Supplementary

Supplement to: Xu J, et al. Camrelizumab in combination with apatinib in patients with advanced hepatocellular carcinoma (RESCUE): a non-randomized, open-label, phase 2 trial.

Table of Contents

Supplementary Methods

Inclusion and exclusion criteria

Inclusion criteria

Subjects must meet all of the following criteria to be eligible for this study:

1. Subjects must participate voluntarily and sign the informed consent form;
2. Aged ≥ 18 years old, males and females;
3. Pathologically confirmed advanced hepatocellular carcinoma (unresectable or metastatic), with at least one measurable lesion that has not been treated locally (must be ≥ 10 mm in longest diameter by spiral CT or ≥ 15 mm in shortest diameter for enlarged lymph nodes, as per RECIST 1.1);
4. Child-Pugh score ≤ 6 (Child-Pugh Class A);
5. BCLC Stage B-C;
6. Refractory to sorafenib or lenvatinib (disease progression or unacceptable toxicity), or unwilling or unable to afford sorafenib;
7. Ability to swallow tablets;
8. ECOG score of 0-1 (refer to Appendix I for ECOG criteria);
9. Expected survival ≥ 12 weeks;
10. Vital organ functions meet the following requirements (not including any use of blood components and cell growth factors within 14 days before the first dose):

Absolute neutrophil count ≥ 1.5 × 109/L;

Platelets ≥ 80 × 109/L;

Hemoglobin ≥ 90 g/L;

Serum albumin ≥ 28 g/L;

Thyroid stimulating hormone (TSH) ≤ 1×ULN (In case of abnormalities, FT3 and FT4 levels should be measured at the same time. If FT3 and FT4 levels are normal, the subject can be enrolled);

Bilirubin ≤ 1.5 × ULN (within 7 days prior to the first dose);

ALT and AST ≤ 3 × ULN (within 7 days prior to the first dose);

AKP ≤ 2.5 × ULN;

Serum creatinine ≤ 1.5 × ULN;

11. For female patients of childbearing potential or female patients who are not sterilized by surgical operations, they need to use a medically approved contraceptive measure (such as an intra-uterine contraceptive device, contraceptive pills or condoms) during the study treatment period and within 3 months after the end of the study treatment; For female patients of childbearing potential who are not sterilized by surgical operations, they must have a negative serum or urine HCG test result within 72 h prior to study enrollment; and they must not be in the lactation period; For male patients with partners of childbearing potential, they should take effective contraceptive measures during the study period and within 3 months after the last dose of SHR-1210;

Exclusion Criteria

Subjects meeting any of the following are not eligible to participate in this study:

1. Subjects with any active autoimmune diseases or a history of autoimmune diseases (including but not limited to the following: autoimmune hepatitis, interstitial pneumonia, uveitis, enteritis, hepatitis, hypophysitis, vasculitis, nephritis, hyperthyroidism; adult subjects with vitiligo or completely relieved childhood asthma can be enrolled if they do not require any intervention; subjects with asthma requiring medical intervention with bronchodilators cannot be enrolled);
2. Subjects who are currently using immunosuppressants, or systemic hormonal therapy for immunosuppression (>10 mg/day of prednisone or an equivalent dose of other therapeutic hormones) and still use the above drugs within 2 weeks prior to enrollment;
3. ≥ 2 lines of systemic treatments;
4. Subjects who have experienced severe allergic reactions to other monoclonal antibodies;
5. Patients with known CNS metastasis or hepatic encephalopathy;
6. Patients with liver tumor burden greater than 50% of total liver in volume, or patients who have previously undergone liver transplantation;
7. Patients with symptomatic ascites requiring paracentesis or drainage or patients who have undergone ascites drainage within the past 3 months, except for those with asymptomatic ascites of a small amount;
8. Patients with hypertension which cannot be well controlled by antihypertensives

(systolic pressure ≥ 140 mmHg or diastolic pressure ≥ 90 mmHg);

9. Uncontrolled cardiac diseases or symptoms, such as: (1) NYHA Class II or above heart failure, (2) unstable angina, (3) myocardial infarction within the past year, (4) clinically significant supraventricular or ventricular arrhythmia requiring treatment or intervention, or (5) QTc > 450 ms (males), or QTc > 470 ms (females);

10. Abnormal coagulation function (INR > 2.0, PT > 16 s), bleeding tendency or receiving thrombolytics or anticoagulant therapy. Prophylactic use of low-dose aspirin or low molecular weight heparin is allowed;

