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Original Article

Artificial intelligence based on serum biomarkers predicts the efficacy of lenvatinib for unresectable hepatocellular carcinoma

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Abstract: Lenvatinib has been effective not only as a first-line but also as a later-line systemic therapy for unresectable hepatocellular carcinoma (uHCC) in real-world clinical practice. How to predict the efficacy of lenvatinib and guide appropriate therapy selection in patients with uHCC have become important issues. This study aimed to investigate the impact of serum biomarkers on the treatment outcomes of patients with uHCC treated with lenvatinib in a real-world setting using an artificial intelligence algorithm. We measured serum biomarkers, including alpha-fetoprotein (AFP), albumin-bilirubin (ALBI) grade, and circulating angiogenic factors (CAFs [i.e., vascular endothelial growth factor, angiopoietin-2, fibroblast growth factor-19 [FGF19], and FGF21]) and analyzed treatment outcomes, including objective response rate (ORR), progression-free survival (PFS), and overall survival (OS) in patients with uHCC treated with lenvatinib. The results of this study demonstrated that an AFP reduction ≥ 40% from baseline within 8 weeks after lenvatinib induction was associated with a higher ORR. With baseline biomarkers using a decision tree-based model, we identified patients with high, intermediate, and low ORRs (84.6%, 21.7% and 0%, respectively; odds ratio, 53.04, $P < 0.001$, high versus intermediate/low groups). Based on the decision tree-based survival predictive model, baseline AFP was the most important factor for OS, followed by ALBI grade and FGF21.

Keywords: Efficacy, hepatocellular carcinoma, lenvatinib

Introduction

Hepatocellular carcinoma (HCC) is one of the most common cancers and a leading cause of cancer-related mortality globally. Therapeutic options for HCC are mainly determined by the cancer stage and hepatic functional reserve. For early-stage HCC, curative therapies such as tumor resection, liver transplantation, and local ablation are reasonable therapeutic strategies. Unfortunately, most patients present with unresectable HCC (uHCC) and have a poor prognosis [1]. For patients with uHCC, systemic therapy is currently the mainstream [2-4].

Lenvatinib, an multikinase inhibitor that acts on vascular endothelial growth factor receptors (VEGFR) 1-3, fibroblast growth factor receptors (FGFR) 1-4, platelet-derived growth factor receptor-α (PDGFR-α), KIT, and RET, has demonstrated its promising therapeutic potency for uHCC. The phase III REFLECT trial revealed that patients receiving lenvatinib had a noninferior overall survival (OS) and a better progression-

free survival (PFS) and objective response rate (ORR) than those taking sorafenib [5]. Thus, lenvatinib has become an alternative first-line treatment for patients with uHCC. Subsequent real-world studies also verified the effectiveness and tolerability of lenvatinib in the clinical setting [6].

In Taiwan, lenvatinib has been approved and reimbursed by the National Health Insurance of Taiwan for advanced-stage or intermediate-stage HCC refractory to transarterial chemoembolization. Owing to the rapidly changing landscape of uHCC treatment and the development of new systemic therapy, lenvatinib is now used not only as a first-line but also as a later-line systemic therapy in real-world clinical practice. How to predict the effectiveness of lenvatinib and guide appropriate therapy selection in patients with uHCC have become important issues. Subsequent analyses from the REFLECT study revealed that higher baseline serum biomarkers, including vascular endothelial growth factor (VEGF), angiopoietin-2 (ANG2), and fibroblast growth factor-21 (FGF21), correlated with shorter OS [7]. The current study aimed to investigate the role of serum biomarkers, including alpha-fetoprotein (AFP), albumin-bilirubin (ALBI) score, and circulating angiogenic factors (CAFs [i.e., VEGF, ANG2, FGF19, and FGF21]), on the efficacy of lenvatinib for uHCC by an artificial intelligence algorithm.

Materials and methods

Study population

We retrospectively analyzed patients who had uHCC and received lenvatinib treatment between January 2020 and August 2021 at Kaohsiung Medical University Hospital. The diagnosis of HCC was established based on the criteria of the American Association for the Study of Liver Diseases [8]. Patients with HCC were enrolled if the following criteria were met: 1) uHCC in Barcelona Clinic Liver Cancer (BCLC) stage B or C; 2) receiving at least one dose of lenvatinib; 3) Child-Pugh class A or class B; and 4) Eastern Cooperative Oncology Group performance status score of 0 or 1 at the time of lenvatinib initiation. We excluded patients who received other concomitant systemic therapy, were concurrent with other cancers, or had poor liver function (Child-Pugh class C). Informed consent was obtained from all individual participants included in the study.

Lenvatinib treatment

The standard initial dose of lenvatinib was 12 mg/day for a bodyweight ≥ 60 kg or 8 mg/day for a bodyweight < 60 kg. For patients with Child-Pugh class B, the initial dose of lenvatinib was 8 mg/day based on a previous early-phase clinical trial [9]. However, the initial dose was allowed to be modified at the physician's discretion. Lenvatinib treatment was continued until tumor progression, death, or unacceptable adverse events. We adjusted the dose of lenvatinib and interrupted treatment according to the protocol of the REFLECT study [5].

Assessment

We assessed the baseline characteristics of patients, therapies prior to, during, and after lenvatinib treatment, as well as adverse events (AEs) during lenvatinib administration. We defined OS as the time from the initiation of lenvatinib to death by any cause. PFS was defined as the time between the initiation of lenvatinib and disease progression or death. Time to progression (TTP) was defined as the time between the initiation of treatment with lenvatinib and the date of disease progression. Tumor response was evaluated by dynamic computed tomography or magnetic resonance imaging according to the modified Response Evaluation Criteria in Solid Tumors (mRECIST) and categorized into complete response (CR), partial response (PR), stable disease (SD), and progressive disease (PD). The ORR was defined as the proportion of patients with CR or PR. The disease control rate (DCR) was defined as the proportion of patients with CR, PR, or SD. AEs were assessed according to the National Cancer Institute Common Terminology Criteria for Adverse Events version 5.0.

Serum biomarker analysis

Serum samples were acquired from patients who consented to serum biomarker assessment before the initiation of lenvatinib treatment. Biomarker assays for VEGF, ANG2, FGF19, and FGF21 were performed on serum samples using enzyme-linked immunosorbent assays (ELISAs). Details of the ELISAs used in the current study are provided in the Supplementary Appendix. The association of VEGF, ANG2, FGF19, and FGF21 data with treatment outcomes of patients who had uHCC treated with lenvatinib were analyzed.

Statistical analysis

The continuous variables and categorical variables of different groups were compared using the independent sample t test and the chi-square test or Fisher's exact test, respectively. We used the area under the receiver operating characteristic curve to evaluate the performance of decision tree-based models and FGF21 values in predicting tumor response or mortality. The optimal cutoff value of FGF21 (2.48 log pg/mL) was determined according to the Youden index. We used logistic regression to identify the factors predictive of tumor response by univariate and multivariate models. Survival analyses were conducted by using the Kaplan-Meier method, the log-rank test, and the Cox proportional-hazard model. Variables with a potential association ($P < 0.1$ for PFS and OS analyses; $P < 0.2$ for tumor response analysis) identified in the univariate analysis were included in the multivariate analysis. A two-tailed P value < 0.05 was considered statistically significant. Database processing and analyses were conducted with SPSS version 24.0 (SPSS, Inc., Chicago, IL, USA). We used the "rpart" package of R software to develop a decision tree model with the classification and regression tree (CART) algorithm. In the decision tree analysis, each node was split into two sub-nodes according to the best threshold obtained by the Gini index. This step continued until a stopping rule was satisfied or a maximum homogeneity was reached in the leaf nodes. We used the RandomForest package of R software to perform random forest analysis. The random forest algorithm allowed each decision tree to randomly sample from the dataset by bootstrap. A total of 500 independently grown trees were built, and all trees were integrated into a more accurate result. The importance of predictors of tumor response was ranked by the decrease in the Gini impurity. Out-of-bag error was used to measure the prediction error of the random forest.

