 |
| Anti-hepatitis virus treatment (no/yes) | 41/40 |
| Child-Pugh grade (A/B) | 79/2 |
| AFP (< 400/≥400μg/L) | 61/20 |
| Tumor number (single/multiple) | 61/20 |
| Tumor size (≤ 5cm/>5cm) | 56/25 |
| BCLC(0-A/B) | 63/18 |
| MVI (without/with) | 45/36 |
| Edmondson grade (I-II/III-IV) | 43/38 |
| Liver fibrosis (S1S2/S3S4) | 20/61 |

The most commonly mutated genes of enrolled patients were *TP53* (n = 46, 56.8%), *TERT* (n = 34, 42.0%) and *CTNNB1* (n = 20, 24.7%). The alteration type of *TERT* included promoter mutation (n = 31, 91.2%), gene amplification (n = 1, 2.9%), gene arrangement (n = 1, 2.9%) and substitution (n = 1, 2.9%). Among them, the most common alteration was the C > T mutation at -124 bp from the *TERT* ATG start site (n = 29, 85.3%) (Table 2). The median TMB value was 3.9 mutations/Mb and the 75% TMB was 8.9 mutations/Mb. TMB values lower than 8.9 mutations/Mb were considered as TMB-low (TMB-L) (n = 61) while TMB values higher than 8.9 mutations/Mb were considered as TMB-high (TMB-H) (n = 20).

Table 2
Genetic alterations of TERT gene in HCC tissues

| Number of patients | Type of genetic alterations | Genetic alterations |
|--------------------|-----------------------------|---------------------|
| 29 | Promoter mutation | c.-124C > T |
| 1 | Promoter mutation | c.-146C > T |
| 1 | Promoter mutation | c.-57A > C |
| 1 | Substitution | c.425G > A |
| 1 | Gene amplification | amplification |
| 1 | Gene arrangement | SLC12A7/TERT |

3.2. Association between clinicopathological characteristics and *TERT* mutations

In this part, we analyzed the correlation between *TERT* mutations and parameters of HCC patients including twelve clinicopathological characteristics, *TP53* mutations, *CTNNB1* mutations and TMB. By Chi-square test or Fisher exact test, we didn't find significantly correlation between *TERT* mutations and these parameters of patients ($p > 0.05$) (Table 3).

Table 3
Comparison of the clinicopathological characteristics, genes and TMB between wild type and mutant groups

| Factors | TERT | | χ^2 | P |
|---|-------|--------|----------|-------|
| | WT | Mutant | | |
| Age (< 56/ \geq 56) | 22/15 | 19/15 | 0.650 | 0.420 |
| Gender (male/female) | 39/8 | 27/7 | 0.166 | 0.683 |
| Hypertension (without/with) | 38/9 | 30/4 | 0.789 | 0.372 |
| Diabetes (without/with) | 43/4 | 30/4 | N/A | 0.715 |
| Family history of cancer (without/with) | 30/17 | 23/11 | 0.127 | 0.721 |
| History of alcoholism (without/with) | 33/14 | 23/11 | 0.061 | 0.805 |
| AFP (< 400/ \geq 400 μ g/L) | 36/11 | 25/9 | 0.100 | 0.752 |
| Tumor number (single/multiple) | 38/9 | 23/11 | 1.850 | 0.174 |
| Tumor size (\leq 5cm/ $>$ 5cm) | 35/12 | 21/13 | 1.492 | 0.222 |
| BCLC(0-A/B) | 39/8 | 24/10 | 1.752 | 0.186 |
| MVI (without/with) | 29/18 | 16/18 | 1.713 | 0.191 |
| Edmondson grade (I-II/III-IV) | 27/20 | 16/18 | 0.855 | 0.355 |
| TP53 (WT/Mutant) | 24/23 | 11/23 | 2.815 | 0.093 |
| CTNNB1 (WT/Mutant) | 37/10 | 24/10 | 0.702 | 0.402 |
| TMB (< 8.9/ \geq 8.9 mutations/Mb) | 36/11 | 25/9 | 0.100 | 0.752 |

3.3 Survival analysis

3.3.1 HCC recurrence

During the follow-up period, 40 patients had tumor recurrence, and among them 39 patients had tumor recurrence in the first 2 years after hepatectomy. Compared to patients without *TERT* mutation, the proportion of recurrence within the first 2 years after hepatectomy was higher in patients with a *TERT* mutation (64.7% vs. 36.2%, $p = 0.011$). Besides, the recurrence rate within 2 years after hepatectomy was higher in patients with BCLC stage B than others (72.2% vs. 41.3%, $p = 0.020$). By logistic binary regression, we found that *TERT* mutations (HR: 2.985, 95%CI: 1.158–7.692, $p = 0.024$) and BCLC stage B (HR: 3.326, 95%CI: 1.019–10.856, $p = 0.046$) were independent risk factors for recurrence within 2 years after hepatectomy (Table 4).