11. Clinically significant bleeding symptoms or clear bleeding tendency within 3 months prior to enrollment, such as coughing > 2.5 mL of blood daily, gastrointestinal bleeding, esophageal varices with bleeding risks, hemorrhagic gastric ulcer, or vasculitis. In case of positive fecal occult blood at baseline, a re-examination is needed, and a gastroscopy is required if it is still positive. The subject will be excluded if the gastroscopy indicates severe esophageal varices (except for those where such conditions have been ruled out by gastroscopy within 3 months prior to enrollment);

12. Events of arterial/venous thrombosis within 6 months prior to enrollment, such as cerebrovascular accidents (including transient ischemic attacks, cerebral hemorrhage, cerebral infarction), deep vein thrombosis, and pulmonary embolism;

13. Known hereditary or acquired hemorrhage and thrombophilia (such as hemophilia, coagulopathy, thrombocytopenia, etc.);

14. The routine urinalysis indicates that urine protein is ≥ ++ and confirms that 24h urine protein is ≥ 1.0 g;

15. Patients who have previously received radiotherapy, chemotherapy, hormone therapy or surgery that is less than 4 weeks before the study after the end of such treatments (last dose); molecular targeted therapy (including oral targeted drugs in other clinical trials) is less than 5 drug half-lives from the first dose; or patients with adverse events caused by previous treatment (except for alopecia) that have not returned to ≤ CTCAE grade 1;

16. Patients with active infection, fever ≥ 38.5 °C of unknown causes within 7 days prior to administration, or WBC count > 15 × 109/L at baseline;

17. Patients with congenital or acquired immunodeficiency (such as HIV positive);

18. HBV DNA > 2000 IU/mL (or 104 copies/mL); or HCV RNA > 103 copies/mL; or HBsAg+ and anti-HCV antibody positive;

19. Patients with other malignancies currently or within the past 3 years (except for cured basal cell carcinoma and cervical carcinoma in situ);

20. Patients with bone metastasis who have received a palliative radiotherapy in an area of > 5% of bone marrow area within 4 weeks prior to the participation in this study;

21. Patients who have previously received other anti-PD-1 antibody therapies or other immunotherapies targeting PD-1/PD-L1, or have previously received apatinib treatments;

22. Patients who have received live vaccines within less than 4 weeks before the first dose or would probably receive during the study;

23. Patients with other potential factors that may affect the study results or result in the premature termination of the study as determined by the investigator, such as alcoholism, drug abuse, other serious diseases (including mental illness) requiring concomitant treatment, serious laboratory abnormalities, accompanied by family or social factors that could affect the safety of the patients.

PD-L1 expression assessment

Two continuous slices at 3-5 μm were obtained from formalin fixed paraffin-embedded (FFPE) tissues of Hepatocellular Carcinoma (HCC) tumor and held in the dark at 2-8 °C. One slice was stained by Hematoxylin and Eosin (H&E) method firstly to assess the tissue histology and preservation quality, and ensure there were at least 100 viable tumor cells in this slice. Then, the other continuous slice was stained by PD-L1 IHC 22C3 pharmDx kit (Agilent Technologies, Santa Clara), an immunohistochemistry using Monoclonal Mouse Anti-PD-L1 to measure the expression of PD-L1 protein. The PD-L1 stain procedure was performed using EnVision FLEX visualization system on Autostainer Link 48 according to the instruction. Slide with two FFPE cell lines NCI-H226 with moderate PD-L1 protein expression and MCF-7 with negative PD-L1 protein expression was used as control in each staining run. The expression of PD-L1 protein was determined by Tumor Proportion Score (TPS), which is the percentage of viable tumor cells showing partial or complete membrane staining. Normal cells and tumor associated immune cells were excluded in the scoring for

PD-L1 expression. Ten representative tumor areas with or without cell membrane staining were observed and counted under 40 × high magnification. The TPS of each specimen was estimated independently by two experienced pathologists (pathologist A and B). Staining results were divided into four groups according to different TPS: <1%, ≥1% - <25%, ≥ 25% - <50%, ≥50%. Little difference was defined if the two pathologists' results fell into same or adjacent groups, TPS scored by pathologist A would be recorded on the report uniformly. On the contrary, controversial results would be discussed with third pathologist to reach an agreement.

Figure S1. Percentage change from baseline in target lesion assessed by IRC per RECIST v1.1

(A) First-line cohort. (B) Second-line cohort.