Results

Baseline characteristics of 82 patients with uHCC treated with lenvatinib

We enrolled 82 patients who received lenvatinib treatment for uHCC. The baseline characteristics of patients are presented in **Table 1**. The median age was 68.5 (range, 46-93) years, and 56 patients (68.3%) were men. Hepatitis B virus infection was the most prevalent etiology of HCC (n = 35, 42.7%), followed by hepatitis C virus infection (n = 30, 36.6%), and non-B, non-C status (n = 17, 20.7%). Sixty-nine patients (84.1%) were classified as Child-Pugh class A, and 64 patients (78.0%) had BCLC stage C disease. Of the 82 patients, 68 patients (82.9%) and 14 patients (17.1%) received lenvatinib as first-line and second- or later-line systemic therapies, respectively. Twenty patients (24.4%) had a high tumor burden, which was defined as Vp4 invasion (tumor invasion into the main trunk of the portal vein or a portal vein branch contralateral to the primary involved lobe, or both), bile duct invasion, or 50% or higher liver occupation by an intrahepatic tumor, which conformed to the exclusion criteria in the REFLECT study.

Efficacy of lenvatinib

The median lenvatinib treatment duration was 4.1 (range, 0.2-20.5) months. During a mean follow-up duration of 9.0 (range, 1.2-23.2 months) months, the median OS was 12.8 months (95% CI, 10.1-15.5), the median PFS was 5.3 months (95% CI, 3.2-7.4), and the median TTP was 7.5 months (95% CI, 5.6-9.4) (**Table 2** and **Figure 1A**, **1B**). Seventy-four patients (90.2%) received follow-up imaging tests for assessment of tumor response. No patients achieved CR, and 18 patients (24.3%) achieved PR, leading to an ORR of 24.3%. Thirty-six patients (48.6%) had SD, making a DCR of 73.0% (n = 54).

We stratified patients according to a baseline AFP level of 200 ng/mL and ALBI grade. Patients with AFP < 200 ng/mL had markedly better ORR (31.4% vs. 8.7%, P = 0.035), DCR (80.4% vs. 56.5%, P = 0.032), TTP (median, 9.6 months vs. 6.4 months, P = 0.003), PFS (median, 8.9 months vs. 3.0 months, P < 0.001), and OS (median 16.2 months vs. 6.5 months, P < 0.001) than those with AFP ≥ 200 ng/mL (**Table 2** and **Figure 1C**, **1D**). Patients with ALBI grade 1 exhibited a longer OS (median, not reached vs. 9.9 months, P = 0.010) but similar ORR, DCR, TTP, and PFS compared to those with ALBI grade 2 or 3 (**Table 2** and **Figure 1E**, **1F**).

Factors predictive of treatment outcomes on lenvatinib

Factors predictive of tumor response: We evaluated the factors predictive of tumor response

Table 1. Characteristics of the HCC patients

Characteristics	At initiation of Lenvatinib N = 82
Age (years), median (range)	68.5 (46-93)
Male, n (%)	56 (68.3)
Etiology, n (%)	
Hepatitis B	35 (42.7)
Hepatitis C	30 (36.6)
Non-hepatitis B and C	17 (20.7)
Child-Pugh score, n (%)	
A (5)	45 (54.9)
A (6)	24 (29.3)
B	13 (15.8)
ALBI score, n (%)	
Grade 1	32 (39.0)
Grade 2	49 (59.8)
Grade 3	1 (1.2)
BCLC stage, n (%)	
B	18 (22.0)
C	64 (78.0)
Largest tumor size ≥ 5 cm, n (%)	30 (36.6)
HCC ≥ 50% liver occupation	6 (7.3)
Macrovascular invasion, n (%)	29 (35.4)
Vp3	11 (13.4)
Vp4	15 (18.3)
Other	3 (3.7)
Clear bile duct invasion, n (%)	3 (3.7)
High tumor burden	20 (24.4)
Extrahepatic metastasis, n (%)	31 (37.8)
Lung	16 (19.5)
Bone	6 (7.3)
Adrenal gland	5 (6.1)
Others	15 (18.3)
AFP (ng/mL), median (range)	44.7 (1.1-303000.0)
AFP ≥ 200 ng/mL, n (%)	29 (35.4)
AFP ≥ 400 ng/mL, n (%)	21 (25.6)
Prior treatment before lenvatinib, n (%)	73 (89.0)
TACE	63 (76.8)
Local ablation	33 (40.2)
Surgery	27 (32.9)
Systemic therapy	14 (17.1)
Sorafenib	13 (15.9)
Regorafenib	7 (8.5)
Nivolumab	3 (3.7)
Atezolizumab + Bevacizumab	1 (1.2)
Radiation therapy	9 (11.0)
Liver transplantation	1 (1.2)
Concurrent treatment, n (%)	21 (25.6)
TACE	10 (12.2)

(Supplementary Table 1). Baseline AFP < 200 ng/mL, extrahepatic metastasis, and relative dose intensity (RDI) > 70% during the first 8 weeks were potential predictors of lower ORR by univariate analysis. However, none of them remained independent predictors on multivariate analysis. Of the 82 patients, 68 patients possessed available serum samples for CAF analyses at the baseline of lenvatinib treatment. We conducted logistic regression to assess the correlation between CAFs and tumor response. Higher baseline FGF21 and ANG2 had a numerically higher chance of tumor response (OR [95% CI], 2.19 [0.71-6.73] and 2.11 [0.34-13.15], respectively); however, none of them reached statistical significance (**Table 3**).

Of the 45 patients who possessed a baseline AFP ≥ 10 ng/mL and assessable imaging tests, 20 subjects had an AFP response, which was defined as an AFP reduction ≥ 40% from baseline within 8 weeks after lenvatinib administration [10]. Patients with an AFP response demonstrated a remarkably higher ORR than those without an AFP response (40% vs. 8%, P = 0.014, Supplementary Table 2). By using multivariate analysis, AFP response was an independent factor predictive of tumor response (OR, 11.18, P = 0.011, Supplementary Table 3).

Decision tree-based tumor response predictive model: We used CART analysis to make the most feasible model predicting the tumor response to lenvatinib (**Figure 2A**). The decision tree model was constructed based on 12 clinical characteristics (age, sex, etiology of HCC, ALBI grade, BCLC stage, largest

Radiation therapy	8 (9.8)
Others	7 (8.5)
Treatment after lenvatinib, (%)	24 (29.3)
Systemic therapy	12 (14.6)
Atezolizumab + Bevacizumab	4 (4.9)
Sorafenib	5 (6.1)
Nivolumab	4 (4.9)
Regorafenib	1 (1.2)
Cabozantinib	1 (1.2)
Nivolumab + Ipilimumab	1 (1.2)
Pembrolizumab	1 (1.2)
TACE	8 (9.8)
Others	6 (7.3)
Radiation therapy	5 (6.1)
Line of systemic therapy for lenvatinib	
1	68 (82.9)
2	5 (6.1)
3	8 (9.8)
4	1 (1.2)
Duration of therapy (months), median (range)	4.1 (0.2-20.5)
Did not fulfill REFLECT criteria	40 (48.8)

Note: HCC, Hepatocellular Carcinoma; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; Vp3, Tumor Invasion into the First-Order Branches of the Portal Vein; Vp4, Tumor Invasion into the Main Trunk of the Portal Vein or A Contralateral Portal Vein Branch or Both; AFP, Alpha-Fetoprotein; TACE, Transarterial Chemoembolization; High tumor burden, ≥ 50% liver occupation, clear bile duct invasion, or portal vein invasion at the main portal vein.

tumor size ≥ 5 cm, macrovascular invasion, extrahepatic metastasis, AFP < 200 ng/mL, RDI > 70% during the first 8 weeks, concurrent treatment with lenvatinib, and previous systemic therapy) and CAFs (VEGF, ANG2, FGF19, and FGF21). The minimum split size at each node was set at two to avoid overfitting. The best predictor in the root node was the baseline FGF21 level, using < 82 pg/mL versus ≥ 82 ng/mL thresholds for the first step. Baseline FGF21 ≥ 82 pg/mL, AFP < 200 ng/mL, ANG2 ≥ 5411 ng/mL, age < 76 years, VEGF between 76 and 101 pg/mL, FGF19 between 191 and 325 pg/mL, and BCLC stage C were factors associated with a higher ORR. Ten terminal nodes were computed and categorized into three groups: high, intermediate, and low ORR groups. This decision tree-based model had excellent performance with an area under the curve (AUC) up to 0.906 (95% CI, 0.827-0.985; P = 1.7×10^{-6}) (Supplementary Figure 1). The tumor response rate of the high ORR group (84.6%) was significantly higher than that of the intermediate and low ORR groups (21.7% and 0%, respectively) (Supplementary Table 4). High ORR classification remained a strong predictor on multivariate analysis (OR, 53.04, *P* < 0.001, Supplementary Table 5). The relative importance of the tumor response predictors was assessed by the random forest algorithm. Baseline FGF21 was identified as the most important predictor of tumor response, followed by ANG2, AFP, FGF19, and VEGF (**Figure 2B**). The out-of-bag error rate was 30.0% in estimating the true prediction error based on the choices of random forest variables.