Table 4

Association of *TERT* mutations and different factors with recurrence within 2 years after hepatectomy

| Variables | Univariate Analysis | | | Multivariate Analysis | | |
|---|---------------------|----------|----------|-----------------------|--------------|----------|
| | n/n | χ^2 | <i>P</i> | HR | 95%CI | <i>P</i> |
| Age(< 56/≥56) | 22/17 | 1.010 | 0.315 | | | |
| Gender (male/female) | 32/7 | 0.016 | 0.899 | | | |
| Hypertension (no/yes) | 35/4 | 1.873 | 0.171 | | | |
| Diabetes (no/yes) | 34/5 | N/A | 0.472 | | | |
| Family history of cancer (no/yes) | 23/16 | 1.387 | 0.239 | | | |
| History of alcoholism (no/yes) | 28/11 | 0.249 | 0.618 | | | |
| Anti-hepatitis virus treatment (no/yes) | 20/19 | 0.013 | 0.908 | | | |
| AFP (< 400/≥400μg/L) | 26/13 | 3.021 | 0.082 | | | |
| BCLC(0-A/B) | 26/13 | 5.372 | 0.020 | 3.326 | 1.019–10.856 | 0.046 |
| Liver fibrosis (S1S2/S3S4) | 7/32 | 1.839 | 0.175 | | | |
| Edmondson grade (I-II/III-IV) | 21/18 | 0.017 | 0.895 | | | |
| MVI (no/yes) | 19/20 | 1.424 | 0.233 | | | |
| TP53 (WT/Mutant) | 17/22 | 0.004 | 0.947 | | | |
| TERT (WT/Mutant) | 17/22 | 6.435 | 0.011 | 2.985 | 1.158–7.692 | 0.024 |
| CTNNB1 (WT/Mutant) | 27/12 | 1.494 | 0.222 | | | |
| TMB (< 8.9/≥8.9Muts/Mb) | 28/11 | 0.499 | 0.480 | | | |

3.3.2 TTP and OS

Kaplan-Meier analysis and log-rank test were used in survival analysis. In analysis of TTP, the 75% TTP was 11.9 months and 5.4 months in patients with a *TERT* mutation and others respectively. The difference between two groups was statistically significant ($p = 0.003$) (Fig. 1). In analysis of OS, the 75% OS of patients with a *TERT* mutation was 36.9 months, while patients without *TERT* mutation didn't reach the 75% OS. There were significant difference between two groups ($p = 0.013$) (Fig. 2). Sixteen variables were examined in cox regression model to reveal the correlated risk factors for TTP and OS. For TTP, serum AFP level $\geq 400\mu\text{g/L}$, BCLC stage B and *TERT* mutations were significantly correlated with shorter TTP of HCC patients in the univariate analysis ($p < 0.05$). Multivariate analysis showed that *TERT* mutations (HR: 2.245, 95%CI: 1.185–4.252, $p = 0.013$) and BCLC stage B (HR: 2.132, 95%CI: 1.082–4.198, $p = 0.029$) were independent risk factors for poor TTP (Table 5). For OS, serum AFP level $\geq 400\mu\text{g/L}$, MVI, Edmondson grade I-II, BCLC stage B and *TERT* mutations were significantly correlated with shorter OS of

HCC patients in the univariate analysis. Multivariate analysis showed that BCLC stage B (HR: 4.071, 95%CI: 1.266–13.099, $p = 0.019$) and MVI (HR: 5.040, 95%CI: 1.090-23.298, $p = 0.038$) were independent risk factors for OS of HCC patients underwent hepatectomy while the TERT mutation was not ($p = 0.063$) (Table 6).

Table 5
Univariate and multivariate analysis of indexes for TTP by cox regression model

| Variables | Univariate Analysis | | | Multivariate Analysis | | |
|---|---------------------|-------------|-------|-----------------------|-------------|-------|
| | HR | 95%CI | P | HR | 95%CI | P |
| Age(< 56/≥56) | 1.626 | 0.868–3.047 | 0.129 | | | |
| Gender (male/female) | 0.918 | 0.406–2.076 | 0.837 | | | |
| Hypertension (no/yes) | 0.533 | 0.189–1.498 | 0.232 | | | |
| Diabetes (no/yes) | 1.448 | 0.566–3.701 | 0.440 | | | |
| Family history of cancer (no/yes) | 1.023 | 0.572–1.830 | 0.939 | | | |
| History of alcoholism (no/yes) | 1.373 | 0.729–2.587 | 0.327 | | | |
| Anti-hepatitis virus treatment (no/yes) | 1.048 | 0.564–1.949 | 0.882 | | | |
| AFP (< 400/≥400µg/L) | 1.977 | 1.018–3.841 | 0.044 | - | - | 0.199 |
| BCLC(0-A/B) | 2.504 | 1.285–4.878 | 0.007 | 2.132 | 1.082–4.198 | 0.029 |
| Liver fibrosis (S1S2/S3S4) | 1.363 | 0.628–2.960 | 0.433 | | | |
| Edmondson grade (Ⅰ-Ⅱ/Ⅲ-Ⅳ) | 1.030 | 0.552–1.921 | 0.926 | | | |
| MVI (no/yes) | 1.623 | 0.872–3.021 | 0.126 | | | |
| TP53 (WT/Mutant) | 0.985 | 0.528–1.838 | 0.963 | | | |
| TERT (WT/Mutant) | 2.502 | 1.335–4.690 | 0.004 | 2.245 | 1.185–4.252 | 0.013 |
| CTNNB1 (WT/Mutant) | 1.257 | 0.638–2.476 | 0.509 | | | |
| TMB (< 8.9/≥8.9Muts/Mb) | 1.369 | 0.683–2.745 | 0.376 | | | |