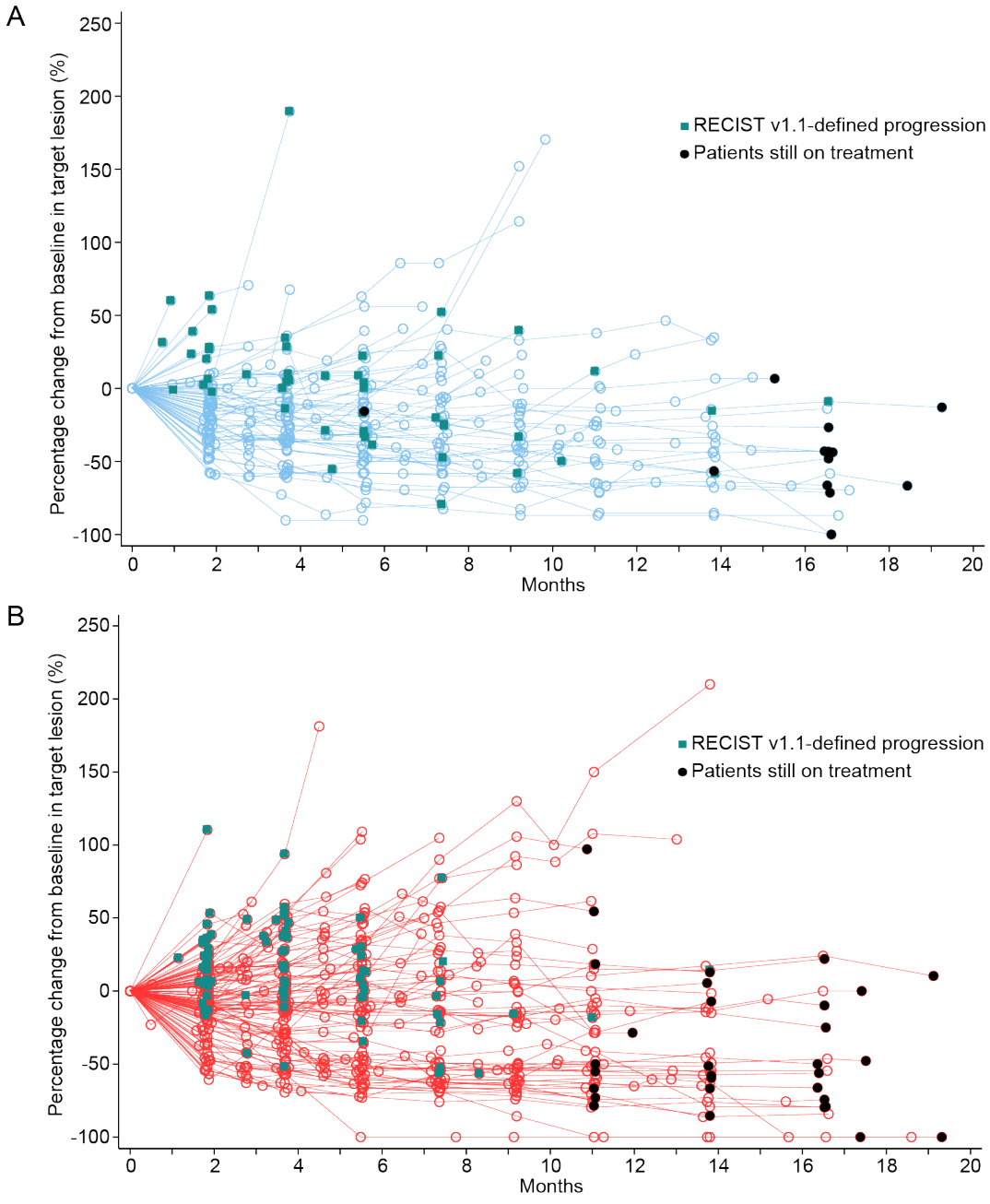
