Presenter Disclosure Information

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The following relationships exist related to this presentation:

No Relationships to Disclose

The Predictive Value of Centre Tumour CD8$^+$ T Cells in Patients with Hepatocellular Carcinoma: Comparison with Immunoscore

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National Harbor, MD
Society for Immunotherapy of Cancer 2015

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TNM Staging of Liver Cancer
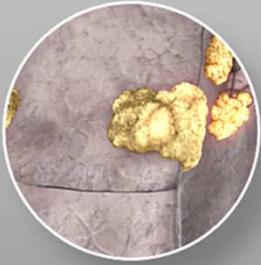
Tumor size

T1 – A tumor is less than 3 cm (1 ½ inches) in size

T2 – The tumor is greater than 3 cm

T3 – The tumor can be any size, but is near the airway or has spread to local areas such as the chest wall or diaphragm

T4 – The tumor is any size, but is located in the airway, or has invaded local structures such as the heart or the esophagus.

Lymph Nodes

N0 – No lymph nodes are affected

N1 – The tumor has spread to nearby nodes on the same side of the body

N2 – The tumor has spread to nodes further away but on the same side of the body

N3 – Cancer cells are present in lymph nodes on the other side of the chest from the tumor, or in nodes near the collarbone or neck muscles

Metastases

M0 – No metastases are present

M1 – The tumor has spread (metastasized) to other regions of the body or the other lung

SCIENTIFIC ANIMATIONS. ASHISH KHERA on September , 2015

Immune infiltrate

Cancer-associated fibroblast

Tumour cell

Pericyte

Lymphatic vessel

Blood vessel

Vascular network
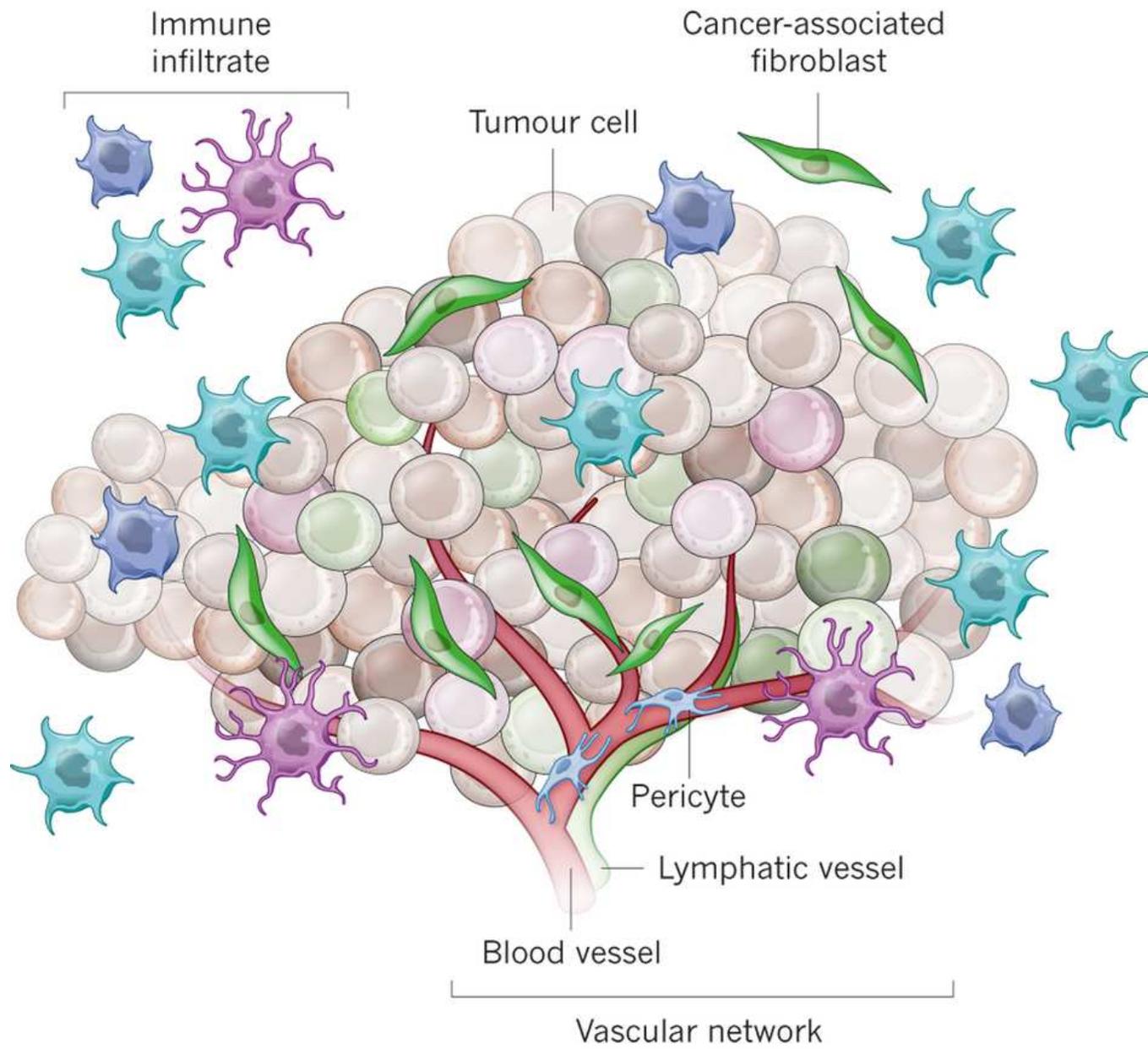
Immunoscore on Colon Cancer
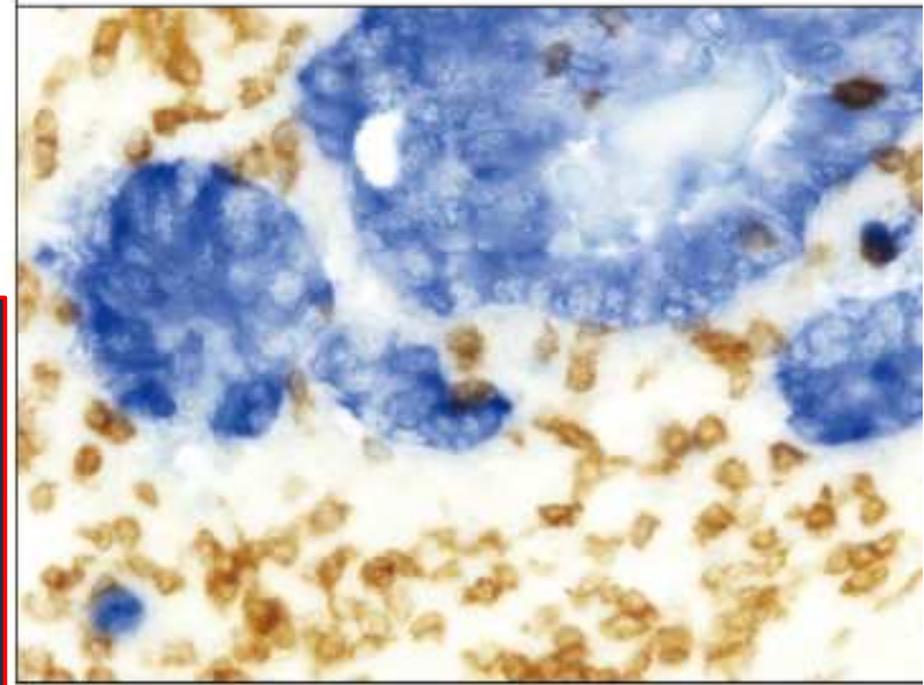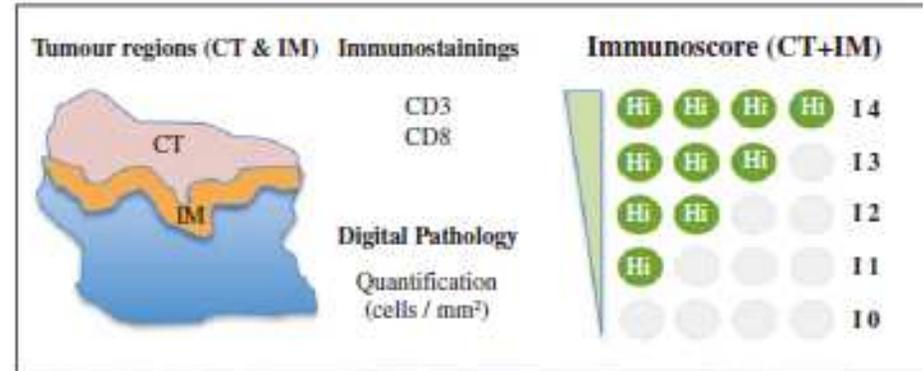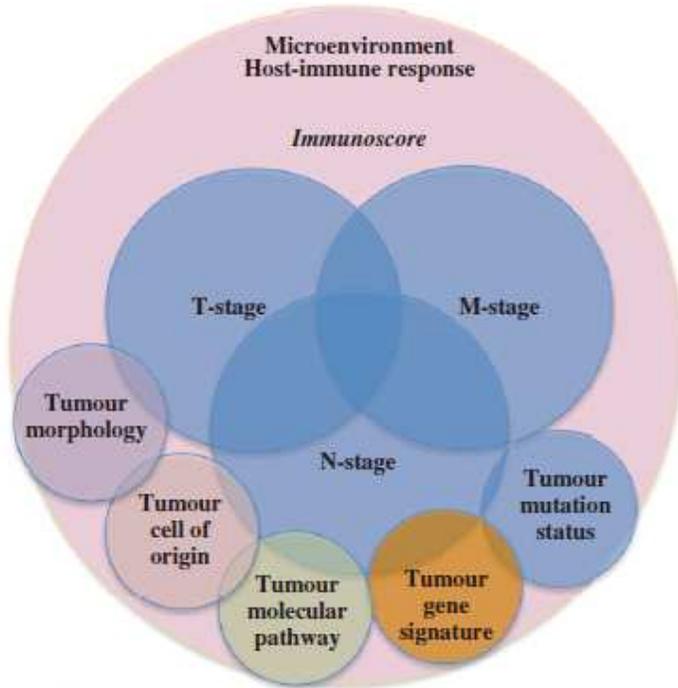
Immunoscore®CC
Indication: Colon Cancer
Clinical Utility: Improve the classification & identify patients at risk
**Material:** FFPE slides from tumor resection
Target: T cells CD3 + and CD8 +
Location: Centre and periphery of the tumor
Technology: Immunohistochemistry and digital pathology