Factors predictive of survival outcomes: The factors predictive of PFS and OS based on univariate and multivariate analyses are shown in Supplementary Tables 6 and 7 and summarized in **Table 4**. Baseline AFP level < 200 ng/mL (HR, 0.47; P = 0.008), extrahepatic metastasis (HR, 2.03; P = 0.010), largest tumor size ≥ 5 cm (HR, 1.96; P = 0.026), and RDI > 70% during the first 8 weeks (HR, 0.50; P = 0.027) were independent predictors of PFS. Baseline AFP level < 200 ng/mL (HR, 0.33; P = 0.002), ALBI grade 2 or 3 (vs. grade 1; HR, 3.83; P = 0.002), largest tumor size ≥ 5 cm (HR, 3.14; P = 0.003), and RDI > 70% during the first 8 weeks (HR, 0.30; P = 0.003) were independent predictors of OS.

We analyzed the correlation between CAFs and survival outcomes (**Table 3**). For PFS, increased CAFs including VEGF, ANG2, FGF19, and FGF21, had numerically higher hazard rates; however, none of them reached statistical significance. For OS, increased VEGF, ANG2, FGF19, and FGF21 also showed numerically higher hazard rates, but only FGF21 had a significant association with worse OS (HR, 2.04, P = 0.017). The optimal cutoff value of FGF21 (2.48 log pg/mL) to predict mortality was obtained based on the Youden index. By multivariate analysis, FGF21 ≥ 2.48 log pg/mL was predictive of worse OS (HR, 2.30, P = 0.028) (Supplementary Table 8). Patients who had FGF21 < 2.48

Table 2. Treatment efficacy of lenvatinib

Variable	All patients n = 82	AFP < 200 ng/mL n = 53	AFP ≥ 200 ng/mL n = 29	*p* value	ALBI Gr.1 n = 32	ALBI Gr.2/3 n = 50	*p* value
Response by mRECIST, n (%)†							
Complete response	0 (0)	0 (0)	0 (0)		0 (0)	0 (0)	
Partial response	18 (24.3)	16 (31.4)	2 (8.7)		9 (29.0)	9 (20.9)	
Stable disease	36 (48.6)	25 (49.0)	11 (47.8)		14 (45.2)	22 (51.2)	
Progressive disease	20 (27.0)	10 (19.6)	10 (43.5)		8 (25.8)	12 (27.9)	
Objective response rate, n (%)	18 (24.3)	16 (31.4)	2 (8.7)	0.035	9 (29.0)	9 (20.9)	0.423
Disease control rate, n (%)	54 (73.0)	41 (80.4)	13 (56.5)	0.032	23 (74.2)	31 (72.1)	0.841
Time to progression, median, months (95% CI)†	7.5 (5.6-9.4)	9.6 (5.3-13.9)	6.4 (2.0-10.8)	0.003	6.9 (4.5-9.2)	7.5 (3.3-11.7)	0.428
Progression-free survival, median, months (95% CI)	5.3 (3.2-7.4)	8.9 (4.0-13.8)	3.0 (2.7-3.3)	< 0.001	6.7 (5.3-8.2)	4.7 (3.9-5.5)	0.108
Overall survival, median, months (95% CI)	12.8 (10.1-15.5)	16.2 (12.0-20.3)	6.5 (3.3-9.6)	< 0.001	NE	9.9 (3.6-16.3)	0.010

Note: mRECIST, modified Response Evaluation Criteria in Solid Tumors; 95% CI, 95% Confidence Interval; AFP, Alpha-Fetoprotein; ALBI grade, Albumin-Bilirubin grade; NE, Not Estimable. †Available imaging tests for response assessment in 74 patients: 51 patients had baseline AFP < 200 ng/mL, and 23 patients had baseline AFP ≥ 200 ng/mL; 31 patients had ALBI grade 1, and 43 patients had ALBI grade 2 or 3.

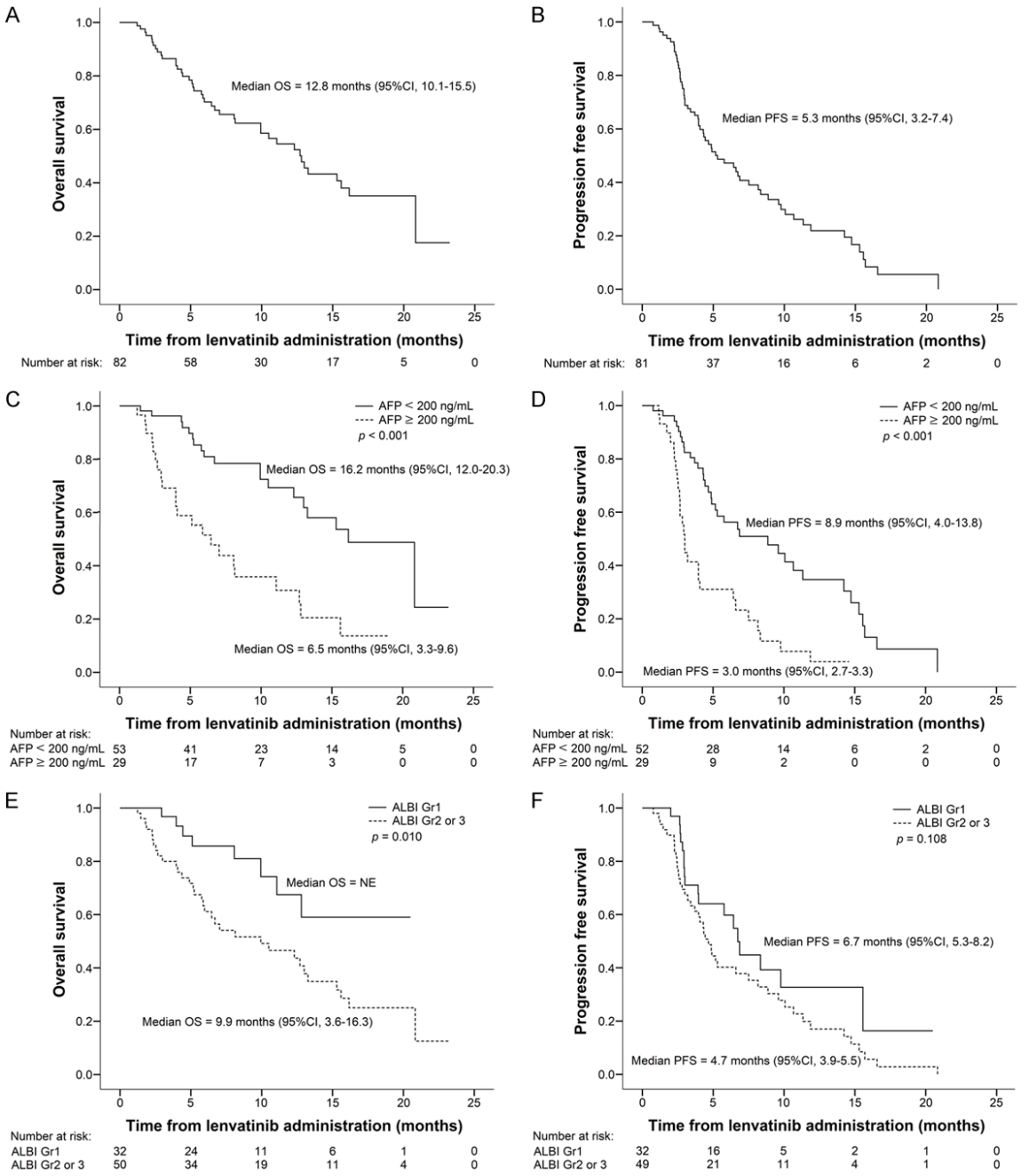

Figure 1. Survival outcomes in patients who received lenvatinib therapy. (A) Overall survival and (B) progression-free survival. (C) Overall survival and (D) progression-free survival based on baseline AFP; (E) overall survival and (F) progression-free survival based on ALBI grade. AFP, Alpha-Fetoprotein; ALBI, Albumin-Bilirubin.

log pg/mL demonstrated a significantly better OS than those with FGF21 ≥ 2.48 log pg/mL (median, 15.3 vs. 8.1 months, P = 0.006, Supplementary Figure 2).