Table 6
Univariate and multivariate analysis of indexes for OS by cox regression model

| Variables | Univariate Analysis | | | Multivariate Analysis | | |
|---|---------------------|------------------|-------|-----------------------|------------------|-------|
| | HR | 95%CI | P | HR | 95%CI | P |
| Age(< 56/≥56) | 0.341 | 0.092– 1.258 | 0.106 | | | |
| Gender (male/female) | 1.473 | 0.397– 5.457 | 0.562 | | | |
| Hypertension (no/yes) | 2.822 | 0.836– 9.527 | 0.095 | | | |
| Diabetes (no/yes) | 1.801 | 0.393– 8.262 | 0.449 | | | |
| Family history of cancer (no/yes) | 1.292 | 0.409– 4.079 | 0.662 | | | |
| History of alcoholism (no/yes) | 0.454 | 0.099– 2.072 | 0.308 | | | |
| Anti-hepatitis virus treatment (no/yes) | 0.700 | 0.222– 2.209 | 0.543 | | | |
| AFP (< 400/≥400µg/L) | 4.755 | 1.508– 14.997 | 0.008 | - | - | 0.113 |
| BCLC(0-A/B) | 5.967 | 1.891– 18.830 | 0.002 | 4.071 | 1.266– 13.099 | 0.019 |
| Liver fibrosis (S1S2/S3S4) | 0.945 | 0.255– 3.499 | 0.933 | | | |
| Edmondson grade (Ⅰ-Ⅱ/Ⅲ-Ⅳ) | 6.495 | 1.419– 29.717 | 0.016 | - | - | 0.162 |
| MVI (no/yes) | 6.753 | 1.478– 30.854 | 0.014 | 5.040 | 1.090- 23.298 | 0.038 |
| TP53 (WT/Mutant) | 4.052 | 0.887– 18.503 | 0.071 | | | |
| TERT (WT/Mutant) | 4.532 | 1.225– 16.763 | 0.024 | - | - | 0.063 |
| CTNNB1 (WT/Mutant) | 1.453 | 0.436– 4.836 | 0.543 | | | |
| TMB (< 8.9/≥8.9Muts/Mb) | 2.224 | 0.705– 7.020 | 0.173 | | | |

3.3.3 A predictive model

In order to evaluate the predictive value of 2 independent risk factors (*TERT* mutations and BCLC stage B) for recurrence within 2 years after curative hepatectomy in hepatitis B-related HCC patients, we established a predictive model. In this model, the study population was divided into 3 groups: group 1 (without any risk factor), group 2 (with 1 risk factor) and group 3 (with 2 risk factors). ROC curves were generated to evaluate the ability of the model to forecast postoperative tumor relapse. As shown in Table 7 and Fig. 3, the model possessed the greater AUC than *TERT* mutations or BCLC stage (AUC: 0.688 vs. 0.639, 0.688 vs. 0.607, respectively). Therefore, the model had better ability to predict postoperative recurrence than *TERT* mutations or BCLC stage. Compared to patients in group 1 and group 2, the proportion of recurrence within 2 years after hepatectomy was higher in group 3 ($p = 0.006$) (Table 8). Besides, Kaplan-Meier curves showed that patients in group 3 had poorer TTP and OS than other groups ($p = 0.001$, $p < 0.001$, respectively) (Fig. 4, 5).

Table 7
AUCs, 95%CI, and P values of different parameters in predicting recurrence for HCC patients after curative surgery

| Factors | AUC | 95%CI | P |
|----------------|-------|-------------|-------|
| TERT mutations | 0.639 | 0.517–0.761 | 0.031 |
| BCLC stage | 0.607 | 0.483–0.731 | 0.097 |
| The model | 0.688 | 0.571–0.804 | 0.004 |

Table 8
The predictive model and recurrence within 2 years after hepatectomy

| Factor | Group 1 | Group 2 | Group 3 | P |
|--|---------|---------|---------|-------|
| Recurrence within 2 years after surgery | 12 | 19 | 8 | 0.006 |
| No recurrence within 2 years after surgery | 27 | 13 | 2 | |

4. Discussion

TERT has been reported to be the most common mutated gene in HCC patients, with the frequency ranged from 51–60% [11–14]. In this study, the mutation frequency of *TERT* was 42.0%, which was lower than TP53 (56.8%). The difference between our study and previous studies may be attributed to the different hepatitis backgrounds. In previous studies, HCV infection was the most common in enrolled patients [11–14]. While in our study, all patients were HBsAg positive and without HCV infection. HBV could active the telomerase by inverting into the *TERT*, which provided an alternative mechanism of reactivating telomerase and might cause the reduced rate of *TERT* mutations [21, 22].