Galon J, Mlecnik B, Bindea G, Angell HK, Berger A, Lagorce C, et al. Towards the introduction of the 'Immunoscore' in the classification of malignant tumours. The Journal of pathology 2014;232:199-209.

Immunostainings

CD3
CD8

Digital Pathology

Quantification
(cells / mm²)
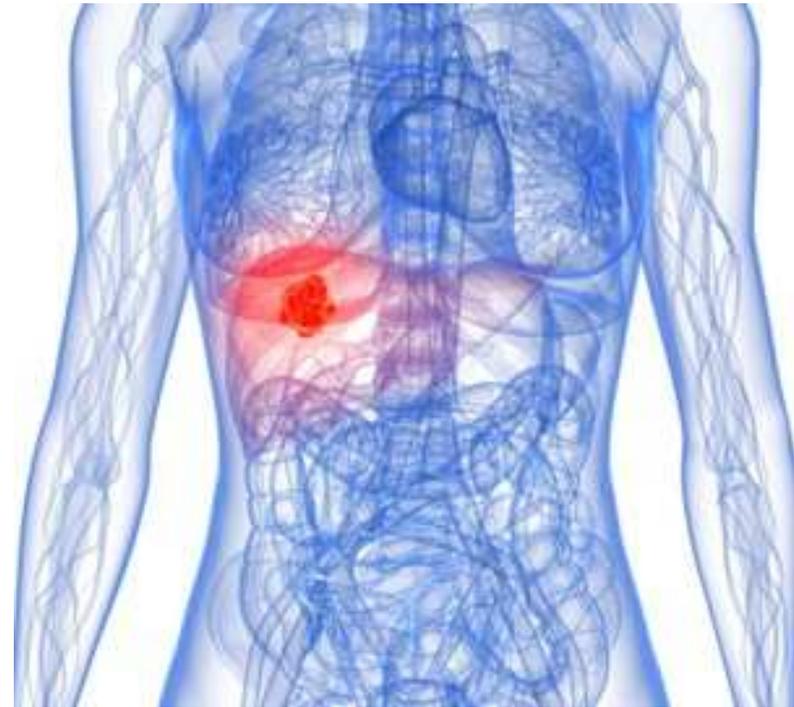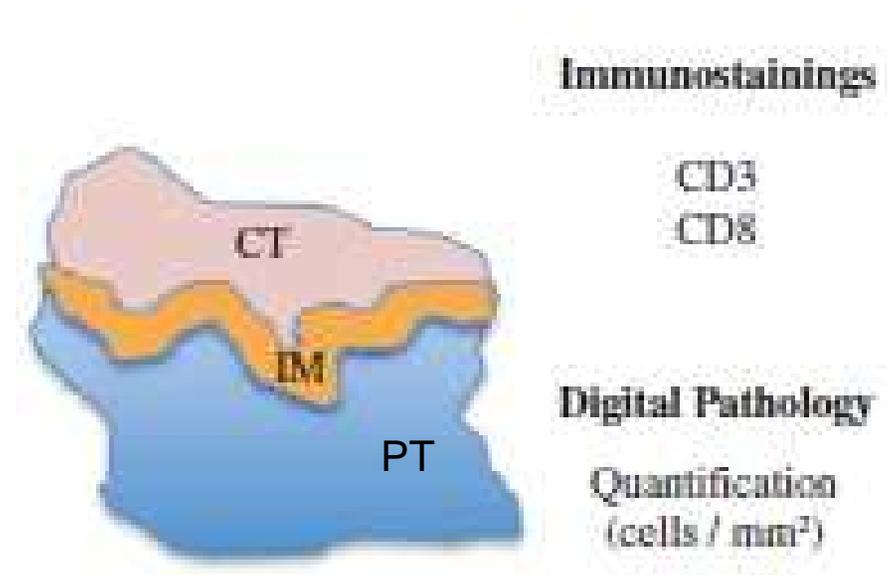
CT: Centre Tumour regions

IM: Invasive Margin regions

PT: Peritumour regions

Two HCC cohorts (Total 449)

Center of The Tumor (CT)

