Supplemental Methods

CT Imaging Protocol

CECT scans were performed with a multidetector row four, 16, or 64 slice CT scanner (Light-Speed; GE Healthcare, Piscataway, NJ or Somatom; Siemens Healthcare, Germany), with a collimation of 2.5-5 mm, section thickness of 3-5 mm and reconstruction of 2.5-3 mm using a bi- or triphasic liver protocol. Parameters used for CT varied with patient size and were, on average, 120 kv with mAs 200 to 350. For the triphasic protocol, images were obtained 30-35, 60-70, and 180-300 seconds after the start of intravenous injection of 120 mL of ioversol (Optiray 240; Mallinckrodt, St. Louis, MO), iohexol (Omnipaque 350; Nycomed Amersham, Piscataway, New Jersey) or iodixanol (Visipaque; GE Healthcare, Princeton, New Jersey) at a rate of 5 mL/s. For the biphasic technique, images were obtained at 30-35 seconds and then at 60-70 seconds after the start of contrast injection at a rate of 2 to 3 mL/s. Determination of RVI score was based on the portal venous phase images for both bi- and triphasic studies.

Revised RVI Algorithm

In the initial ‘proof of concept’ study, the RVI algorithm was constructed from the two most strongly associated CT traits, thereby capturing 85% of the available radiogenomic-associated venous invasion gene expression modules.(1) For this study, the third most represented trait, “tumor-liver difference,” was also included to achieve broader gene expression coverage (100%) and improved diagnostic accuracy.

Supplemental Results

Patient Characteristics

The cohort included 117 males and 40 females with a median age of 56 years (IQR, 50 to 64). The etiology of liver disease was hepatitis C in 48 patients (31%), hepatitis B in 38 (24%), alcohol use in 8 (6%) and more than one etiology in 29 (18%) while 34 (21%) had no identifiable etiology. Median AFP value measured at the time of CT was 17.4 ng/mL (IQR, 5 to 136.3 ng/mL). Seventy-seven patients (49%) had AFP values less than or equal to 20 ng/mL, 67 patients (43%) had values greater than 20 ng/mL and 13 patients (8%) had no data available. One hundred and twenty patients (72%) had a single lesion while 37 patients (28%) had two or three lesions. Median diameter of the index tumors evaluated for RVI was 2.8 cm (IQR, 1.8 to 4.5 cm). Tumor size distribution showed that 82 patients (52%) had an index tumor smaller than or equal to 3 cm in diameter and 75 patients (48%) had a tumor greater than 3 cm. AJCC-TNM staging classified 92 patients (59%) as stage 1, 56 (35%) as stage 2 and 9 (6%) as stage 3a. Child-Pugh score (CPS) classified 81 patients (52%) as class A, 57 (36%) as class B, 10 (6%) as class C and 3 patients did not have data for CPS classification.

Seventy-two patients (46%) underwent surgical resection while 85 patients (54%) underwent LT. Median time from CECT assessed for RVI to surgery was 2.3 months (IQR, 0.75 to 5.25 months). Of those who underwent LT, 78 patients (92%) were within Milan criteria while the remaining 7 exceeded Milan criteria but were within UCSF criteria.(2) To maintain eligibility for LT, 35 (45%) of the 78 patients within Milan criteria underwent bridging locoregional therapy following preoperative CT.(3) Of the 7 patients who were within UCSF criteria but outside Milan criteria, 6 (86%) underwent

locoregional therapy to downstage their status to within Milan criteria.(4) Six of the surgical resection patients (8%) also received locoregional therapy.

One hundred and seven patients (68%) had histologic evidence of cirrhosis. Thirty-three lesions (21%) were nuclear grade 1 out of 4, 40 (26%) nuclear grade 2, 47 (30%) nuclear grade 3, 13 (8%) nuclear grade 4 and in 24 patients (15%) grade was not reported. In the 41 patients who underwent locoregional therapy, necrotic tissue did not impede histologic determination of MVI.(5, 6) Pathologic evaluation of explanted tissue revealed no macrovascular vascular invasion undetected by imaging.