In our study, *TERT* mutations consisted of promoter mutation, substitution, amplification and rearrangement. *TERT* promoter mutation (85.3%) was the most common alteration, which was consistent with results of a previous study [22]. The hot spots of *TERT* promoter mutation were situated at the - 124

bp and - 146 bp from the *TERT* ATG start site [22]. The most frequent mutation was c.-124C > T (85.3%) in our cases. But we only found one case with mutation of c.-146C > T. Except for the two hot spots, we also found a A > C substitution at the - 57 bp, which had been reported in melanoma [23]. Other alterations of *TERT* gene just occupied a small proportion.

Telomeres are DNA-protein structure located at the end of chromosomes. Telomeres shorten accelerates along with the round of cell division and leads to cell death finally [24]. Cancer cells keep continuous division by reactivation of telomerase and maintaining of telomere length. Telomerases are in charge of the telomere synthesis, which consist of *TERT*, telomerase RNA component and core enzyme [16]. Reactivation of telomerase is a crucial event in hepatocarcinogenesis [16]. There are four mechanisms of telomerase reactivation: (a) through *TERT* promoter mutations, (b) through HBV insertion into promoter, (c) through *TERT* amplification and (d) through *TERT* translocation [15]. *TERT* controlled the activity of telomerase and is suppressed in most cells of adults [16]. *TERT* mutations would lead to the overexpression of *TERT* [25–27].

In this study, we didn't find the significant correlation between *TERT* mutations and clinicopathological characteristics of HCC patients such as tumor size, tumor number, MVI, BCLC stage and differentiation. The result was similar with the researches of Kwa et al [28] and Lee et al [29]. Previous studies have reported that *TERT* mutations could lead to overexpression of *TERT* in several cancers. For example, transcriptional activity of *TERT* with c.-124C > T, c.-146C > T or c.-57A > C was higher than wild type in melanoma [23, 30]; Cao et al [31] reported that amplification of *TERT* increase the expression of *TERT*; Bayard et al [32] reported that rearrangement of *TERT* could induce overexpression of *TERT*. In survival analysis, we found that *TERT* mutations correlated with shorter TTP and OS of HCC patients underwent hepatectomy. We inferred that this result might be related with the overexpression of *TERT* caused by *TERT* mutation. Jeong et al [33] reported that *TERT* overexpression predicted poor intrahepatic recurrence free survival, this result was similar with ours. Besides, we found that BCLC stage and *TERT* mutations were independent risk factors for recurrence within 2 years after hepatectomy. A predictive model based on the BCLC stage and *TERT* mutations showed better predictive value for early recurrence after curative hepatectomy than any single factor. Patients with both BCLC stage B and *TERT* mutations had poorer prognosis than others. The predictive model may help us to identify patients with high risk of recurrence after hepatectomy. For these patients, more frequent surveillance and intervene in time was of vital importance.

This study has some limitations. First, this is a monocenter and retrospective study, multicenter and prospective studies are needed to acquire more representative and convincing results. Second, all patients enrolled were with HBV infection and without other hepatitis virus. Thus, our result may not applicable to HCC patients infected with other hepatitis virus. Third, due to the limitation of sample size, we couldn't analyze the correlation between different mutations of *TERT* and prognosis of HCC patients.

5. Conclusion

In conclusion, promoter mutation was the most common alteration in *TERT*. *TERT* mutations and BCLC stage B were significantly associated with a high risk of early recurrence and predicted poor TTP in HCC patients underwent curative hepatectomy. The predictive model based on the two factors had better ability to predict short postoperative recurrence of HCC patients than *TERT* mutations or BCLC stage.

Declarations

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Competing interests

The author(s) declare no competing interests.

Data Availability

The datasets analyzed during the current study are available from the corresponding author on reasonable request.

Author contributions

Provision of study material or patients: Liqun Wu; Collection and assembly of data: Liqun Wu, Kangjian Song, Junyu Huo, Fu He, Qingwei Zhu; Data analysis and interpretation: Kangjian Song, Junyu Huo; Manuscript writing: Kangjian Song. All authors contributed to conception and design; took part in drafting and revising the article; gave final approval of the version to be published; and agreed to take responsibility and be accountable for the consents of the article.