Peritumoral Tissues (PT)
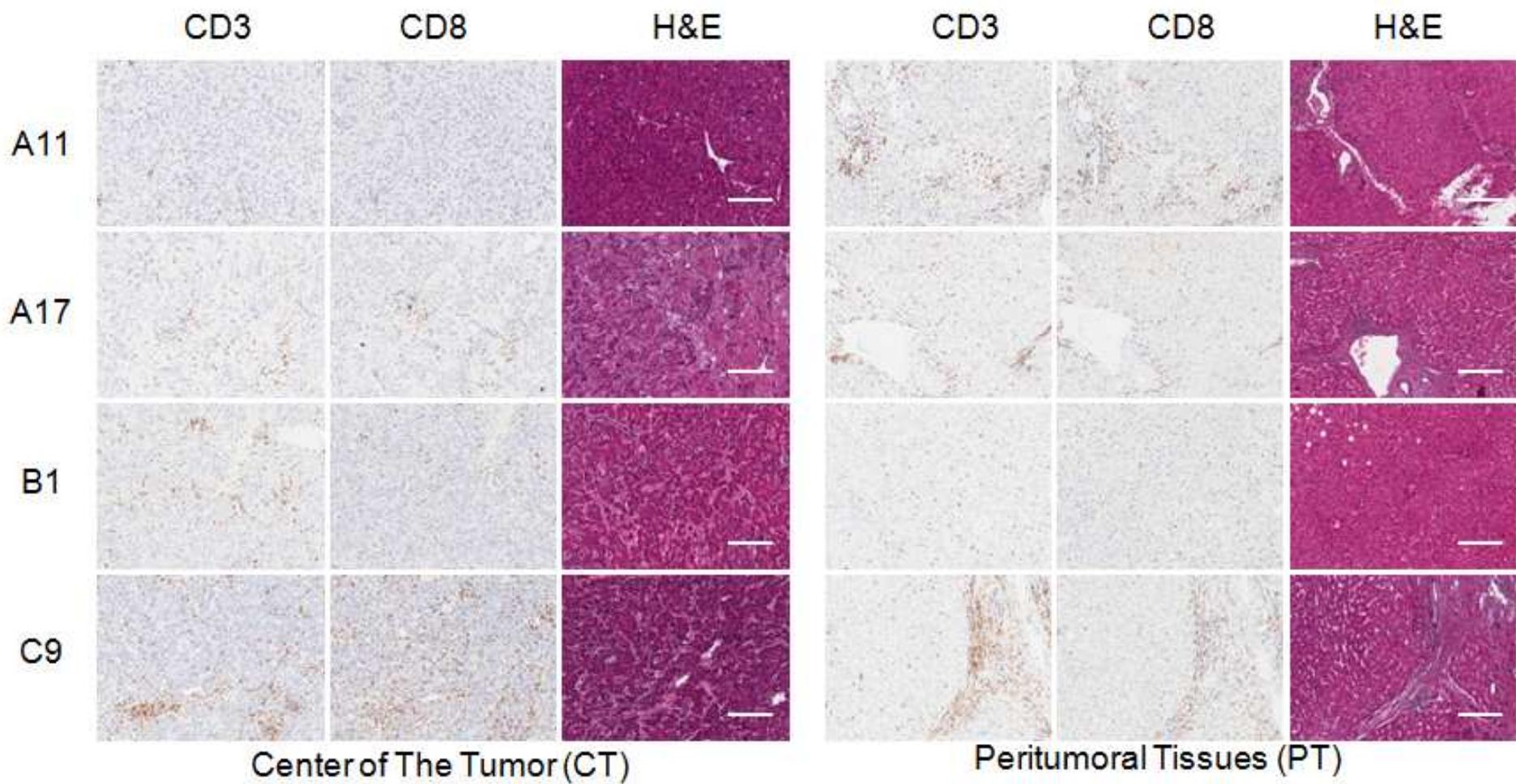
Original magnification: ×10. Bar=200 µm

Positive correlations of the densities of CD3$^+$ or CD8$^+$ cells/mm^2 in centre tumour but not peritumour to overall survival in Cohort 1
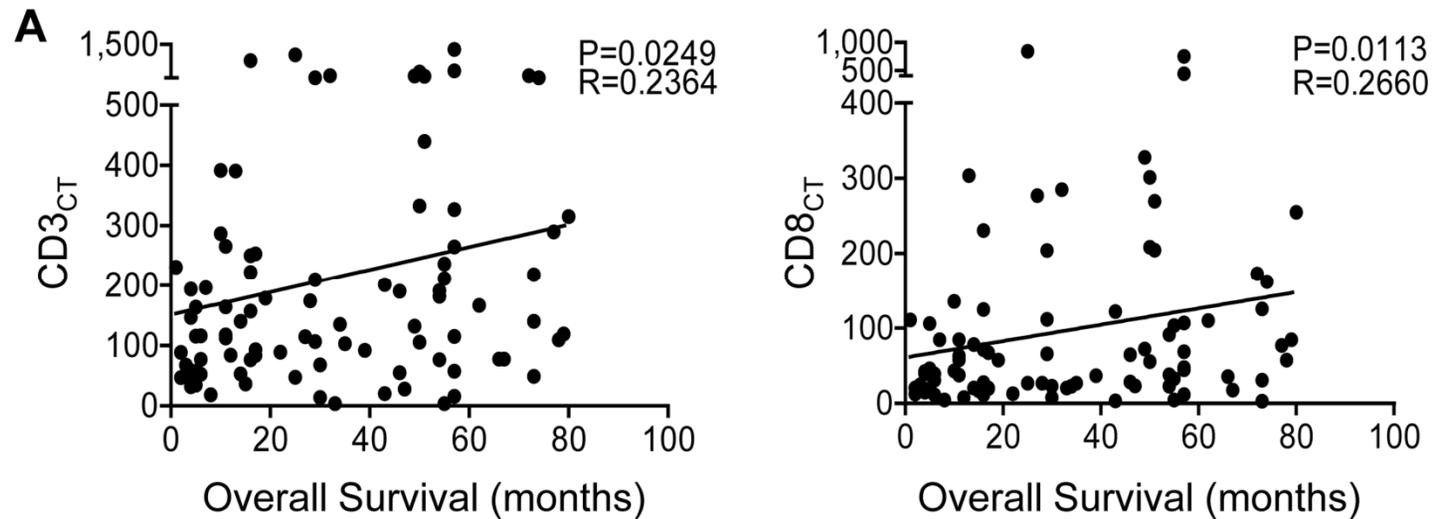
Cohort 1 (90 patients with liver cancer)

Figure 1. Correlation between the number of CD3$^+$/CD8$^+$ cells/mm^2 and OS in CT and PT regions.

Positive correlations of the densities of CD3^{+} or CD8^{+} cells/mm^2 in centre tumour but not peritumour to overall survival in Cohort 1

B
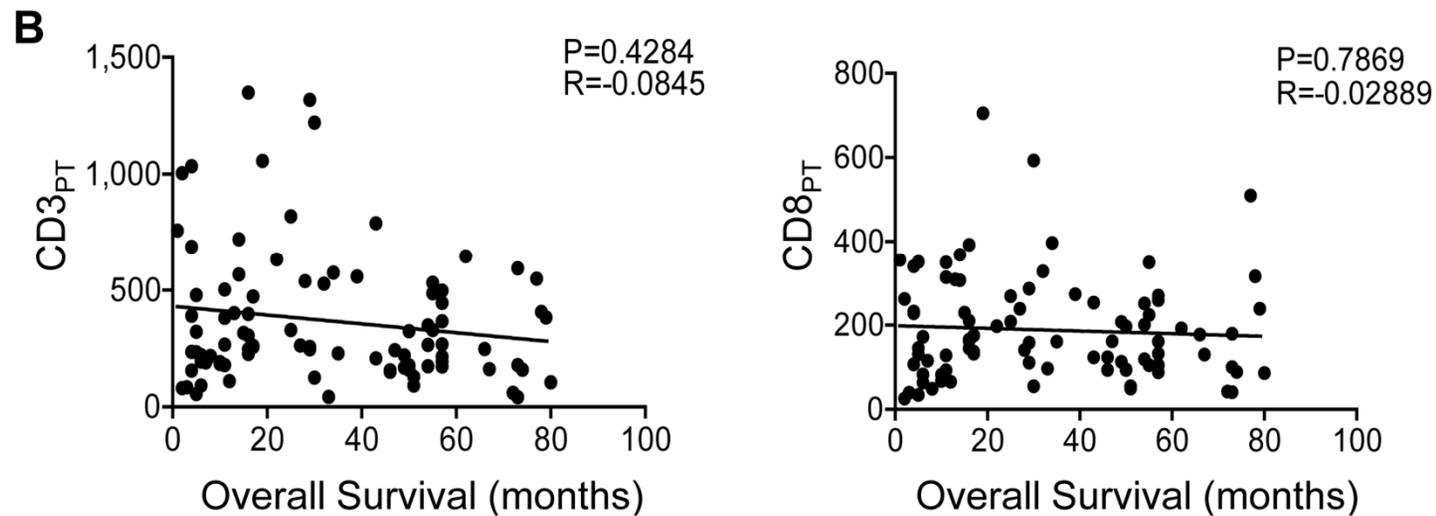
Figure 1. Correlation between the number of CD3^{+}/CD8^{+} cells/mm^2 and OS in CT and PT regions.