Decision tree-based survival predictive model: The decision tree model was constructed based on 13 clinical characteristics (age, sex, etiology of HCC, ALBI score, BCLC stage, largest tumor size ≥ 5 cm, macrovascular invasion, extrahepatic metastasis, AFP < 200 ng/mL, RDI > 70% during the first 8 weeks, concurrent treatment with lenvatinib, previous systemic therapy, and treatment after lenvatinib) and four CAFs. **Figure 3A** demonstrates that the best predictor in the root node was the base-

Table 3. Association of circulating angiogenic factors with treatment outcomes on lenvatinib (n = 68)

Markers	OR (95% CI)	*p* value
Objective response rate†		
VEGF (log pg/mL)	1.07 (0.21-5.44)	0.932
ANG2 (log pg/mL)	2.11 (0.34-13.15)	0.424
FGF19 (log pg/mL)	0.68 (0.18-2.65)	0.580
FGF21 (log pg/mL)	2.19 (0.71-6.73)	0.171
	HR (95% CI)	*p* value
Progression-free survival		
VEGF (log pg/mL)	1.83 (0.71-4.73)	0.212
ANG2 (log pg/mL)	1.59 (0.71-3.57)	0.265
FGF19 (log pg/mL)	1.79 (0.90-3.56)	0.100
FGF21 (log pg/mL)‡	1.46 (0.89-2.39)	0.138
Overall survival		
VEGF (log pg/mL)	1.97 (0.74-5.27)	0.178
ANG2 (log pg/mL)	2.27 (0.82-6.30)	0.116
FGF19 (log pg/mL)	1.66 (0.75-3.69)	0.215
FGF21 (log pg/mL)‡	2.04 (1.14-3.65)	0.017

VEGF, Vascular Endothelial Growth Factor; ANG2, Angiopoietin-2; FGF, Fibroblast Growth Factor; HR, Hazard Ratio; OR, Odds Ratio; 95% CI, 95% Confidence Interval. †Available imaging tests for response assessment in 62 patients. ‡n = 66.

line AFP levels, using < 200 ng/mL versus ≥ 200 ng/mL thresholds for the first step. Seven terminal nodes were computed and categorized into three groups: high, intermediate, and low OS groups. The OS of different groups are presented in **Figure 3B**. This decision tree-based model had excellent performance with an AUC of 0.873 (95% CI, 0.793-0.953; P = 1.8 × 10^{-8}) (Supplementary Figure 3).

Discussion

The landscape of systemic therapy for uHCC has dramatically changed over the past decade with the blooming of new drugs. To date, sorafenib, lenvatinib, and atezolizumab plus bevacizumab have been approved as first-line treatments for advanced HCC [5, 11, 12]. Recent real-world data from China and Japan also demonstrated that lenvatinib was effective in patients who experienced prior systemic therapy [13, 14]. Thus, lenvatinib is also used as a second- or later-line of systemic therapy in the clinical setting. In the current study, the median OS, PFS, and TTP of lenvatinib were 12.8 months, 5.3 months, and 7.5 months, respectively, which were similar to the results of the REFLECT study and previous real-world reports [5, 6]. For the best overall response, ORR (24.3%) and DCR (73.0%) observed in our study were also similar to those in the REFLECT trial (ORR: 24.1%, DCR: 75.5%).

We found that patients with baseline AFP < 200 ng/mL had a markedly higher ORR than their counterparts (31.4% vs. 8.7%). However, AFP < 200 ng/mL was no longer predictive of tumor response after adjusting for other covariates, such as extrahepatic metastasis and RDI. A reduction in AFP ≥ 40% from baseline was found to be an early predictor for tumor response in our study, which was consistent with one previous report [10]. Nonetheless, compared to predicting tumor response during lenvatinib treatment, it might be more valuable to predict tumor response before drug induction and select proper candidates who potentially benefit more from lenvatinib, especially for patients who have the potential to receive subsequent curative therapy. By the decision tree model, we stratified patients into subgroups with different chances of tumor response. This tumor response classification remains a strong predictor for objective response on multivariate analysis. By assessing the cut-off values of serum biomarkers used to split nodes in the decision tree, we observed that higher baseline FGF21 and ANG2 levels were factors related to a higher ORR, while higher baseline VEGF and FGF19 levels were not always predictive of a higher incidence of tumor response. The current findings may explain the longer OS observed in patients who had high baseline levels of FGF21 and ANG2 compared with sorafenib from the REFLECT study, while this phenomenon was not observed with FGF19 [7]. In another HCC study regarding lenvatinib treatment, baseline FGF19 was not a predictor for objective response, while increases in FGF19 and decreases in ANG2 were associated with lenvatinib responses [15].

The decision tree-based survival predictive model demonstrated that baseline AFP levels, ALBI grade, and FGF21 were the three most important serum biomarkers associated with OS. Baseline AFP has been observed as a prognostic factor for patients who underwent sorafenib or regorafenib therapy [16, 17]. However, previous data regarding the correlation between baseline AFP and survival with lenva-

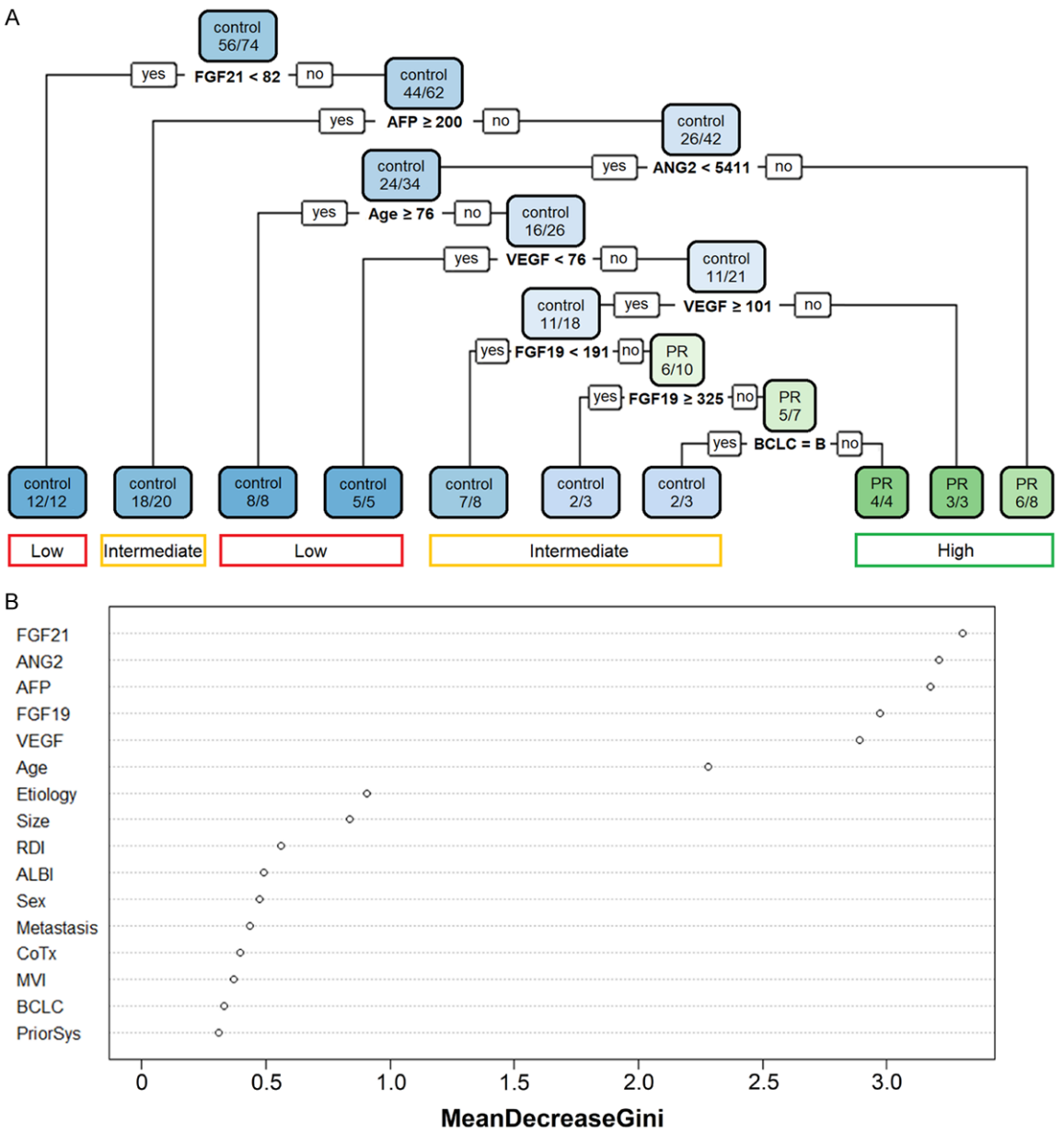

Figure 2. A. Decision tree algorithm to predict tumor response and categorize patients into three groups: high, intermediate, and low ORR groups. In each node, the number in the denominator represents the total number of patients, and the number in the numerator represents the number of peoples with partial response (PR) or control (i.e., stable disease or progressive disease). B. The importance of factors predictive of tumor response determined by the random forest model. RDI, RDI > 70% during the first 8 weeks; ALBI, Albumin-Bilirubin grade; CoTx, Concurrent Treatment with Lenvatinib; MVI, Macrovascular Invasion; PriorSys, Previous Systemic Therapy.

tinib are limited, and only one real-world study from Taiwan described AFP ≥ 200 ng/mL as a risk factor associated with mortality [18].