Ethics approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The study was approved by the Ethics Committee of the Affiliated Hospital of Qingdao University (ethics approval number: QDFYKYL-20161212). Informed consent was obtained from all patients included in our study. All participants and contributors of this study have signed informed consent for publication.

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Figures

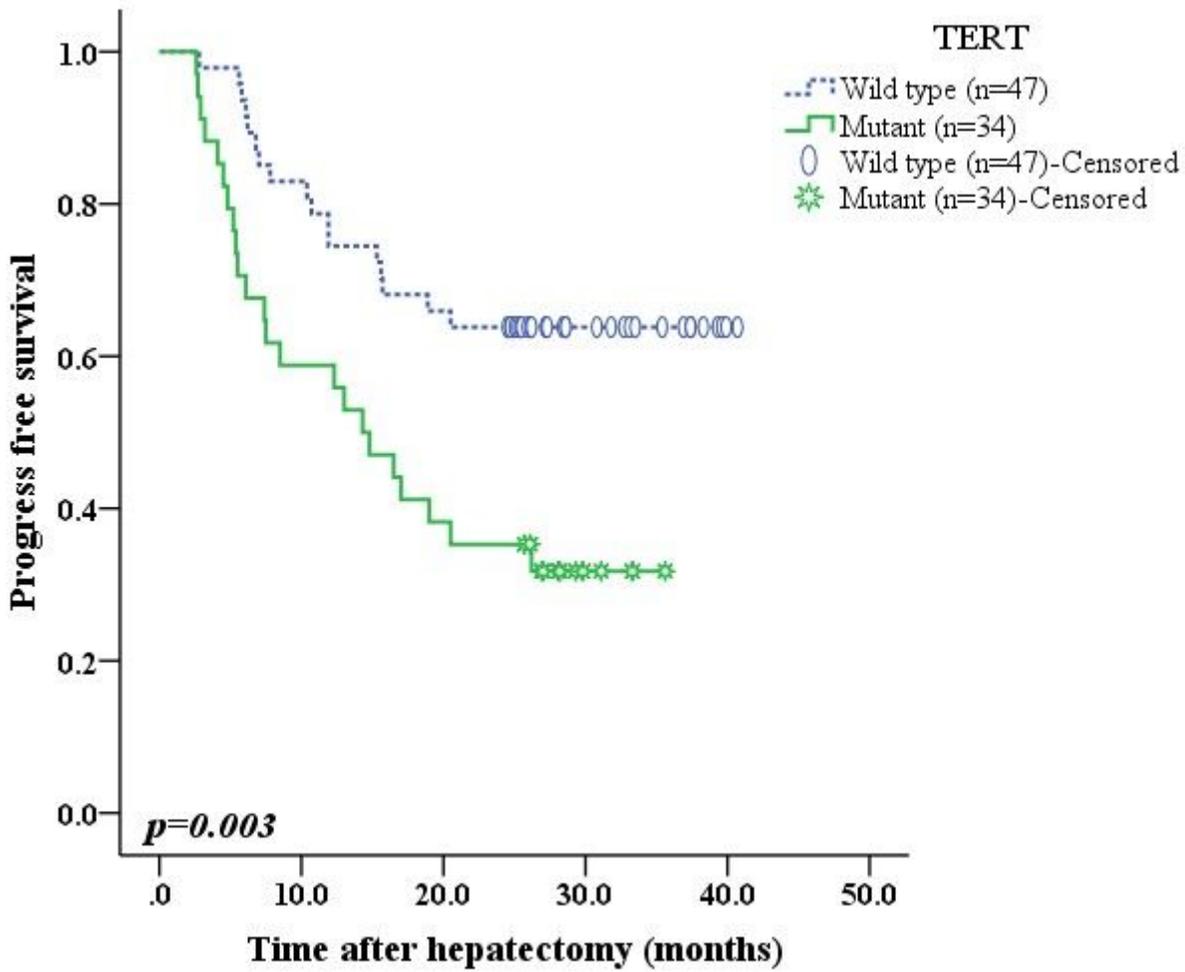


Figure 1

Kaplan-Meier curve showed patients with a TERT mutation had poorer TTP than other patients ($p=0.003$)