Supplemental References

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6. Kim PT, Onaca N, Chinnakotla S, Davis GL, Jennings LW, McKenna GJ, Ruiz RM, et al. Tumor biology and pre-transplant locoregional treatments determine outcomes in patients with T3 hepatocellular carcinoma undergoing liver transplantation. Clin Transplant 2013;27:311-318.

	RVI Positive (N=41)	RVI Negative (N=116)	*P*
Median age, years (IQR)	56 (51-62)	56 (50-63)	0.334
Sex (%)			
Male	75.6	74.1	0.853
Female	24.4	25.9	0.853
Surgery (%)			
Resection	56.1	42.2	0.127
Liver Transplantation	43.9	57.8	0.127
Etiology of Liver Disease (%)			
HCV	34.1	29.3	0.646
HBV	17.1	26.7	0.421
Alcohol	2.4	6	0.187
Multiple	17.1	19	0.85
Unknown	29.3	19	0.11
Median AFP, ng/mL (IQR)	89.8 (11.2-816.5)	14.7 (4.7-73.8)	0.002
Imaging (IQR)			
Median time from imaging to surgery, months†	1.5 (25.5-97.5)	2 (29-155)	0.001
Median tumor size, cm†	3.5 (2.2-3.5)	2.6 (1.6-4.4)	0.024
Median number of lesions	1 (1-1)	1 (1-1)	0.711
Pathology			
Grade (IQR)†	3 (2-3)	2 (1-3)	0.008
Cirrhosis (%)†	57.9	75.2	0.043
Staging (IQR)			
Median AJCC-TNM*	2 (1-2)	1 (1-2)	0.07
Median Child-Pugh Score	7 (6-8)	6 (5-8)	0.706

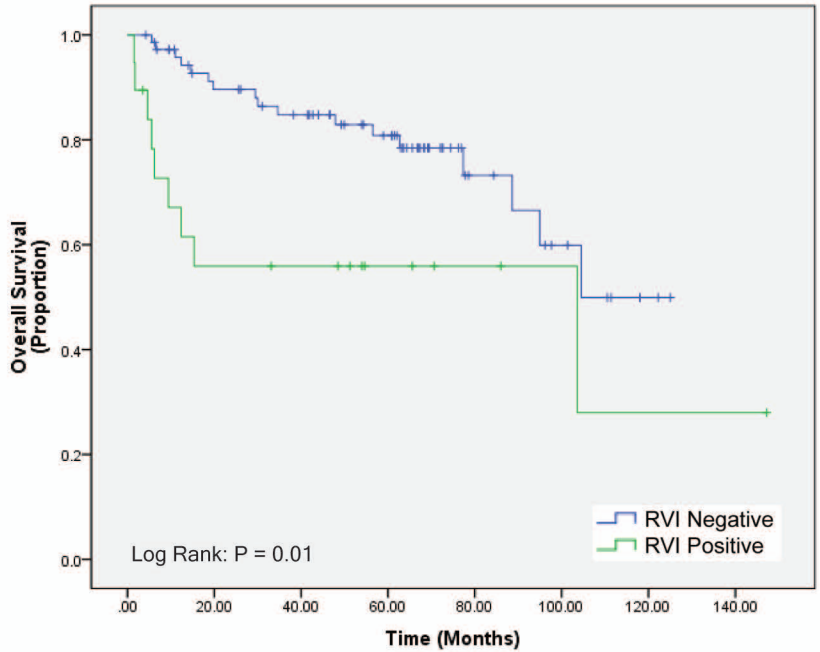
*: AJCC-TNM stages were assigned ascending numerical values

†: $p<0.05$ (Mann–Whitney U test)