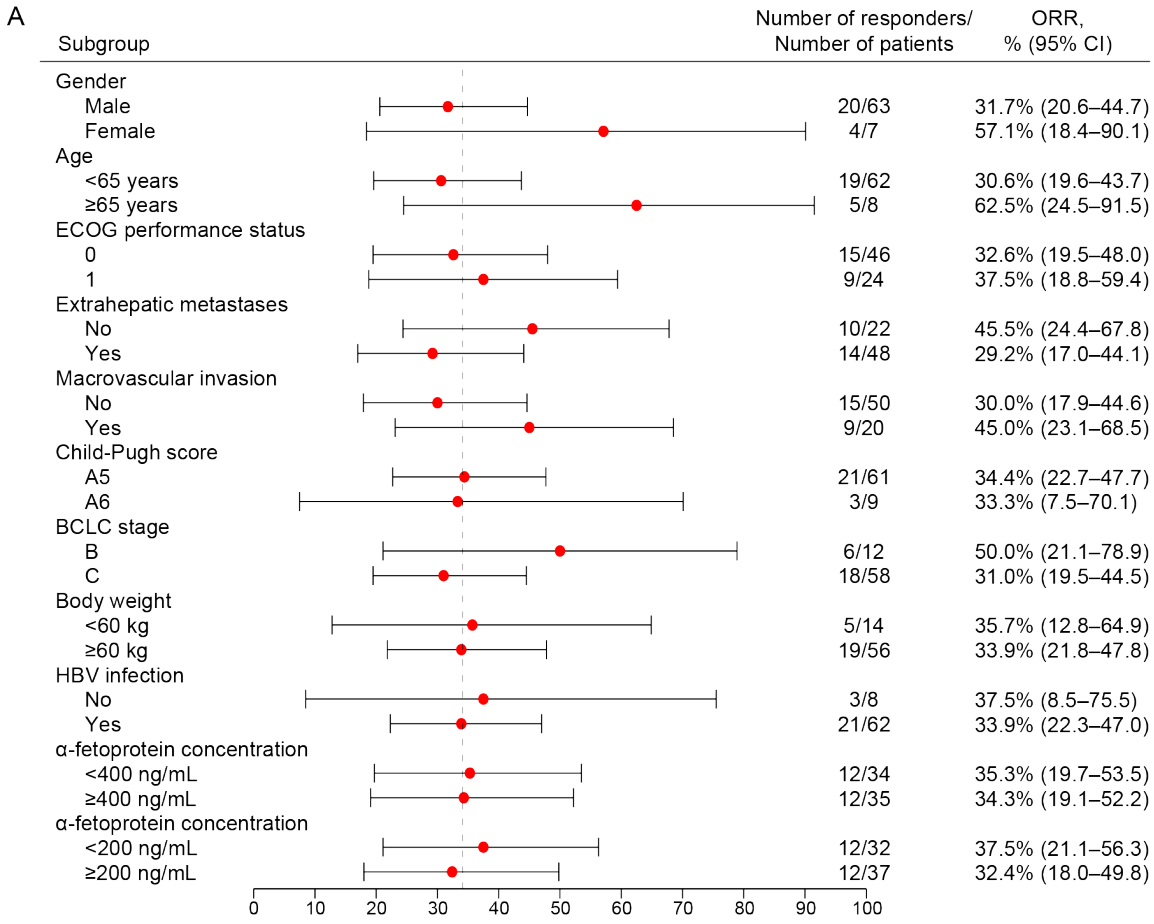