Positive correlations of the densities of CD3$^+$ or CD8$^+$ cells/mm^2 in centre tumour but not peritumour to overall survival in Cohort 1
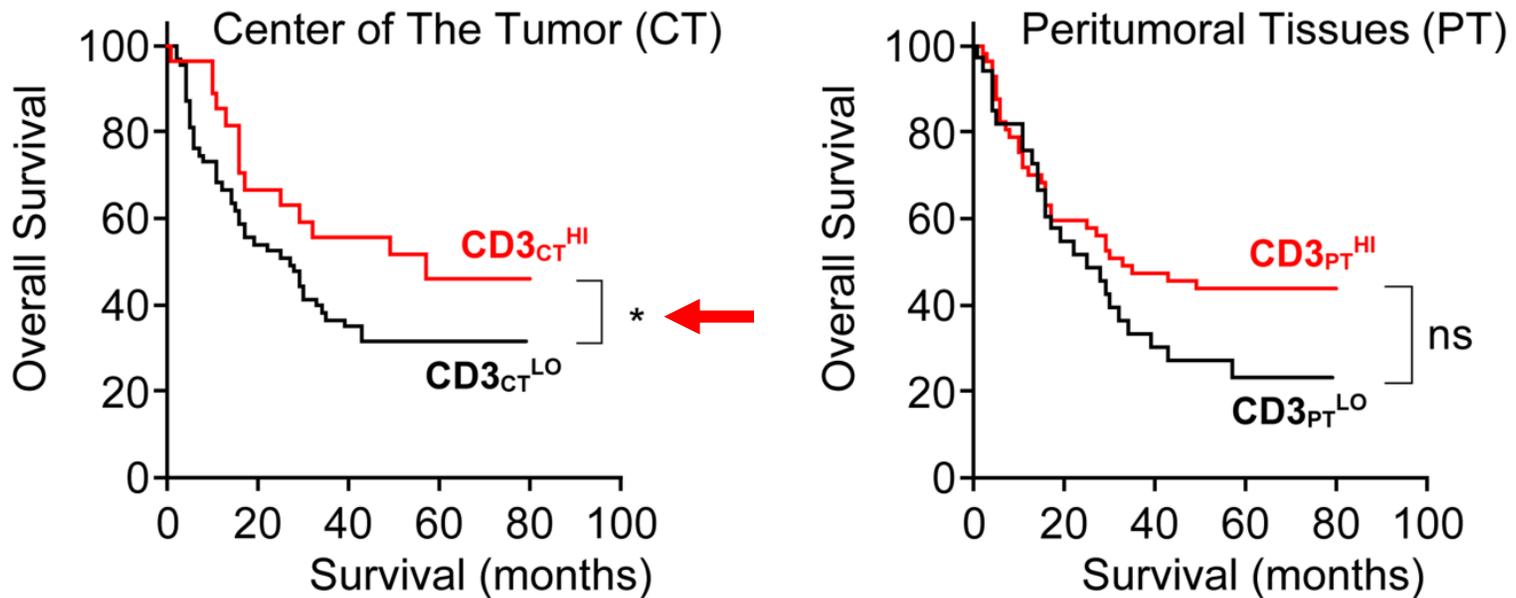
(The minimum P-value Cut-off values were 214 and 375 for CD3$_{CT}$ and CD3$_{PT}$, respectively)

Figure 1. Correlation between the number of CD3$^+$/CD8$^+$ cells/mm^2 and OS in CT and PT regions.

Positive correlations of the densities of CD3$^+$ or CD8$^+$ cells/mm^2 in centre tumour but not peritumour to overall survival in Cohort 1
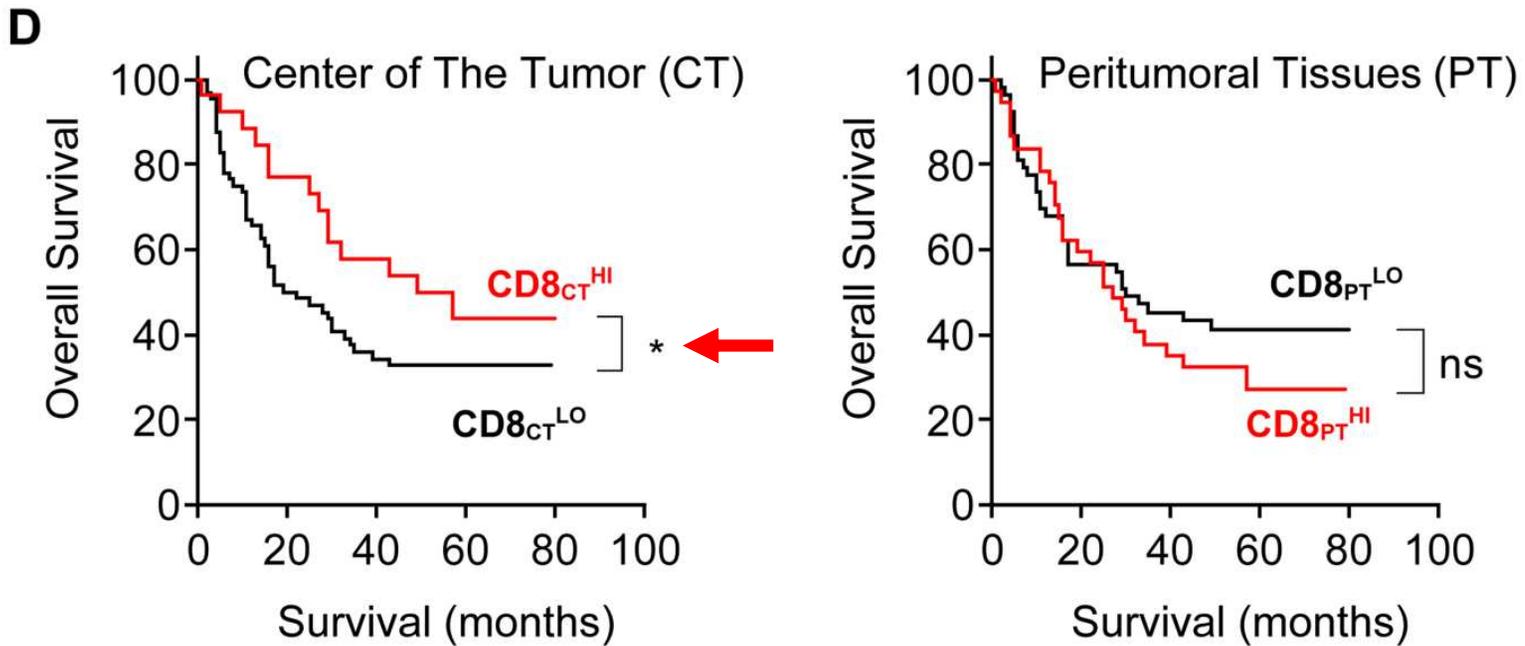

(The minimum P-value Cut-off values were 97 and 186 for CD8$_{CT}$ and CD8$_{PT}$, respectively)

Figure 1. Correlation between the number of CD3$^+$/CD8$^+$ cells/mm^2 and OS in CT and PT regions.