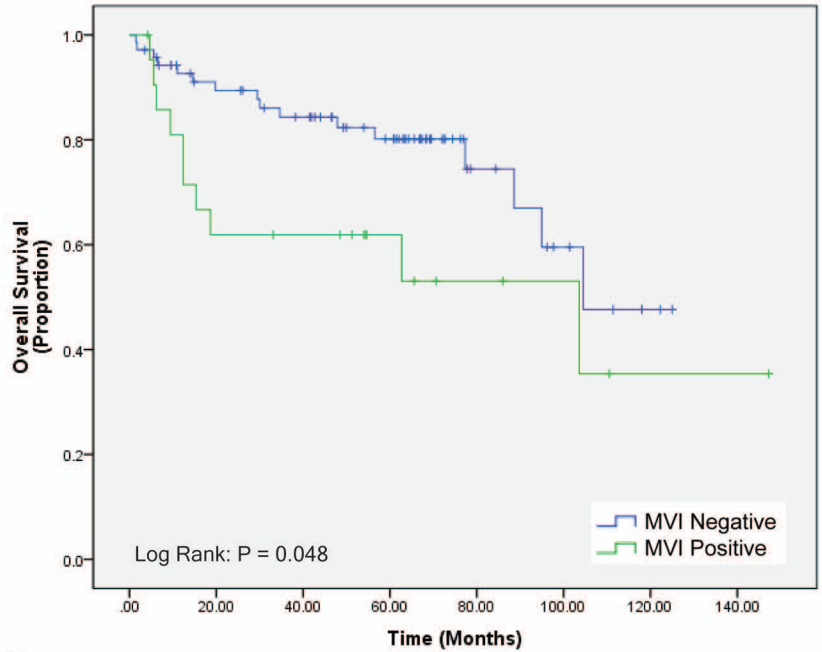
			Univariate		Multivariate (MVI)		Multivariate (RVI)	
	No. of Pts	Recurrence Free Survival (%)	*P*	HR (95% CI)	*P*	HR (95% CI)	*P*	HR (95% CI)
Age	157	53	0.438	0.99 (0.96-1.01)	0.706	1.01 (0.97-1.04)	0.712	1.01 (0.97-1.04)
Sex								
Male	117	53	0.895	0.95 (0.46-1.97)	0.437	0.71 (0.30-1.69)	0.47	0.72 (0.30-1.73)
Female	40	52						
HBV								
Yes	55	62	0.467	1.27 (0.67-2.44)				
No	102	49						
HCV								
Yes	77	55	0.035	0.48 (0.24-0.95)				
No	80	52						
AFP*	144	74	0.051	1.23 (1.00-1.51)	0.269	1.15 (0.92-1.43)	0.09	1.21 (0.97-1.51)
Locoregional therapy								
Downstaged	6	67	0.641	1.65 (0.20-13.4)				
Bridged	35	63	0.155	0.50 (0.19-1.29)				
Tumor size*	157	72	<0.001	1.17 (1.09-1.25)	<0.001	1.18 (1.09-1.28)	<0.001	1.18 (1.09-1.28)
No. of Lesions								
Single	120	51						
Two to three	37	60	0.244	1.33 (0.82-2.17)	0.185	1.49 (0.83-1.77)	0.207	1.45 (0.82-2.57)
Nuclear grade*	133	73	0.086	1.33 (0.96-1.83)	0.285	1.25 (0.86-1.82)	0.147	1.31 (0.91-1.89)
MVI								
Yes	45	33	0.001	2.94 (1.53-5.63)	0.161	1.59 (0.72-3.54)		
No	112	61						
RVI								
Yes	41	27	<0.001	3.48 (1.81-6.70)			0.007	2.74 (1.31-5.73)
No	116	62						

*For continous variables, 3 year RFS is listed for the subjects who had greater than the average of the variable