Figure S2. Subgroup analyses of ORR

(A) First-line cohort. (B) Second-line cohort.



B

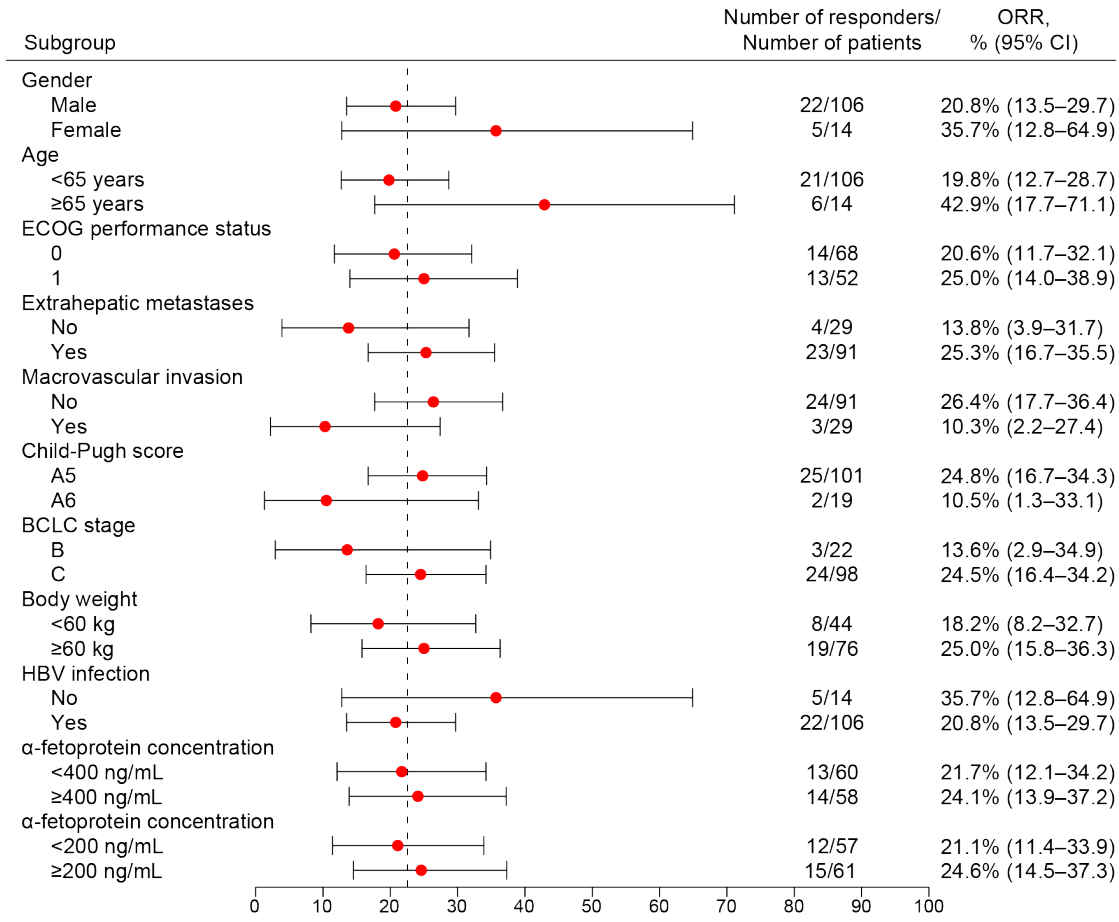

Subgroup
Number of responders/ Number of patients
ORR, % (95% CI)
Gender
Male 22/106 20.8% (13.5–29.7)
Female 5/14 35.7% (12.8–64.9)
Age
<65 years 21/106 19.8% (12.7–28.7)
≥65 years 6/14 42.9% (17.7–71.1)
ECOG performance status
0 14/68 20.6% (11.7–32.1)
1 13/52 25.0% (14.0–38.9)
Extrahepatic metastases
No 4/29 13.8% (3.9–31.7)
Yes 23/91 25.3% (16.7–35.5)
Macrovascular invasion
No 24/91 26.4% (17.7–36.4)
Yes 3/29 10.3% (2.2–27.4)
Child-Pugh score
A5 25/101 24.8% (16.7–34.3)
A6 2/19 10.5% (1.3–33.1)
BCLC stage
B 3/22 13.6% (2.9–34.9)
C 24/98 24.5% (16.4–34.2)
Body weight
<60 kg 8/44 18.2% (8.2–32.7)
≥60 kg 19/76 25.0% (15.8–36.3)
HBV infection
No 5/14 35.7% (12.8–64.9)
Yes 22/106 20.8% (13.5–29.7)
α-fetoprotein concentration
<400 ng/mL 13/60 21.7% (12.1–34.2)
≥400 ng/mL 14/58 24.1% (13.9–37.2)
α-fetoprotein concentration
<200 ng/mL 12/57 21.1% (11.4–33.9)
≥200 ng/mL 15/61 24.6% (14.5–37.3)
0 10 20 30 40 50 60 70 80 90 100

Figure S3. Kaplan-Meier plot of PFS

(A–B) PFS in first-line cohort (A) and second-line cohort (B) assessed by investigator per RECIST v1.1. (C–D) PFS in first-line cohort (C) and second-line cohort (D) assessed by IRC per mRECIST.