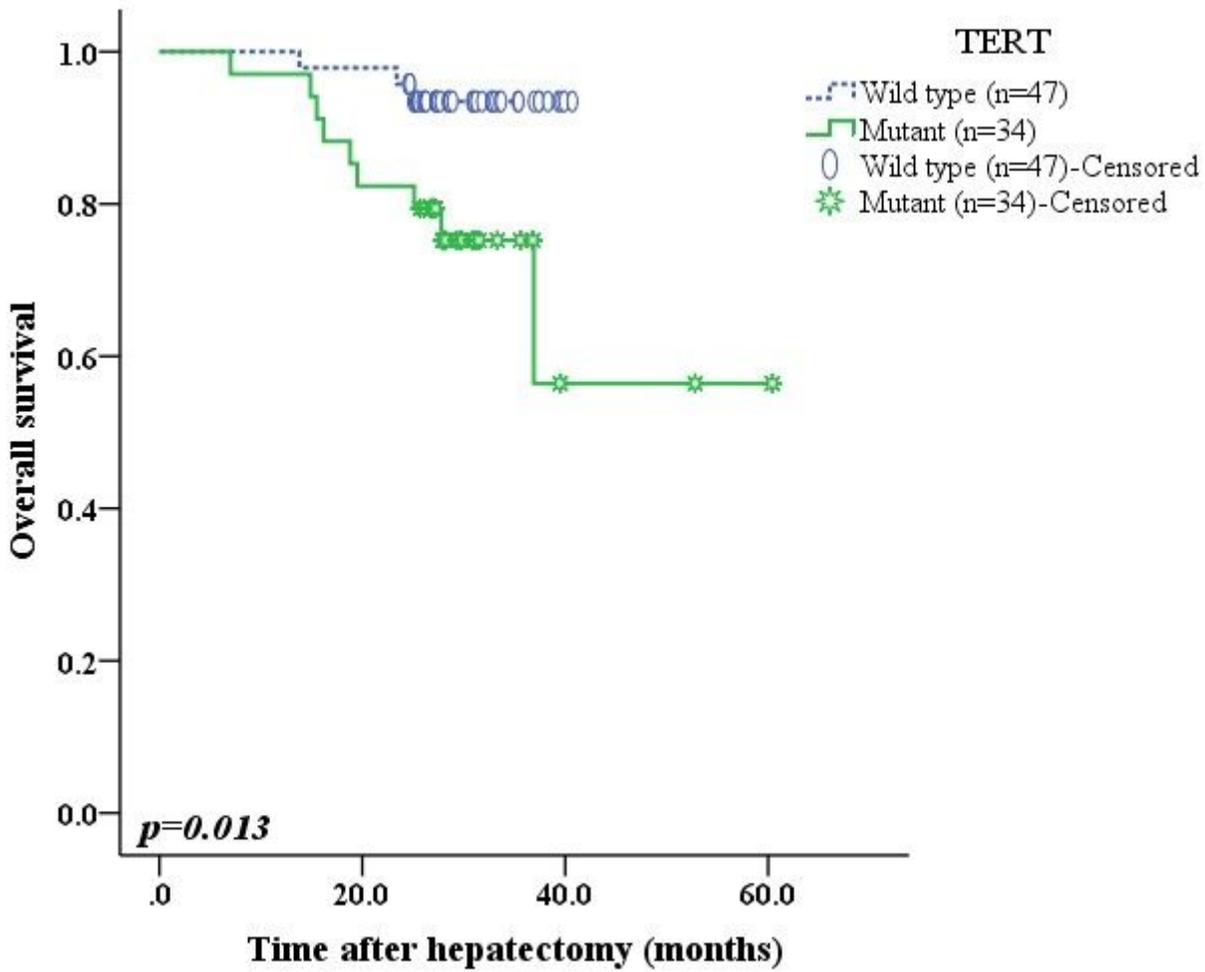


Figure 2

Kaplan-Meier curve showed patients with a TERT mutation had poorer OS than other patients ($p=0.013$)