Result :

Positive correlations of the densities of CD3$^+$ or CD8$^+$ cells/mm^2
in centre tumour but not peritumour to overall survival

Part   1   Centre Tumour but not Peritumour

Invasive Margin

Positive correlations of the densities of CD3$^+$ or CD8$^+$ cells/mm^2 in centre tumour but not peritumour to overall survival in Cohort 1
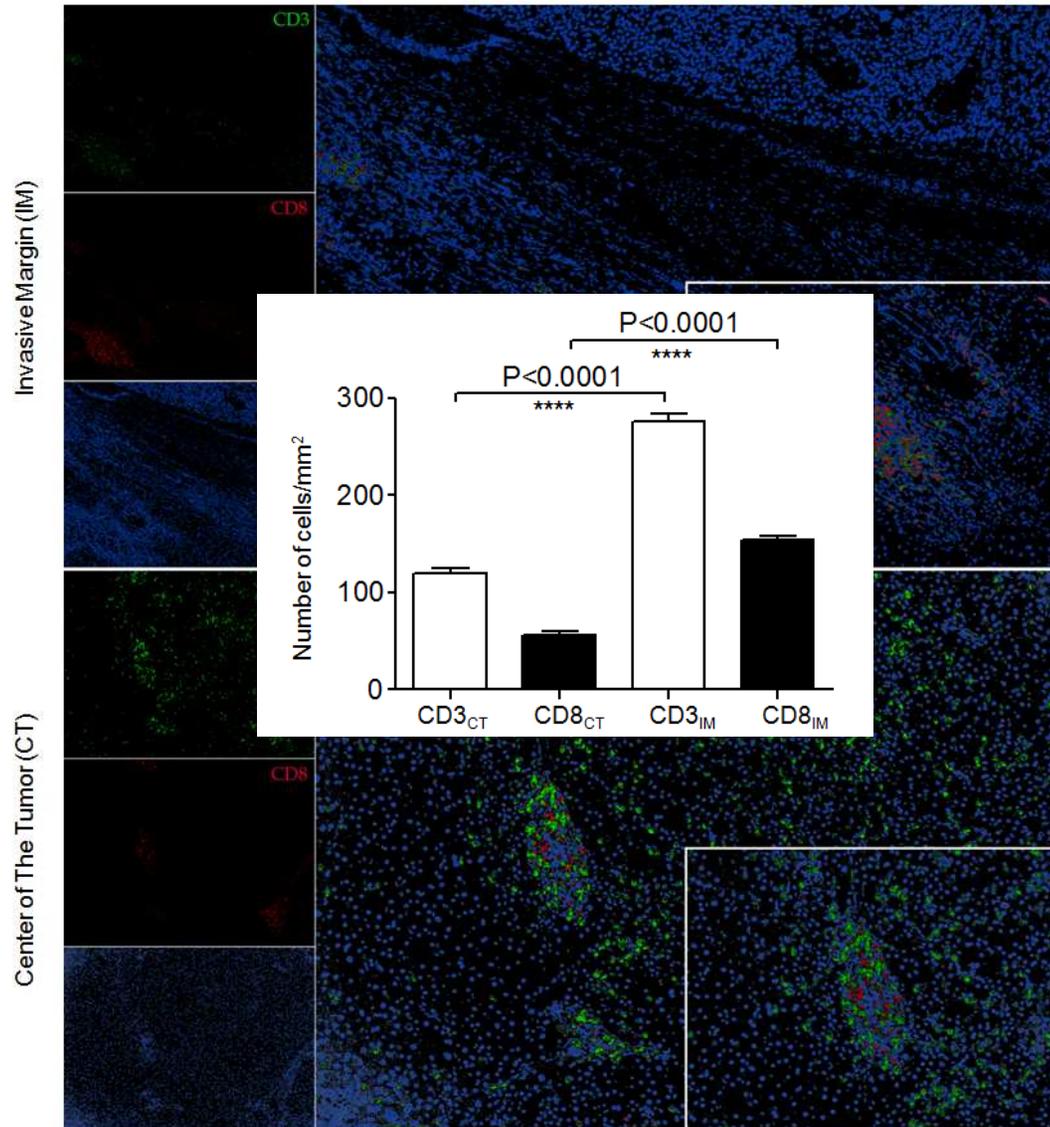
CD3⁺ or CD8⁺ T cells in invasive margin and centre tumour in a large Cohort 2
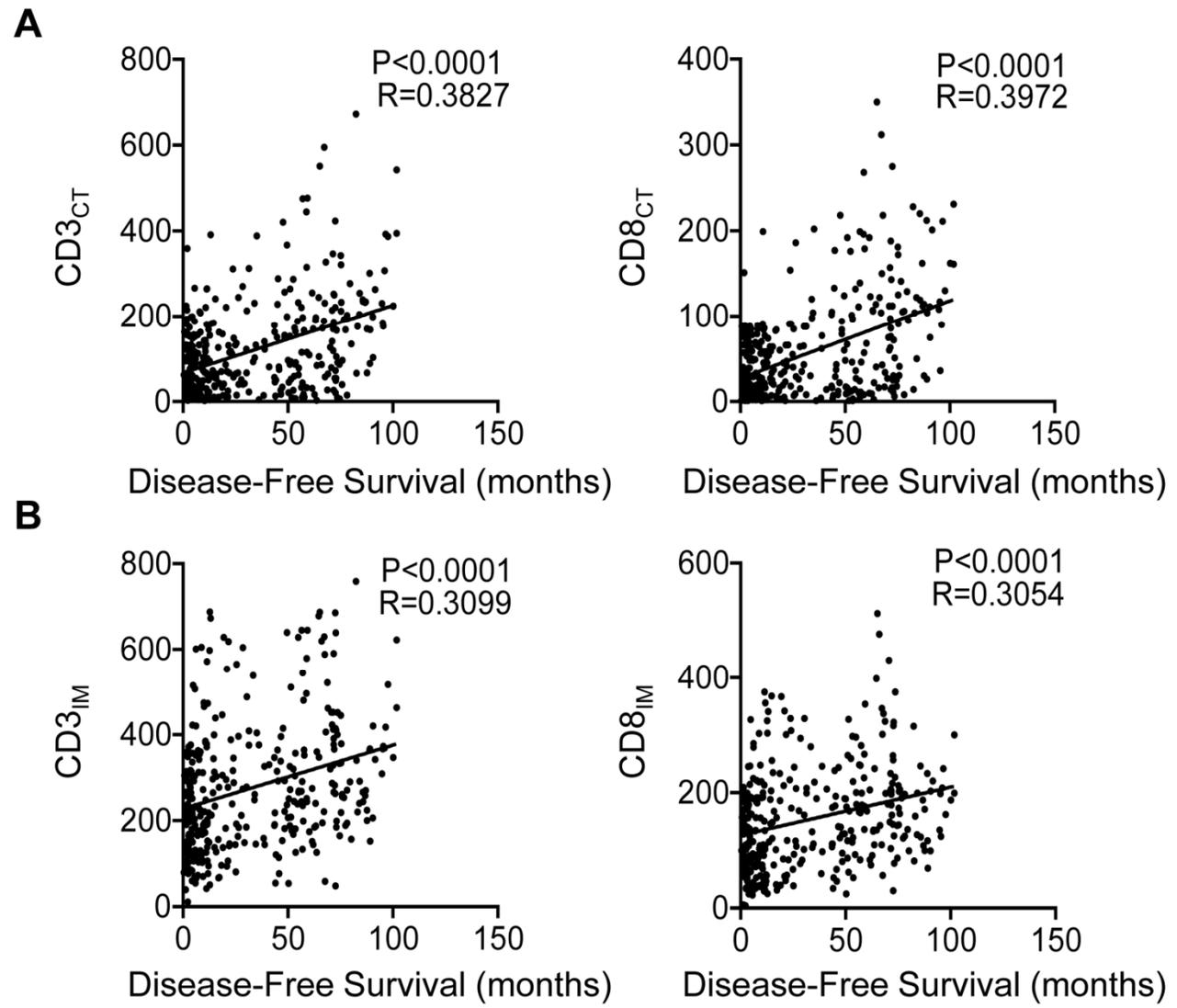
Cohort 2 (359 patients with liver cancer)

Figure 3. Positive correlation between the number of CD3⁺ or CD8⁺ cells/mm² and survival time in HCC patients.