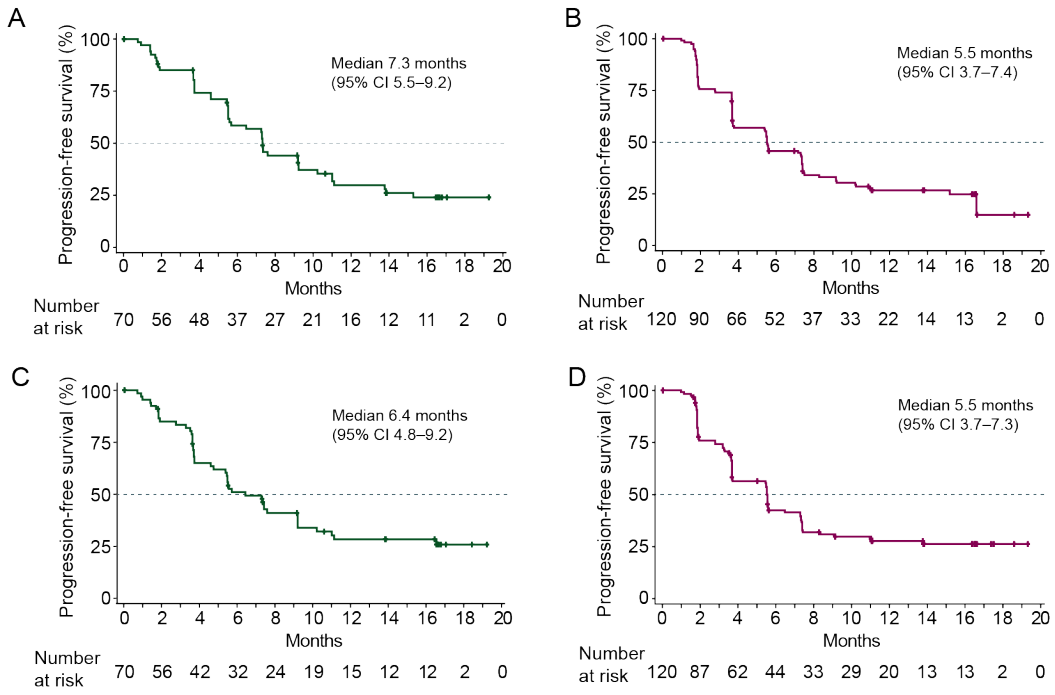


Figure S4. Kaplan-Meier plots of OS in patients with AFP <400 ng/mL and ≥400 ng/mL

(A) First-line cohort. (B) Second-line cohort. P value was calculated using Pearson Chi-Square test.

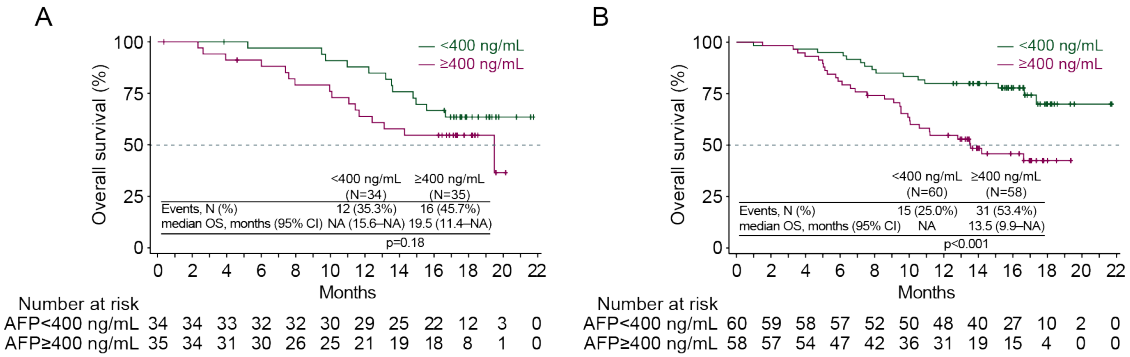


Table S1. Participating investigators

Principal investigator	Institution	Number of patients
Jianming Xu	The Fifth Medical Center, General Hospital of the People's Liberation Army, Beijing, China	26
Jie Shen	The Affiliated Drum Tower Hospital of Nanjing University Medical School, Nanjing, China	20
Shanzhi Gu	Hunan Cancer Hospital, Changsha, China	16
Jian Wu / Lihua Wu	The First Affiliated Hospital, College of Medicine, Zhejiang University, Hangzhou, China	14
Guoliang Shao	Zhejiang Cancer Hospital, Hangzhou, China	12
Yanqiao Zhang	Harbin Medical University Cancer Hospital, Harbin, China	9
Li Xu	Sun Yat-Sen University Cancer Center, Guangzhou, China	9
Zhenggang Ren	Zhongshan Hospital, Fudan University, Shanghai, China	8
Tao Yin	Hubei Cancer Hospital, Affiliated Hubei Cancer Hospital of Huazhong University of Science and Technology, Wuhan, China	8
Jingfeng Liu	Mengchao Hepatobiliary Hospital of Fujian Medical University, Fuzhou, China	8
Jianping Xiong	The First Affiliated Hospital of Nanchang University, Nanchang, China	7
Xianhai Mao	Hunan People's Hospital , Changsha, China	7
Ling Zhang	Henan Cancer Hospital, Zhengzhou, China	7
Jiayin Yang	West China Hospital of Sichuan University, Chengdu, China	5
Xiaoming Chen	Guangdong Provincial People's Hospital, Guangzhou, China	5
Lequn Li	Guangxi Medical University Affiliated Tumor Hospital, Nanning, China	5