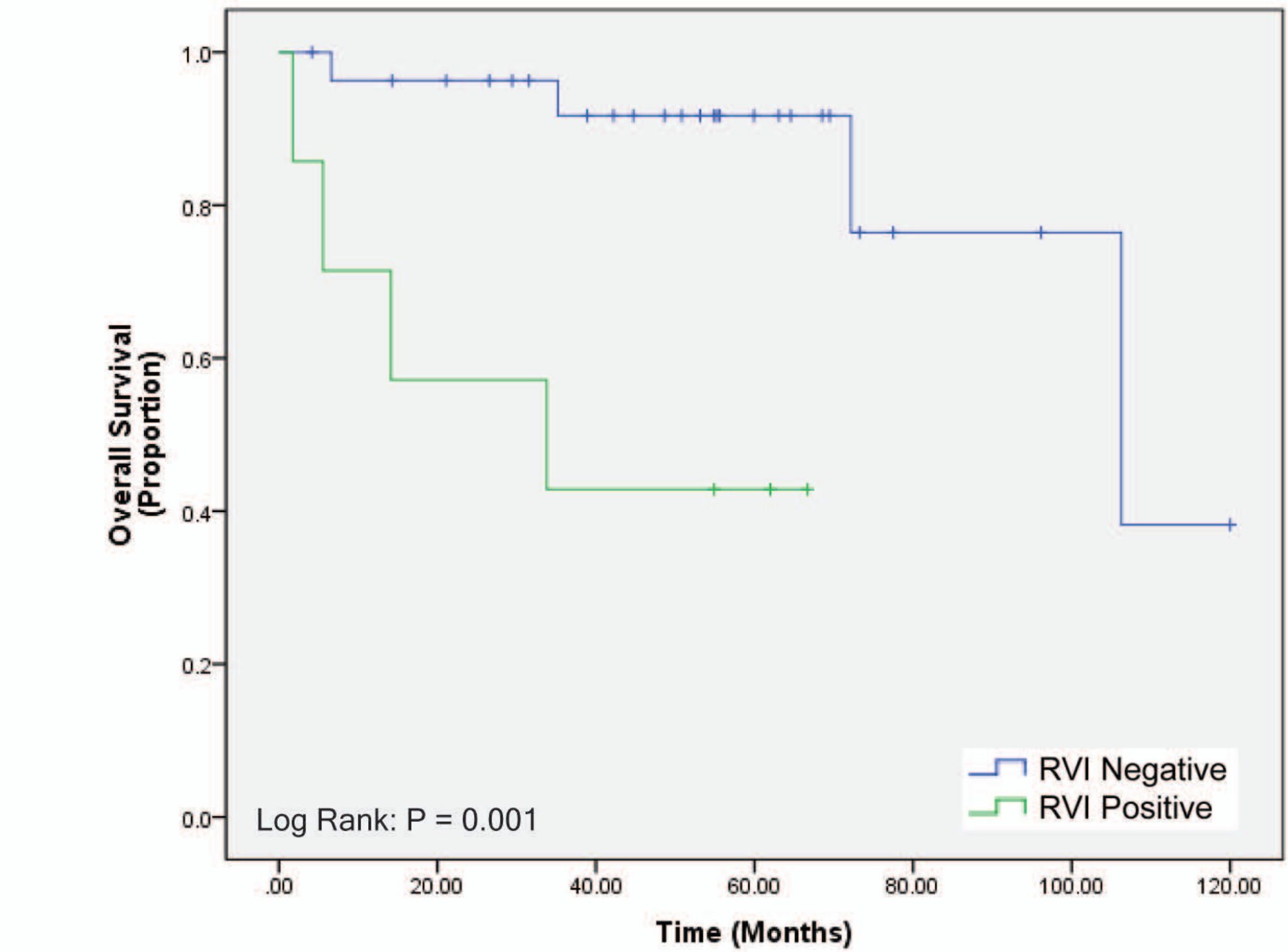
	N	*P*	HR	CI 95%
Tumor Size				
≤ 3 cm	82	0.001	3.87	1.71-8.72
> 3 cm	75	0.011	2.88	1.27-6.53
AFP				
≤ 20 ng/mL	77	0.02	3.15	1.19-8.12
> 20 ng/mL	67	<0.001	3.71	1.84-7.97



No. at Risk							
RVI Negative	72	58	51	38	12	7	2
RVI Positive	18	10	9	5	3	2	1



No. at Risk							
MVI Negative	69	55	48	36	11	6	2
MVI Positive	21	13	12	7	4	3	1



No. at Risk						
RVI Negative	27	25	19	10	3	2
RVI Positive	6	4	3	1		