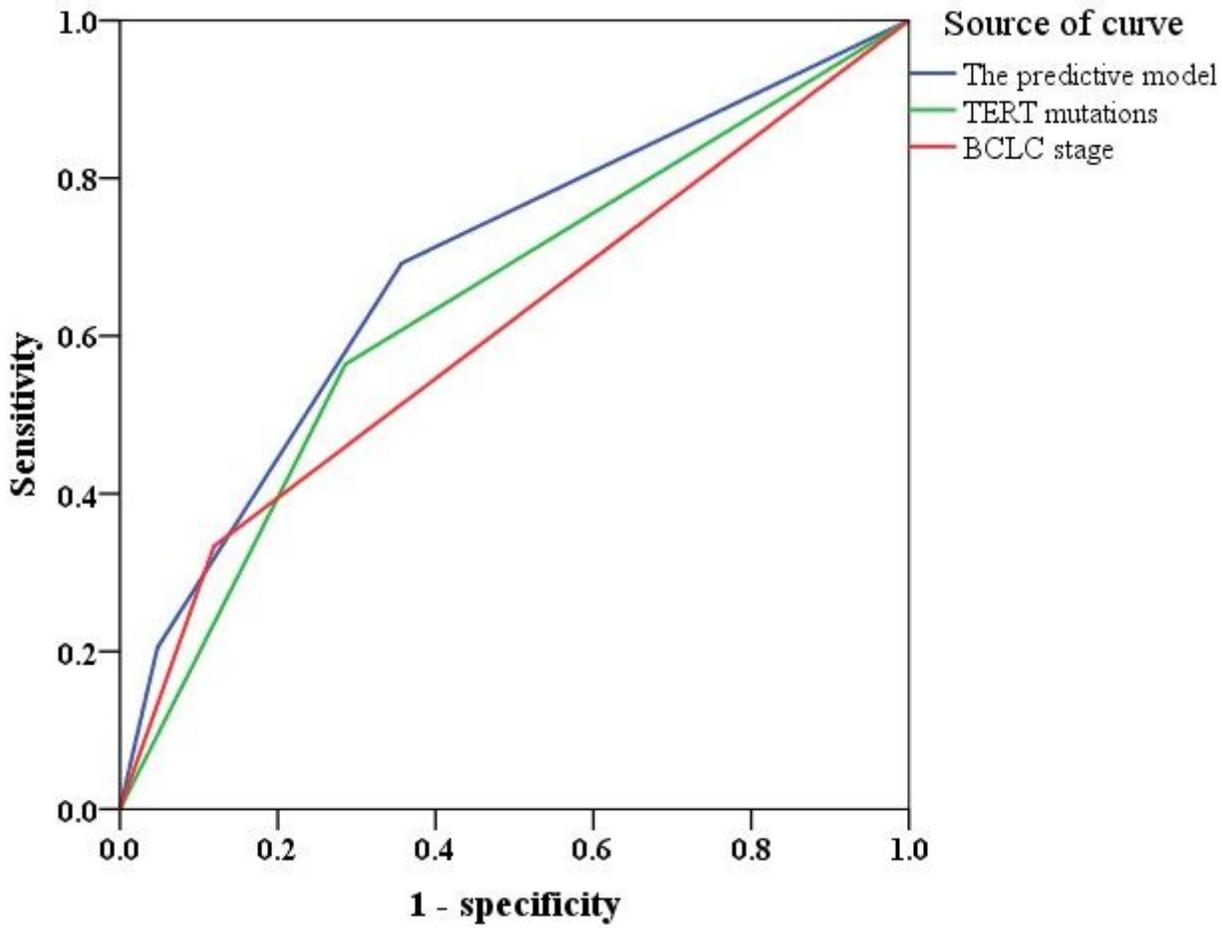


Figure 3

The predictive model had greatest predictive ability for recurrence within 2 years after hepatectomy compared to BCLC stage or TERT mutations