Zhanyu Pan	Tianjin Medical University Cancer Institute and Hospital, Tianjin, China	4
Kangsheng Gu	The First Affiliated Hospital of Anhui Medical University, Hefei, China	4
Zhiming Wang	Xiangya Hospital Central South University, Changsha, China	4
Xi Chen	900 Hospital of the Joint Logistics Support Force, Fuzhou, China	4
KuanSheng Ma	The First Hospital Affiliated to AMU, Chongqing, China	3
Xinmin Zhou	The First Affiliated Hospital of The Fourth Military Medical University, Xi'an, China	2
Guowen Yin	Jiangsu Cancer Hospital, Nanjing, China	1
Enxiao Li	The First Affiliated Hospital of Xi'an Jiaotong University, Xi'an, China	1
Zujiang Yu	First Affiliated Hospital of Zhengzhou University, Zhengzhou, China	1

Table S2. Summary of patients who received post-discontinuation therapy

	First-line (*N*=70)	**Second-line (*N*=120)**
Receiving at least one anti-tumor therapy[a]	31 (44.3)	60 (50.0)
Chemotherapy	2 (2.9)	4 (3.3)
Radiotherapy	7 (10.0)	16 (13.3)
Targeted therapy	24 (34.3)	48 (40.0)
Surgery	3 (4.3)	1 (0.8)
Interventional therapy	9 (12.9)	17 (14.2)
Immunotherapy	6 (8.6)	10 (8.3)
Others	2 (2.9)	3 (2.5)

Data are *N* (%). [a]Post-discontinuation anti-tumor therapies did not include camrelizumab monotherapy, apatinib monotherapy, or combination of camrelizumab and apatinib.

Table S3. Correlation of baseline APF level and ORR assessed by IRC per RECIST v1.1

	First-line cohort		Second-line cohort	
	Responders/ Total	ORR (95% CI)	Responders/ Total	ORR (95% CI)
<400 ng/mL	13/60	21.7% (12.1–34.2)	12/34	35.3% (19.7–53.5)
≥400 ng/mL	14/58	24.1% (13.9–37.2)	12/35	34.3% (19.1–52.2)
P value	/	0.93	/	0.75

P value was calculated using Pearson Chi-Square test.

Table S4. PD-L1 expression results

Patient	Tumor Proportion Score (TPS)		Patient	Tumor Proportion Score (TPS)
#1	0%		#28	1%
#2	1%		#29	0%
#3	0%		#30	0%
#4	2%		#31	20%
#5	0%		#32	0%
#6	0%		#33	10%
#7	0%		#34	5%
#8	0%		#35	0%
#9	0%		#36	0%
#10	1%		#37	3%
#11	2%		#38	1%
#12	0%		#39	0%
#13	0%		#40	3%
#14	3%		#41	0%
#15	0%		#42	3%
#16	0%		#43	0%
#17	3%		#44	0%
#18	2%		#45	0%
#19	4%		#46	0%
#20	0%		#47	1%
#21	10%		#48	0%
#22	0%		#49	0%
#23	0%		#50	2%
#24	0%		#51	0%
#25	5%		#52	2%
#26	2%		#53	0%
#27	0%		#54	0%

Table S5. Correlation of PD-L1 expression and efficacy assessed by IRC per RECIST v1.1

	PD-L1 TPS <1% (*N*=32)	**PD-L1 TPS ≥1% (*N*=22)**
Objective response	6 (18.8)	7 (31.8)
Progression-free survival events	24 (75.0)	14 (63.6)

Data are *N* (%). TPS, tumor proportion score.

Table S6. Adverse event clusters and related preferred terms

Cluster	Preferred Term
Hypertension	Hypertension, Blood pressure increased
Proteinuria	Proteinuria, Protein urine present
Hyperbilirubinaemia	Hyperbilirubinaemia, Jaundice, Yellow skin, Bilirubin conjugated increased, Blood bilirubin increased, Blood bilirubin unconjugated increased
Thrombocytopenia	Platelet count decreased
Leukopenia	White blood cell count decreased
Neutropenia	Neutrophil count decreased, Granulocyte count decreased, Neutrophil percentage decreased
Asthenia	Asthenia, Fatigue
Diarrhoea	Diarrhoea, Diarrhoea infectious
Rash	Rash maculo-papular, Erythema, Rash, Rash papular, Eczema, Drug eruption
Abdominal pain	Abdominal discomfort, Abdominal tenderness, Abdominal pain, Abdominal distension, Hepatic pain, Perihepatic discomfort, Abdominal pain upper, Abdominal pain lower, Gastrointestinal flatulence
Hypothyroidism	Hypothyroidism, Tri-iodothyronine decreased, Blood thyroid stimulating hormone increased, Primary hypothyroidism, Tri-iodothyronine free decreased
Haematuria	Red blood cells urine positive, Blood urine present, Haematuria
Hypoalbuminaemia	Hypoalbuminaemia, Hypoproteinaemia, Blood albumin decreased
Decreased appetite	Hypophagia, Decreased appetite
Anaemia	Red blood cell count decreased, Anaemia, Mean cell haemoglobin concentration decreased, Haemoglobin decreased, Haemorrhagic anaemia
Hepatic function	Hepatic function abnormal, Hepatic enzyme abnormal