ALBI grade, which reflects hepatic functional reserve, plays an important role in the prediction of treatment outcomes in patients with HCC undergoing anticancer treatment [19-21]. In our study, baseline ALBI grade was associated with OS but not with PFS and ORR on lenvatinib, which indicates that baseline liver function can strongly predict patient survival, regardless of its limited impact on tumor response and disease control. The inferior OS in patients with ALBI grade 2 or 3 might be ascribed to the higher incidence of liver function worsening during lenvatinib treatment, leading to a higher treatment discontinuation

Table 4. Factors predictive of survival outcomes on lenvatinib

	Univariate HR (95% CI)	*p* value	Multivariate HR (95% CI)	*p* value
Progression-free survival				
AFP < 200 ng/mL	0.36 (0.21-0.61)	< 0.001	0.47 (0.27-0.82)	0.008
Extrahepatic metastasis	2.35 (1.38-4.02)	0.002	2.03 (1.18-3.50)	0.010
Largest tumor size ≥ 5 cm	1.65 (0.99-2.76)	0.055	1.96 (1.09-3.53)	0.026
RDI > 70% during the first 8 weeks	0.61 (0.36-1.04)	0.067	0.50 (0.27-0.92)	0.027
Overall survival				
AFP < 200 ng/mL	0.31 (0.16-0.57)	< 0.001	0.33 (0.16-0.66)	0.002
ALBI grade 2 or 3 (vs. grade 1)	2.66 (1.22-5.77)	0.014	3.83 (1.66-8.86)	0.002
Largest tumor size ≥ 5 cm	2.39 (1.28-4.44)	0.006	3.14 (1.49-6.62)	0.003
RDI > 70% during the first 8 weeks	0.44 (0.23-0.86)	0.016	0.30 (0.14-0.65)	0.003

Note: ALBI grade, Albumin-Bilirubin grade; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; HR, Hazard Ratio; OR, Odds Ratio; 95% CI, 95% Confidence Interval.

rate (Supplementary Table 9). Poorer baseline liver function status has been related to higher dose reduction and treatment interruption or discontinuation due to AEs [22].

Despite the potential role in predicting tumor response with lenvatinib, higher levels of baseline FGF21 were observed to be associated with worse OS in the current study. We used the Youden index to acquire the optimal cutoff value of FGF21 (2.48 log pg/mL) to predict mortality. FGF21 < 2.48 log pg/mL was an independent factor for OS in univariate and multivariate analyses. One recent prospective cohort study reported that higher serum FGF21 was predictive of worse survival in patients with HCC [23], which suggested that FGF21 may be a metabolism-related prognostic biomarker not only for patients receiving lenvatinib treatment but also for whole cohort of patient with HCC. Taken together, even though high FGF21 was related to poor survival, its impact on tumor shrinkage during lenvatinib therapy might diminish the adverse association. Considering that no significant changes in FGF21 levels from baseline were observed in the patients from the REFLECT trial [7], the underlying mechanism by which FGF21 influences tumor response and survival with lenvatinib remains unclear. More investigations are warranted to elucidate the contradictory roles of FGF21 levels in predicting the ORR and OS observed in the current study.

Our study had some limitations. First, it was a retrospective study, which may have led to unintended biases in patient selection and the evaluation of AEs and treatment outcomes. Second, the number of patients in this study was relatively small, which may limit the interpretation of the heterogeneous and complex interactions of serum biomarkers in HCC. However, we used an artificial intelligence algorithm to overcome this challenge. The random forest algorithm provided internal validation and avoided overfitting. Finally, we did not analyze the dynamic change in ALBI grade and CAFs. Previous reports have demonstrated that changes in the ALBI score and increases in FGF19 levels after lenvatinib administration might be related to the tumor response rate [7, 10]; however, their roles in predicting survival outcomes with lenvatinib are still unknown. Further studies are needed in the future.

Conclusions

Baseline CAFs and early AFP decline were associated with a higher ORR, while baseline levels of FGF21, AFP and ALBI grade were factors predictive of longer OS with lenvatinib by decision tree-based models. The current study successfully deployed an artificial intelligence-based model with noninvasive serum biomarkers to predict treatment responses and long-term outcomes among patients with uHCC treated with lenvatinib.

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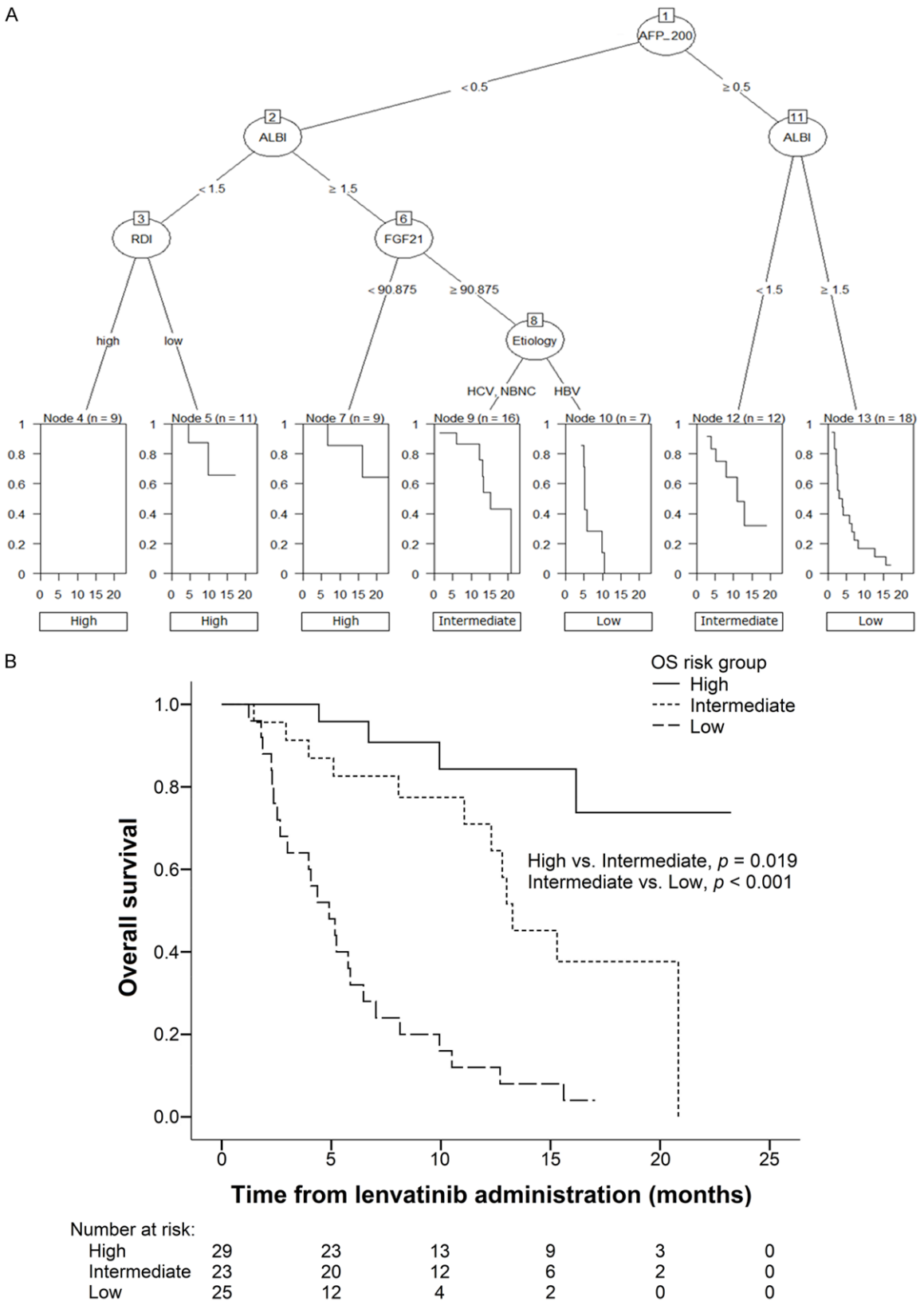

Figure 3. A. Decision tree algorithm to predict the overall survival (OS) and categorize patients into three groups: high, intermediate, and low OS groups. B. Kaplan-Meier survival analysis based on OS groups.