Result :

**CD3+ or CD8+ T cells in invasive margin and centre tumour
in a large Cohort 2**

Part   2   Invasive Margin

                                       Immunoscore

CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC

Disease-Free Survival vs **Survival (months)**

Groups	P
I0 v I1	.0024 S
I0 v I2	< .0001 S
I0 v I3	< .0001 S
I0 v I4	< .0001 S
I1 v I2	.1357
I1 v I3	.0884
I1 v I4	.0001 S
I2 v I3	.3534
I2 v I4	.0004 S
I3 v I4	.0078 S

Overall Survival vs **Survival (months)**

Groups	P
I0 v I1	.1175
I0 v I2	< .0001 S
I0 v I3	.0002 S
I0 v I4	< .0001 S
I1 v I2	.0106 S
I1 v I3	.0089 S
I1 v I4	< .0001 S
I2 v I3	.3034
I2 v I4	.0020 S
I3 v I4	.0192 S
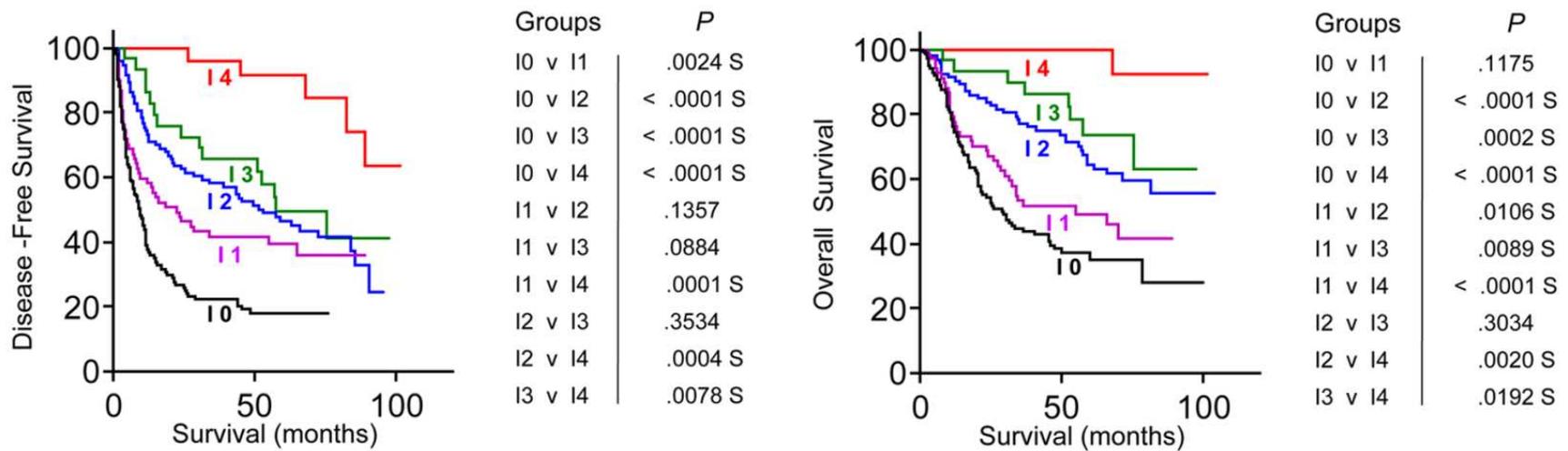
359 HCC sections were divided into IS-0 (n=143), IS-1 (n=68), IS-2 (n=95), IS-3 (n=29) and IS-4 (n=24)

Figure 5. Increased survival time for the patient with high immunoscore (IS).

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Table 1. Univariate Analysis of DFS and OS Among Patients With Liver Cancer (cohort 2) According to Clinical or Immune Parameters

Parameter	No. of pts (%)	DFS			OS		
		HR	95% CI	$P*$	HR	95% CI	$P*$
Immune parameters							
Immune score		1.63	1.44 to 1.85	<0.0001†	1.69	1.45 to 1.96	<0.0001†
0	143(39.8)		4.44 to 27.48			4.05 to 210.15	
1	68(18.9)		2.27 to 14.8			2.83 to 151.01	
2	95(26.5)		1.7 to 10.69			1.56 to 82.97	
3	29(8.1)		1.2 to 9.1			0.95 to 61.04	
4	24(6.7)		(reference)			(reference)	
CD3$_{CT/IM}$							
(LoLo)	191(53.2)	1.0	(reference)		1.0	(reference)	
(Het)	123(34.3)	0.46	0.34 to 0.62	<0.0001†	0.5	0.35 to 0.7	<0.0001†
(HiHi)	45(12.5)	0.23	0.14 to 0.39	<0.0001†	0.18	0.09 to 0.38	<0.0001†
CD8$_{CT/IM}$							
(LoLo)	171(47.6)	1.0	(reference)		1.0	(reference)	
(Het)	152(42.3)	0.46	0.35 to 0.61	<0.0001†	0.46	0.33 to 0.64	<0.0001†
(HiHi)	36(10)	0.16	0.09 to 0.31	<0.0001†	0.1	0.04 to 0.27	<0.0001†

CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC
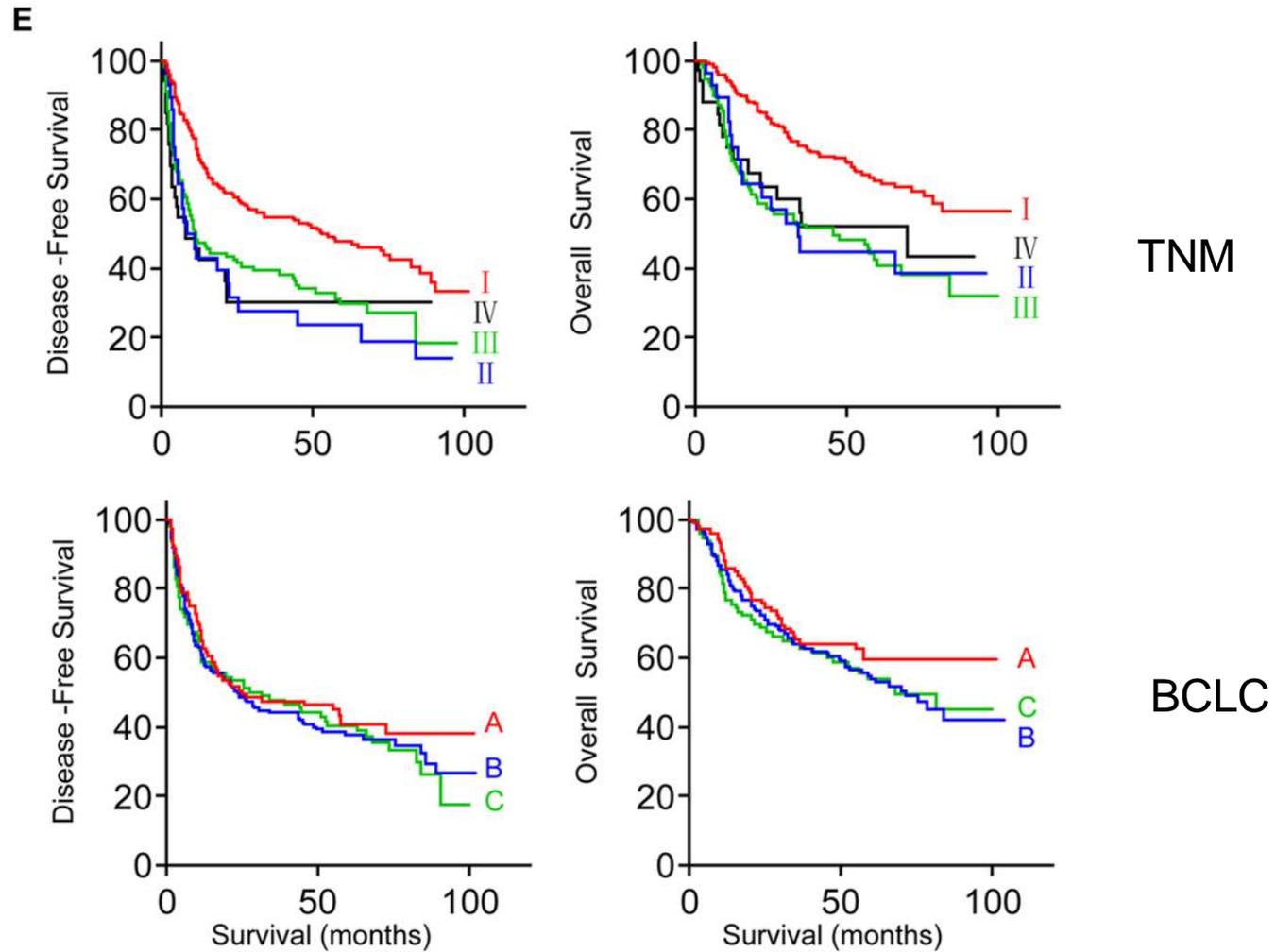
Figure 5. Increased survival time for the patient with high immunoscore (IS).