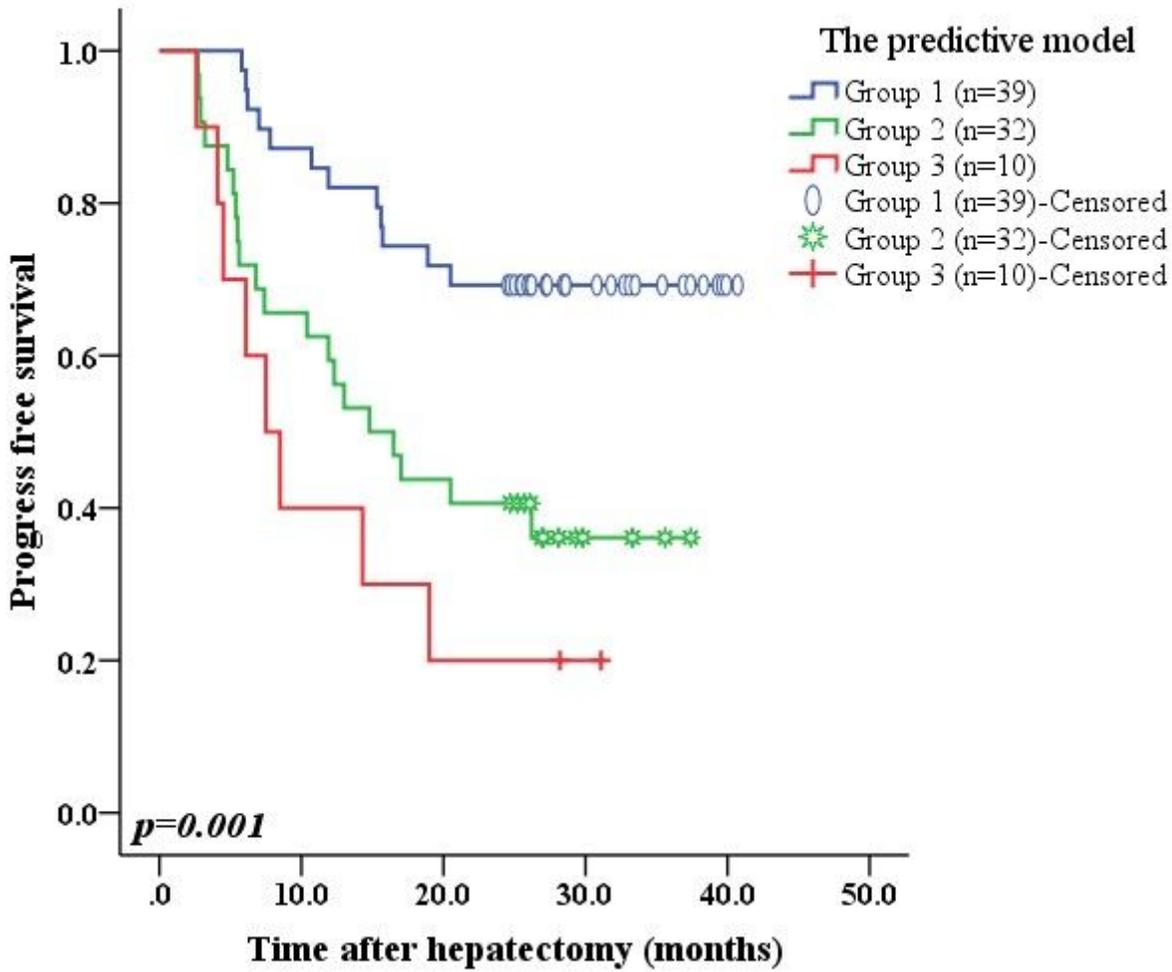


Figure 4

Kaplan-Meier curve showed the difference of TTP between three groups was statistically significant ($p=0.001$)

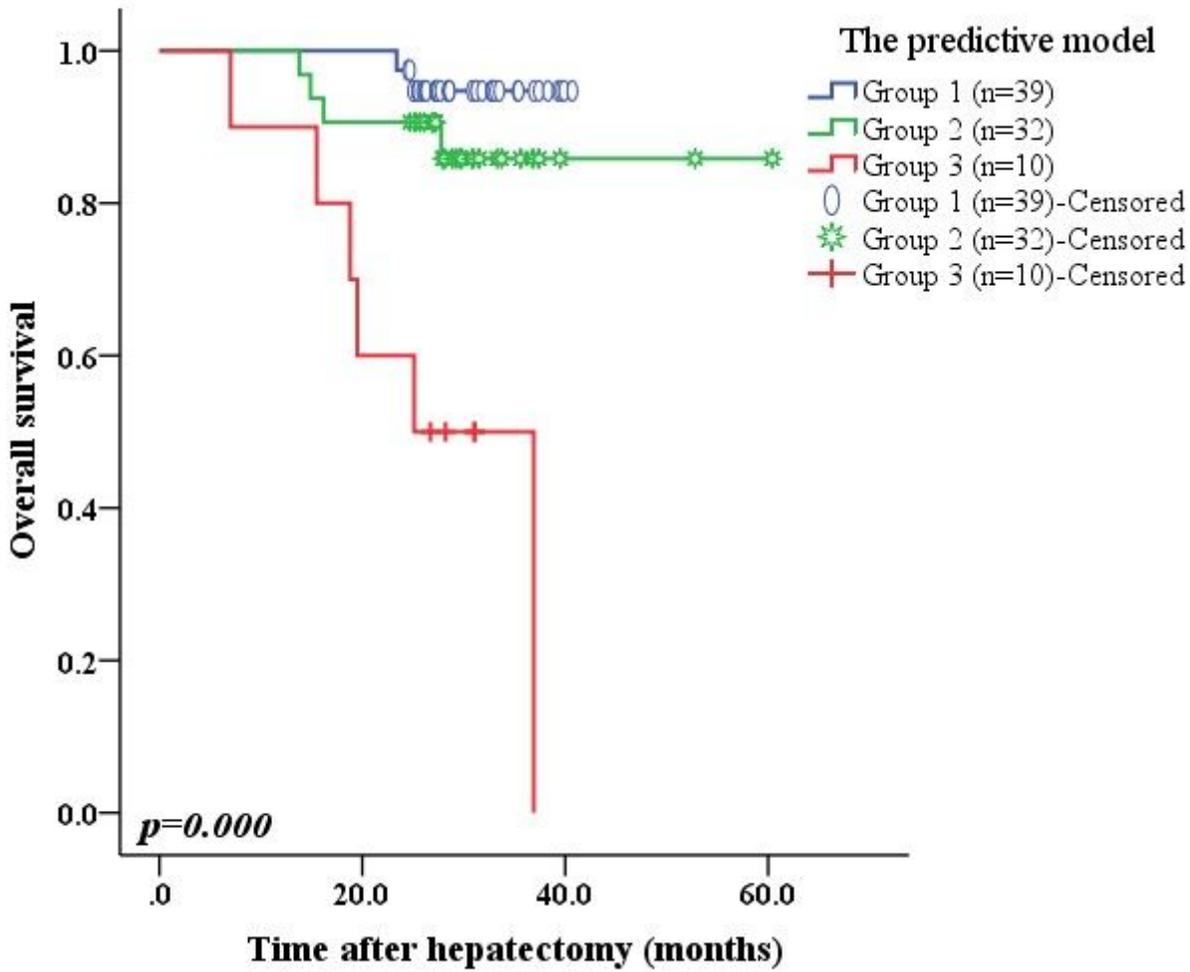


Figure 5

Kaplan-Meier curve showed the difference of OS between three groups was statistically significant ($p=0.000$)