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Informed consent was obtained from all individual participants included in the study.

Disclosure of conflict of interest

Ming-Lung Yu has served as a speaker for AbbVie, Abbott, Bristol-Myers Squibb, Gilead, Merck, a consultant for AbbVie, Abbott, Bristol-Myers Squibb, Gilead, Merck and PharmaEssentia, and has received research funding from AbbVie, Abbott, Bristol-Myers Squibb, Gilead, and Merck. Chung-Feng Huang has served as a speaker for AbbVie, Bristol-Myers Squibb, Gilead, Merck, and Roche. Jee-Fu Huang has served as a speaker for AbbVie, Bristol-Myers Squibb, Gilead, Merck, Sysmex, Roche, a consultant for Roche, Bristol-Myers Squibb, Gilead, Merck, Sysmex, PharmaEssentia, and Polaris Pharmaceuticals.

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Supplementary appendix

The following ELISAs were used for serum biomarker assays and all from R&D systems, Minneapolis, MN, USA:

Human VEGF Quantikine ELISA Kit (DVE00) for VEGF

Human Angiopoietin-2 Quantikine ELISA Kit (DANG20) for ANG2

Human FGF-19 Quantikine ELISA Kit (DF1900) for FGF19

Human FGF-21 Quantikine ELISA Kit (DF2100) for FGF21

Supplementary Table 1. Factors associated with tumor response on lenvatinib

	Univariate		Multivariate	
	OR (95% CI)	*p* value	OR (95% CI)	*p* value
Age > 68 years	1.25 (0.43-3.63)	0.682		
Male	1.13 (0.35-3.68)	0.835		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	0.62 (0.16-2.37)	0.481		
Hepatitis C	0.66 (0.16-2.67)	0.560		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	0.65 (0.22-1.88)	0.424		
BCLC stage				
Stage B		Reference		
Stage C	1.06 (0.30-3.78)	0.931		
Largest tumor size ≥ 5 cm	1.59 (0.52-4.83)	0.413		
Macrovascular invasion	1.34 (0.45-4.04)	0.599		
Extrahepatic metastasis	0.44 (0.13-1.52)	0.194	0.47 (0.13-1.69)	0.246
AFP < 200 ng/mL	4.80 (1.01-22.99)	0.049	4.47 (0.92-21.83)	0.064
RDI > 70% during the first 8 weeks	2.25 (0.77-6.62)	0.141	2.08 (0.68-6.41)	0.201
Concurrent treatment with lenvatinib	1.05 (0.32-3.45)	0.934		
Previous systemic therapy history	0.51 (0.10-2.56)	0.415		

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; OR, Odds Ratio; 95% CI, 95% Confidence Interval.

Supplementary Table 2. Treatment outcomes with lenvatinib according to AFP response in patients with baseline AFP ≥ 10 ng/mL

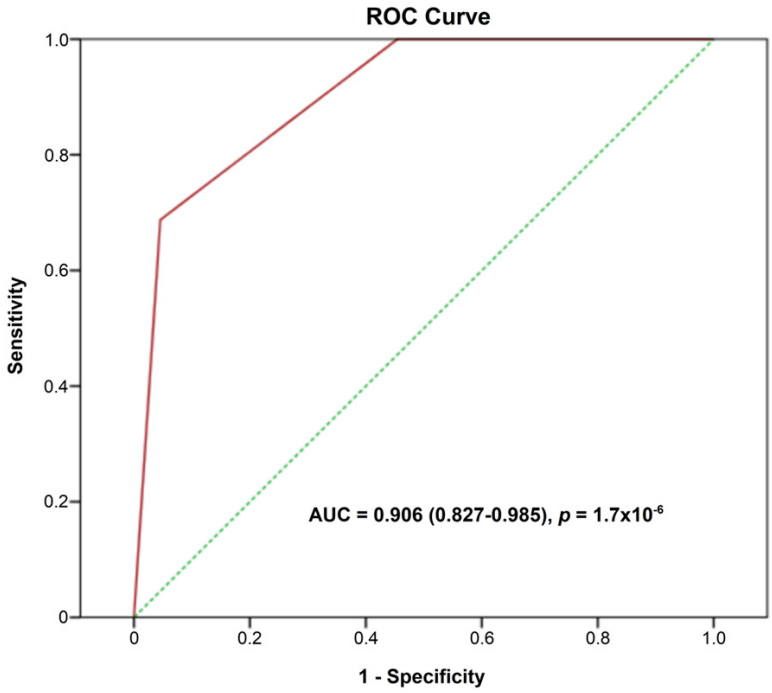
Variable	All patients n = 51	AFP responder n = 23	AFP non-responder n = 28	*p* value
Response by mRECIST, n (%)†				
Complete response	0 (0)	0 (0)	0 (0)	
Partial response	10 (22.2)	8 (40.0)	2 (8.0)	
Stable disease	22 (48.9)	7 (35.0)	15 (60.0)	
Progressive disease	13 (28.9)	5 (25.0)	8 (32.0)	
Objective response rate, n (%)	10 (22.2)	8 (40.0)	2 (8.0)	0.014
Disease control rate, n (%)	32 (71.1)	15 (75.0)	17 (68.0)	0.607
Time to progression, median, months (95% CI)†	6.9 (4.4-9.3)	8.2 (5.8-10.6)	5.3 (2.0-8.6)	0.997
Progression-free survival, median, months (95% CI)	4.4 (3.1-5.8)	6.4 (3.5-9.3)	4.3 (3.0-5.6)	0.885
Overall survival, median, months (95% CI)	9.9 (6.4-13.5)	9.9 (4.5-15.4)	9.9 (3.4-16.4)	0.467

Note: mRECIST, modified Response Evaluation Criteria In Solid Tumors; AFP responder was defined as the patients with an AFP reduction ≥ 40% from baseline within 8 weeks after lenvatinib induction; 95% CI, 95% Confidence Interval; AFP, Alpha-Fetoprotein; ALBI grade, Albumin-Bilirubin grade. †Available imaging tests for response assessment in 45 patients: 20 AFP responders and 25 AFP non-responder.

Supplementary Table 3. Factors associated with tumor response on lenvatinib in patients with baseline AFP ≥ 10 ng/mL, n = 45

	Univariate		Multivariate	
	OR (95% CI)	*p* value	OR (95% CI)	*p* value
Age > 68 years	0.64 (0.14-2.92)	0.567		
Male	0.44 (0.10-1.97)	0.286		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	3.00 (0.30-29.94)	0.349		
Hepatitis C	3.27 (0.31-34.72)	0.325		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	0.67 (0.16-2.74)	0.574		
BCLC stage				
Stage B		Reference		
Stage C	1.86 (0.20-17.58)	0.587		
Largest tumor size ≥ 5 cm	2.18 (0.52-9.12)	0.285		
Macrovascular invasion	1.13 (0.27-4.76)	0.869		
Extrahepatic metastasis	0.51 (0.11-2.30)	0.380		
AFP < 200 ng/mL	4.75 (0.88-25.64)	0.070	7.52 (1.15-49.27)	0.035
AFP response	7.67 (1.40-41.94)	0.019	11.18 (1.75-71.29)	0.011
RDI > 70% during the first 8 weeks	1.46 (0.34-6.22)	0.613		
Concurrent treatment with lenvatinib	1.24 (0.26-5.84)	0.787		
Previous systemic therapy history	0.44 (0.05-4.12)	0.475		

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; AFP response was defined as an AFP reduction ≥ 40% from baseline within 8 weeks after lenvatinib induction; RDI, Relative Dose Intensity; OR, Odds Ratio; 95% CI, 95% Confidence Interval.



Supplementary Figure 1. Receiver operating characteristic curve analysis based on tumor response classification: high (score = 3), intermediate (score = 2), and low ORR groups (score = 1).