abnormal	
Lymphopenia	Lymphocyte count decreased
Hypokalaemia	Hypokalaemia, Blood potassium decreased
Hepatotoxicity	Hepatic failure, Hepatic encephalopathy, Chronic hepatic failure, Immune-mediated hepatitis, Drug-induced liver injury

Table S7. Immune-related adverse events (irAEs)

	First-line (*N*=70)	Second-line (*N*=120)	Total (*N*=190)
Requiring corticoids due to irAE	12 (17.1)	14 (11.7)	26 (13.7)
Leading to camrelizumab interruption	9 (12.9)	9 (7.5)	18 (9.5)
Leading to camrelizumab discontinuation	1 (1.4)	3 (2.5)	4 (2.1)
Any	24 (34.3)	29 (24.2)	53 (27.9)
Hypothyroidism	5 (7.1)	11 (9.2)	16 (8.4)
Rash	4 (5.7)	3 (2.5)	7 (3.7)
Hyperglycaemia	4 (5.7)	2 (1.7)	6 (3.2)
Diabetes mellitus	3 (4.3)	1 (0.8)	4 (2.1)
Pneumonitis	1 (1.4)	3 (2.5)	4 (2.1)
Hyperthyroidism	1 (1.4)	2 (1.7)	3 (1.6)
Immune-mediated hepatitis	0	3 (2.5)	3 (1.6)
Hepatic function abnormal	1 (1.4)	1 (0.8)	2 (1.1)
Cystitis	0	2 (1.7)	2 (1.1)
Cystitis haemorrhagic	0	2 (1.7)	2 (1.1)
Lymphocytic hypophysitis	1 (1.4)	0	1 (0.5)
Ketoacidosis	1 (1.4)	0	1 (0.5)
Eczema	1 (1.4)	0	1 (0.5)
Drug eruption	1 (1.4)	0	1 (0.5)
Drug-induced liver injury	1 (1.4)	0	1 (0.5)
Interstitial lung disease	0	1 (0.8)	1 (0.5)
Immune-mediated pneumonia	0	1 (0.8)	1 (0.5)
Hepatic enzyme abnormal	1 (1.4)	0	1 (0.5)
Aspartate aminotransferase increased	1 (1.4)	0	1 (0.5)
Blood bilirubin increased	1 (1.4)	0	1 (0.5)
Blood alkaline phosphatase increased	1 (1.4)	0	1 (0.5)
Platelet count decreased	1 (1.4)	0	1 (0.5)

Asthenia	1 (1.4)	0	1 (0.5)
Immune-mediated enterocolitis	1 (1.4)	0	1 (0.5)

Data are *N* (%).

Table S8. Immune-related adverse events of grade 3 or greater

Immune-related adverse events	First-line (*N*=70)	Second-line (*N*=120)	Total (*N*=190)
Grade 3 or greater	8 (11.4)	6 (5.0)	14 (7.4)
Immune-mediated hepatitis	0	3 (2.5)	3 (1.6)
Pneumonitis	1 (1.4)	2 (1.7)	3 (1.6)
Hepatic function abnormal	1 (1.4)	1 (0.8)	2 (1.1)
Hepatic enzyme abnormal	1 (1.4)	0	1 (0.5)
Hyperglycaemia	1 (1.4)	0	1 (0.5)
Diabetes mellitus	1 (1.4)	0	1 (0.5)
Aspartate aminotransferase increased	1 (1.4)	0	1 (0.5)
Ketoacidosis	1 (1.4)	0	1 (0.5)
Blood alkaline phosphatase increased	1 (1.4)	0	1 (0.5)
Platelet count decreased	1 (1.4)	0	1 (0.5)
Drug-induced liver injury	1 (1.4)	0	1 (0.5)
Drug eruption	1 (1.4)	0	1 (0.5)

Data are *N* (%).