Supplementary Table 4. Best overall response with lenvatinib according to tumor response classification

	Total n = 60	Low n = 24	Intermediate n = 23	High n = 13	*p* value
Response by mRECIST, n (%)					
Complete response	0 (0)	0 (0)	0 (0)	0 (0)	
Partial response	16 (26.7)	0 (0)	5 (21.7)	11 (84.6)	
Stable disease	30 (50.0)	16 (66.7)	12 (52.2)	2 (15.4)	
Progressive disease	14 (23.3)	8 (33.3)	6 (26.1)	0 (0)	
Objective response rate, n (%)†	16 (26.7)	0 (0)	5 (21.7)	11 (84.6)	< 0.001
Disease control rate, n (%)	46 (76.7)	16 (66.7)	17 (73.9)	13 (100.0)	0.067

Note: †The adjusted standardized residual was greater than 2 which indicates the column proportions were significantly different at P < 0.05 level.

Supplementary Table 5. Factors including tumor response classifications predictive of tumor response with lenvatinib, n = 60

	Univariate		Multivariate	
	OR (95% CI)	*p* value	OR (95% CI)	*p* value
Age > 68 years	1.17 (0.37-3.71)	0.785		
Male	1.03 (0.30-3.52)	0.967		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	0.44 (0.10-2.00)	0.286		
Hepatitis C	0.54 (0.11-2.72)	0.452		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	0.81 (0.25-2.58)	0.721		
BCLC stage				
Stage B		Reference		
Stage C	0.88 (0.23-3.35)	0.854		
Largest tumor size ≥ 5 cm	2.33 (0.70-7.75)	0.167	0.86 (0.13-5.72)	0.876
Macrovascular invasion	0.97 (0.28-3.34)	0.967		
Extrahepatic metastasis	0.58 (0.16-2.11)	0.412		
AFP < 200 ng/mL	3.62 (0.73-18.07)	0.117	0.79 (0.11-5.90)	0.817
Tumor response classification				
Low and intermediate		Reference		
High	46.20 (7.88-270.99)	< 0.001	53.04 (6.35-442.82)	< 0.001
RDI > 70% during the first 8 weeks	1.86 (0.58-5.90)	0.294		
Concurrent treatment with lenvatinib	0.89 (0.24-3.30)	0.860		
Previous systemic therapy history	0.56 (0.11-2.90)	0.486		

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; OR, Odds Ratio; 95% CI, 95% Confidence Interval.

Supplementary Table 6. Factors predictive of progression free survival (PFS) with lenvatinib

	Univariate		Multivariate	
	HR (95% CI)	*p* value	HR (95% CI)	*p* value
Age > 68 years	0.77 (0.46-1.28)	0.307		
Male	0.72 (0.43-1.22)	0.219		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	1.30 (0.64-2.64)	0.467		
Hepatitis C	1.30 (0.64-2.67)	0.470		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	1.57 (0.90-2.72)	0.112		
BCLC stage				
Stage B		Reference		
Stage C	1.41 (0.73-2.71)	0.306		
Largest tumor size ≥ 5 cm	1.65 (0.99-2.76)	0.055	1.96 (1.09-3.53)	0.026
Macrovascular invasion	1.01 (0.60-1.72)	0.969		
Extrahepatic metastasis	2.35 (1.38-4.02)	0.002	2.03 (1.18-3.50)	0.010
AFP < 200 ng/mL	0.36 (0.21-0.61)	< 0.001	0.47 (0.27-0.82)	0.008
RDI > 70% during the first 8 weeks	0.61 (0.36-1.04)	0.067	0.50 (0.27-0.92)	0.027
Concurrent treatment with lenvatinib	1.12 (0.64-1.95)	0.692		
Previous systemic therapy history	1.19 (0.64-2.21)	0.582		

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; HR, Hazard Ratio; 95% CI, 95% Confidence Interval.

Supplementary Table 7. Factors predictive of overall survival (OS) with lenvatinib

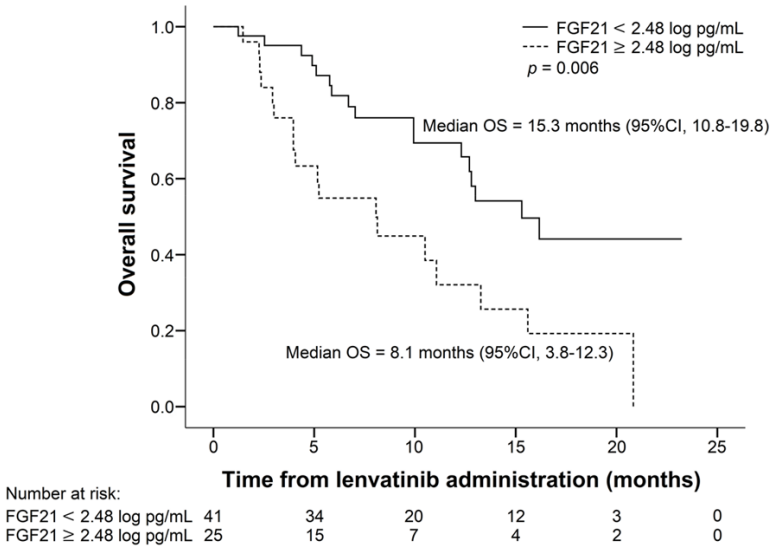
	Univariate		Multivariate	
	HR (95% CI)	*p* value	HR (95% CI)	*p* value
Age > 68 years	0.88 (0.47-1.63)	0.673		
Male	0.87 (0.46-1.64)	0.661		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	1.97 (0.79-4.92)	0.148		
Hepatitis C	1.70 (0.65-4.42)	0.280		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	2.66 (1.22-5.77)	0.014	3.83 (1.66-8.86)	0.002
BCLC stage				
Stage B		Reference		
Stage C	2.68 (1.04-6.87)	0.040	0.80 (0.22-2.93)	0.734
Largest tumor size ≥ 5 cm	2.39 (1.28-4.44)	0.006	3.14 (1.49-6.62)	0.003
Macrovascular invasion	1.85 (0.99-3.45)	0.053	1.66 (0.68-4.06)	0.264
Extrahepatic metastasis	1.71 (0.92-3.19)	0.091	1.32 (0.55-3.15)	0.538
AFP < 200 ng/mL	0.31 (0.16-0.57)	< 0.001	0.33 (0.16-0.66)	0.002
RDI > 70% during the first 8 weeks	0.44 (0.23-0.86)	0.016	0.30 (0.14-0.65)	0.003
Concurrent treatment with lenvatinib	0.93 (0.46-1.87)	0.840		
Previous systemic therapy history	0.49 (0.20-1.19)	0.114		
Treatment after lenvatinib	0.73 (0.37-1.43)	0.357		

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; HR, Hazard Ratio; 95% CI, 95% Confidence Interval.

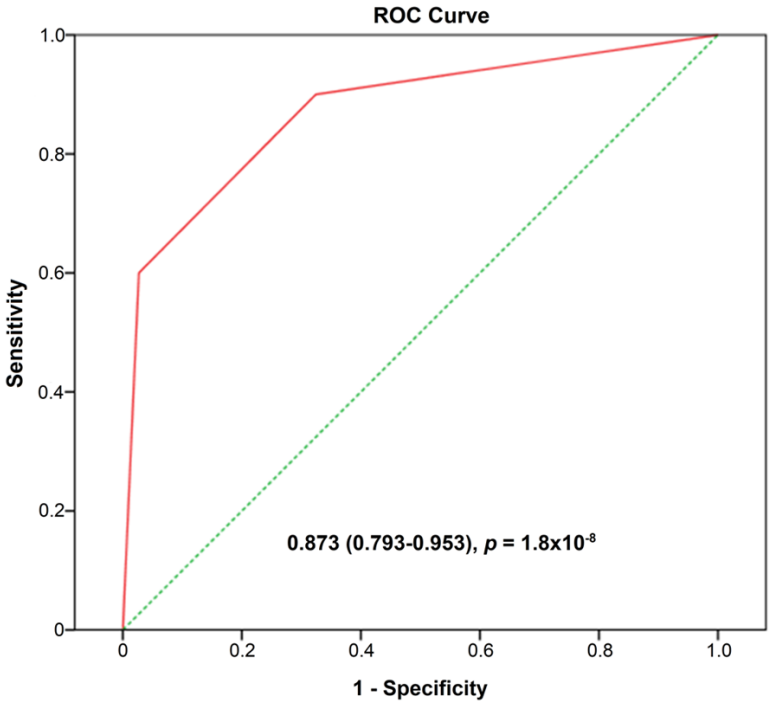
Supplementary Table 8. Factors predictive of overall survival (OS) with lenvatinib in patients who had available FGF21 data, n = 66

	Univariate		Multivariate	
	HR (95% CI)	*p* value	HR (95% CI)	*p* value
Age > 68 years	0.81 (0.41-1.58)	0.530		
Male	1.01 (0.50-2.05)	0.969		
Etiology				
Non-hepatitis B and C		Reference		
Hepatitis B	1.62 (0.60-4.37)	0.345		
Hepatitis C	1.27 (0.44-3.73)	0.658		
ALBI score				
Grade 1		Reference		
Grade 2 or 3	2.24 (0.97-5.15)	0.058	4.06 (1.60-10.31)	0.003
BCLC stage				
Stage B		Reference		
Stage C	3.13 (1.09-8.93)	0.033	0.69 (0.21-2.25)	0.540
Largest tumor size ≥ 5 cm	1.81 (0.92-3.56)	0.085	2.25 (0.88-5.74)	0.091
Macrovascular invasion	1.75 (0.88-3.46)	0.108		
Extrahepatic metastasis	1.85 (0.94-3.65)	0.077	2.17 (0.97-4.86)	0.059
AFP < 200 ng/mL	0.29 (0.14-0.57)	< 0.001	0.30 (0.13-0.68)	0.004
RDI > 70% during the first 2 weeks	0.38 (0.19-0.77)	0.007	0.33 (0.13-0.83)	0.019
Concurrent treatment with lenvatinib	0.99 (0.47-2.07)	0.970		
Previous systemic therapy history	0.31 (0.11-0.89)	0.030	0.14 (0.04-0.48)	0.002
Treatment after lenvatinib	0.73 (0.35-1.52)	0.393		
FGF21 ≥ 2.48 log pg/mL	2.47 (1.27-4.83)	0.008	2.30 (1.09-4.82)	0.028

Note: NBNC, Non-HBV and Non-HCV; HBV, Hepatitis B Virus; HCV, Hepatitis C Virus; ALBI score, Albumin-Bilirubin score; BCLC stage, Barcelona Clinic Liver Cancer stage; AFP, Alpha-Fetoprotein; RDI, Relative Dose Intensity; HR, Hazard Ratio; 95% CI, 95% Confidence Interval.



Supplementary Figure 2. Overall survival based on baseline FGF21 (FGF21 < 2.48 log pg/mL vs. ≥ 2.48 log pg/mL). FGF21, Fibroblast Growth Factor-21.



Supplementary Figure 3. Receiver operating characteristic curve analysis based on overall survival classification.

Supplementary Table 9. Adverse events during lenvatinib therapy

AEs, n (%)	All patients n = 82		AFP < 200 ng/mL n = 53		AFP ≥ 200 ng/mL n = 29		ALBI Gr.1 n = 32		ALBI Gr.2 or 3 n = 50	
	Any Gr.	Gr. ≥ 3	Any Gr.	Gr. ≥ 3	Any Gr.	Gr. ≥ 3	Any Gr.	Gr. ≥ 3	Any Gr.	Gr. ≥ 3
All AEs	81 (98.8)	30 (36.6)	52 (98.1)	21 (39.6)	29 (100.0)	9 (31.0)	32 (100.0)	11 (34.4)	49 (98.0)	21 (42.0)
Fatigue	50 (61.0)	7 (8.5)	34 (64.2)	4 (7.5)	16 (55.2)	3 (10.3)	19 (59.4)	3 (9.4)	31 (62.0)	4 (8.0)
Proteinuria	35 (42.7)	4 (4.9)	22 (41.5)	3 (5.7)	13 (44.8)	1 (3.4)	15 (46.9)	1 (3.1)	20 (40.0)	3 (6.0)
Blood bilirubin elevation	30 (36.6)	12 (14.6)	18 (34.0)	7 (13.2)	12 (41.4)	5 (17.2)	5 (15.6)	3 (9.4)	25 (50.0)	9 (18.0)
Hypothyroidism	28 (34.1)	1 (1.2)	18 (34.0)	1 (1.9)	10 (34.5)	0 (0)	15 (46.9)	0 (0)	13 (26.0)	1 (2.0)
Hand-foot skin reaction	24 (29.3)	3 (3.7)	16 (30.2)	3 (5.7)	8 (27.6)	0 (0)	14 (43.8)	2 (6.3)	10 (20.0)	1 (2.0)
Thrombocytopenia	22 (26.8)	6 (7.3)	14 (26.4)	4 (7.5)	8 (27.6)	2 (6.9)	11 (34.4)	2 (6.3)	11 (22.0)	4 (8.0)
Aminotransferase elevation	19 (23.2)	2 (2.4)	9 (17.0)	1 (1.9)	10 (34.5)	1 (3.4)	5 (15.6)	1 (3.1)	14 (28.0)	1 (2.0)
Hypertension	18 (22.0)	0 (0.0)	12 (22.6)	0 (0)	6 (20.7)	0 (0)	8 (25.0)	0 (0)	10 (20.0)	0 (0)
Diarrhea	17 (20.7)	2 (2.4)	11 (20.8)	1 (1.9)	6 (20.7)	1 (3.4)	9 (28.1)	2 (6.3)	8 (16.0)	0 (0)
Anemia	12 (14.6)	2 (2.4)	9 (17.0)	1 (1.9)	3 (10.3)	1 (3.4)	2 (6.3)	1 (3.1)	10 (20.0)	1 (2.0)
Leukopenia	8 (9.8)	1 (1.2)	4 (7.5)	1 (1.9)	4 (13.8)	0 (0)	5 (15.6)	1 (3.1)	3 (6.0)	0 (0)
Anorexia	5 (6.1)	0 (0)	4 (7.5)	0 (0)	1 (3.4)	0 (0)	1 (3.1)	0 (0)	4 (8.0)	0 (0)
Skin rash	4 (4.9)	1 (1.2)	2 (3.8)	1 (1.9)	2 (6.9)	0 (0)	1 (3.1)	0 (0)	3 (6.0)	1 (2.0)
Nausea/Vomiting	3 (3.7)	0 (0)	1 (1.9)	0 (0)	2 (6.9)	0 (0)	1 (3.1)	0 (0)	2 (4.0)	0 (0)
Alopecia	3 (3.7)	0 (0)	1 (1.9)	0 (0)	2 (6.9)	0 (0)	1 (3.1)	0 (0)	2 (4.0)	0 (0)
Mucositis	1 (1.2)	0 (0)	1 (1.9)	0 (0)	0 (0)	0 (0)	1 (3.1)	0 (0)	0 (0)	0 (0)
AEs leading to dose reduction	35 (42.7)		23 (43.4)		12 (41.4)		16 (50.0)		19 (38.0)	
Proteinuria	11 (13.4)		4 (7.5)		7 (24.1)		6 (18.8)		5 (10.0)	
Hand-foot skin reaction	8 (9.8)		6 (11.3)		2 (6.9)		6 (18.8)		2 (4.0)	
Fatigue	6 (7.3)		4 (7.5)		2 (6.9)		3 (9.4)		3 (6.0)	
Liver function worsening	5 (6.1)		4 (7.5)		1 (3.4)		0 (0)		5 (10.0)	
Hypothyroidism	5 (6.1)		4 (7.5)		1 (3.4)		2 (6.3)		3 (6.0)	
Diarrhea	3 (3.7)		2 (3.8)		1 (3.4)		3 (9.4)		0 (0)	
Thrombocytopenia	1 (1.2)		1 (1.9)		0 (0)		0 (0)		1 (2.0)	
Anorexia	1 (1.2)		1 (1.9)		0 (0)		0 (0)		1 (2.0)	
Skin rash	1 (1.2)		1 (1.9)		0 (0)		0 (0)		1 (2.0)	
AEs leading to drug discontinuation	25 (30.5)	-	16 (30.2)		9 (31.0)		4 (8%)		21 (42.0)	
Liver function worsening	14 (17.1)		9 (17.0)		5 (17.2)		2 (6.3)		12 (24.0)	
Fatigue	7 (8.5)		4 (7.5)		3 (10.3)		1 (3.1)		6 (12.0)	
Proteinuria	2 (2.4)		2 (3.8)		0 (0)		0 (0)		2 (4.0)	
Gastrointestinal bleeding	1 (1.2)		0 (0)		1 (3.4)		0 (0)		1 (2.0)	
Skin rash	1 (1.2)		1 (1.9)		0 (0)		0 (0)		1 (2.0)	
Thrombocytopenia	1 (1.2)	-	1 (1.9)		0 (0)		1 (3.1)		0 (0)	

Note: Gr, Grade; AE, Adverse Event.