# CD3^{+} or CD8^{+} T cells in invasive margin and centre tumour in a large Cohort 2
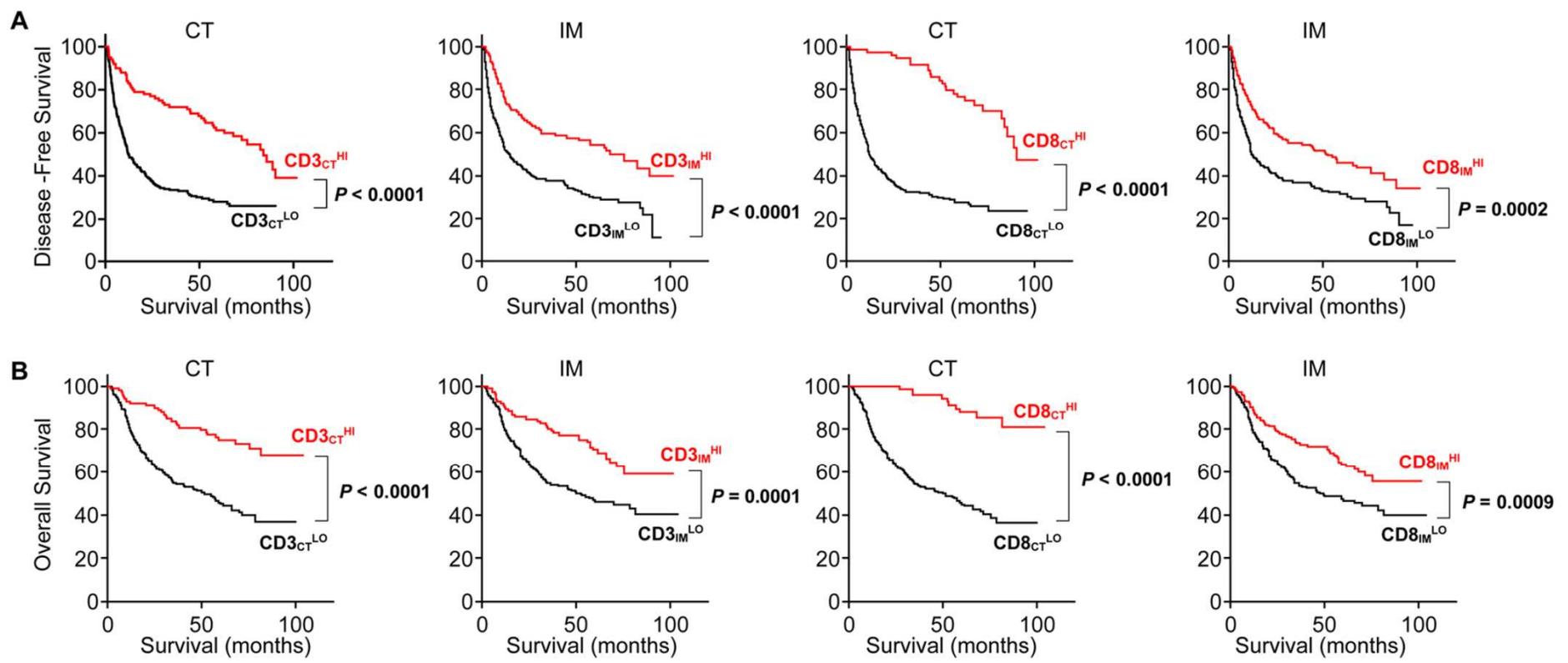
Figure 4. Increased survival time for the patients with high densities of CD3^{+} or CD8^{+} cells.

Table 1. Univariate Analysis of DFS and OS Among Patients With Liver Cancer (cohort 2) According to Clinical or Immune Parameters

			Parameters				
		DFS			OS		
Parameter	No. of pts (%)	HR	95% CI	$P*$	HR	95% CI	$P*$
Immune parameters							
$CD3_{IM}$							
(Low)	246(68.5)	1.0	(reference)		1.0	(reference)	
(High)	113(31.5)	0.5	0.37 to 0.67	<0.0001†	0.49	0.34 to 0.71	0.0002†
$CD8_{IM}$							
(Low)	206(47.4)	1.0	(reference)		1.0	(reference)	
(High)	153(42.6)	0.6	0.46 to 0.79	0.0003†	0.58	0.42 to 0.81	0.0011†
$CD3_{CT}$							
(Low)	259(72.4)	1.0	(reference)		1.0	(reference)	
(High)	100(27.6)	0.38	0.27 to 0.53		0.35	0.23 to 0.53	
$CD8_{CT}$							
(Low)	288(80.2)	1.0	(reference)		1.0	(reference)	
(High)	71(19.8)	0.23	0.15 to 0.36		0.16	0.08 to 0.31	

Result :

CD3+ or CD8+ T cells in invasive margin and centre tumour in a large Cohort 2

Part   3   Immunoscore

Which one is the most important

Table 2. Multivariate Cox Proportional Hazard Analysis for DFS and OS Among Patients With Liver Cancer From Cohort 2

Variable	DFS			OS		
	HR	95% CI	*P* *	HR	95% CI	*P* *
Before backward selection						
T stage	1.09	0.97 to 1.23	0.1676	1.17	1.01 to 1.35	0.0313†
N stage	1.57	1 to 2.48	0.0520	1.53	0.88 to 2.65	0.1312
Tumour thrombus	1.98	1.32 to 2.97	0.0010†	3.35	2.14 to 5.25	<0.0001†
$CD3_{IM}$	0.44	0.26 to 0.75	0.0022†	0.66	0.35 to 1.25	0.1979
$CD8_{IM}$	0.58	0.39 to 0.86	0.0069†	0.63	0.39 to 1.01	0.0538
$CD3_{CT}$	0.95	0.59 to 1.55	0.8407	1.18	0.66 to 2.11	0.5725
$CD8_{CT}$	0.14	0.07 to 0.28	<0.0001†	0.14	0.06 to 0.36	<0.0001†
Immune score	0.56	0.3 to 1.04	0.0671	0.87	0.41 to 1.86	0.7183
After backward selection						
T stage				1.16	1.01 to 1.33	0.0369†
N stage	1.51	0.96 to 2.37	0.0742			
Tumour thrombus	2.16	1.46 to 3.19	0.0001†	3.44	2.21 to 5.37	<0.0001†
$CD3_{IM}$	0.45	0.28 to 0.74	0.0017†			
$CD8_{IM}$	0.6	0.4 to 0.89	0.0107†	0.56	0.39 to 0.8	0.0013†
[obscured]	0.14	0.08 to 0.27	[obscured]	0.17	0.09 to 0.34	[obscured]
[obscured]	0.6	0.34 to 1.05	[obscured]			

CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC
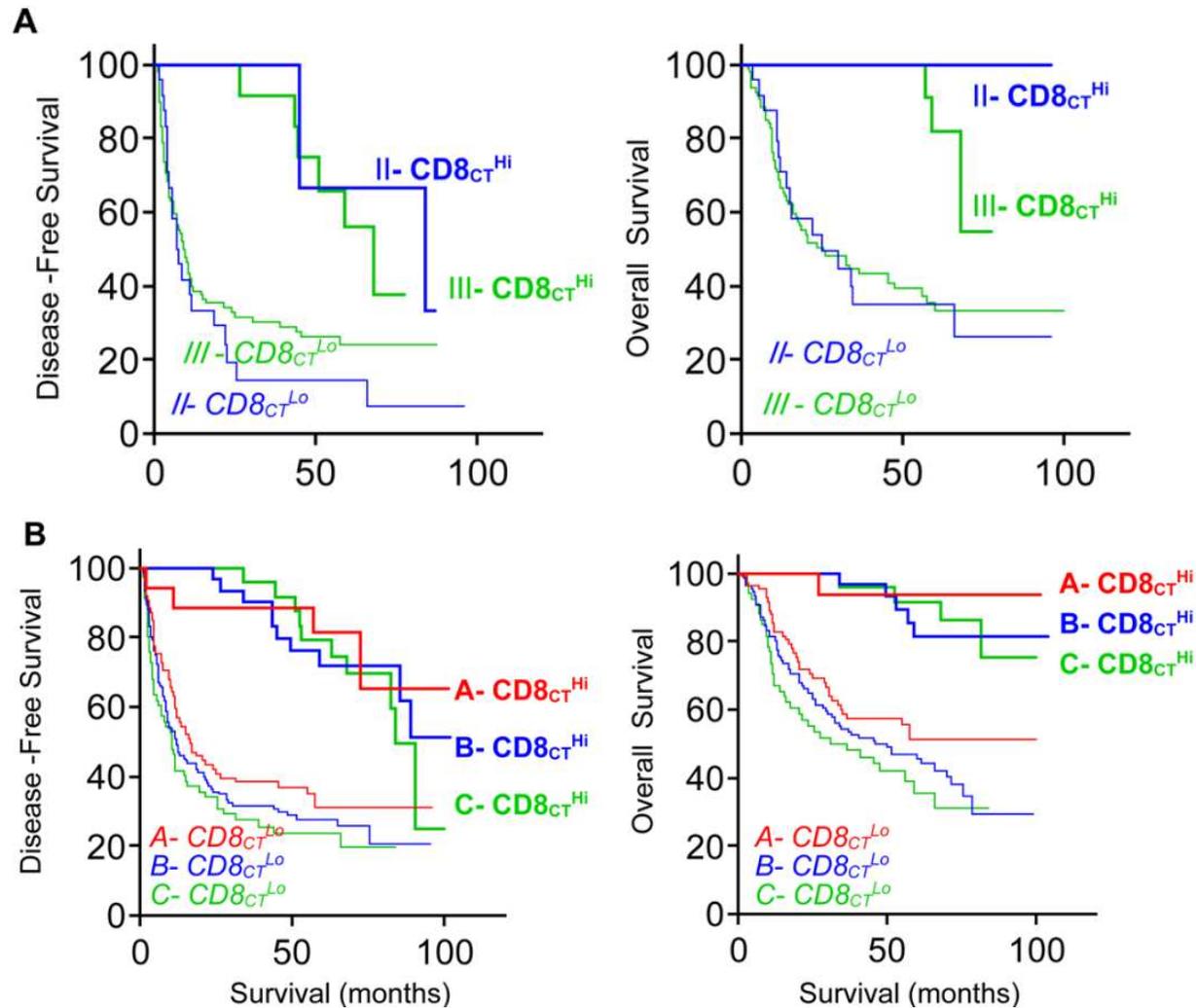

Figure 6. CD8$_{CT}$ directly predicts survival and associates with clinical characteristics of HCC.

Lessons and Take Home Messages

Key points

➢Immunoscore was highly related to the outcome of HCC patients.

➢Immunoscore seems not to be the optimal prognostic factor when compared with the $CD8_{CT}$.

Potential impact on the field

➢The density of $CD8_{CT}$ cells is an independent prognostic indicator in evaluating the prognosis in HCC patients

Lessons learned

➢Simple analysis of the density of $CD8_{CT}$ already meets the needs for survival prediction, which is economical and easy to be reproduced in clinical routines.

Acknowledgements

Thanks to Society for Immunotherapy of Cancer (SITC)!

# CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC
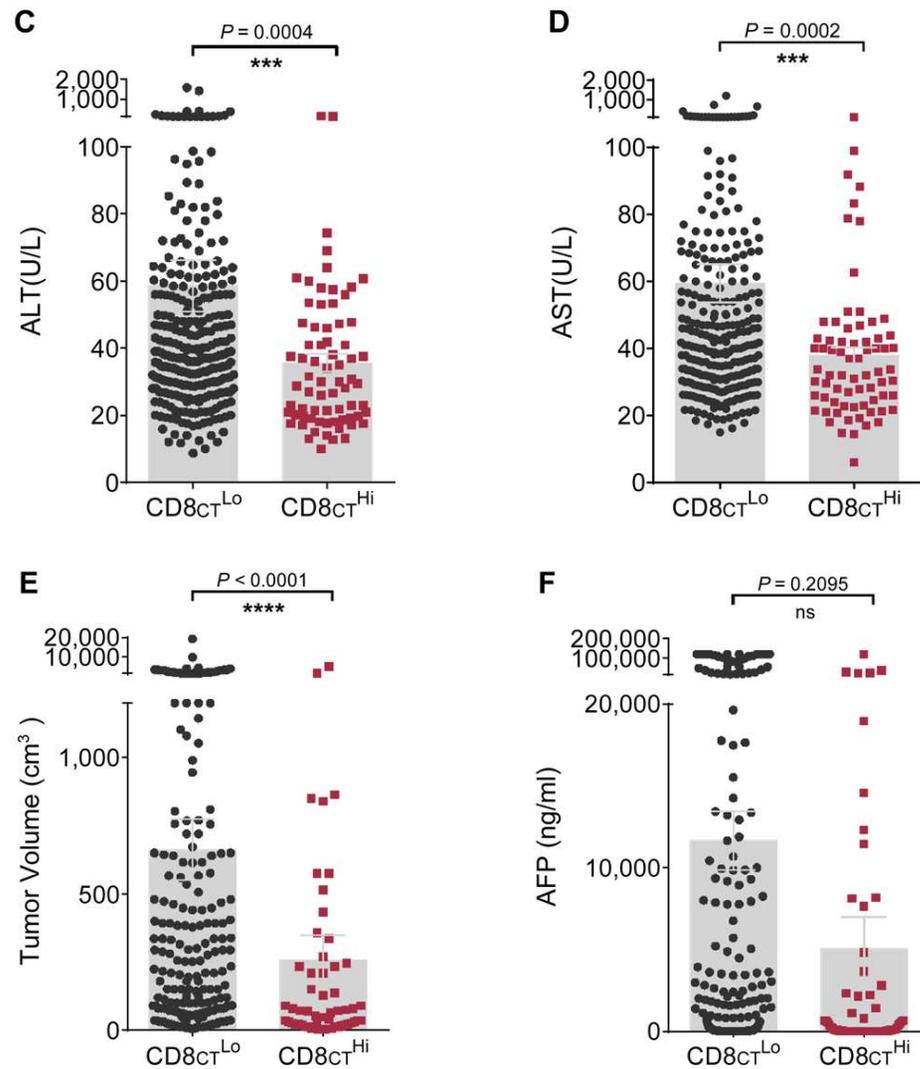
Figure 6. CD8$_{CT}$ directly predicts survival and associates with clinical characteristics of HCC.

CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC
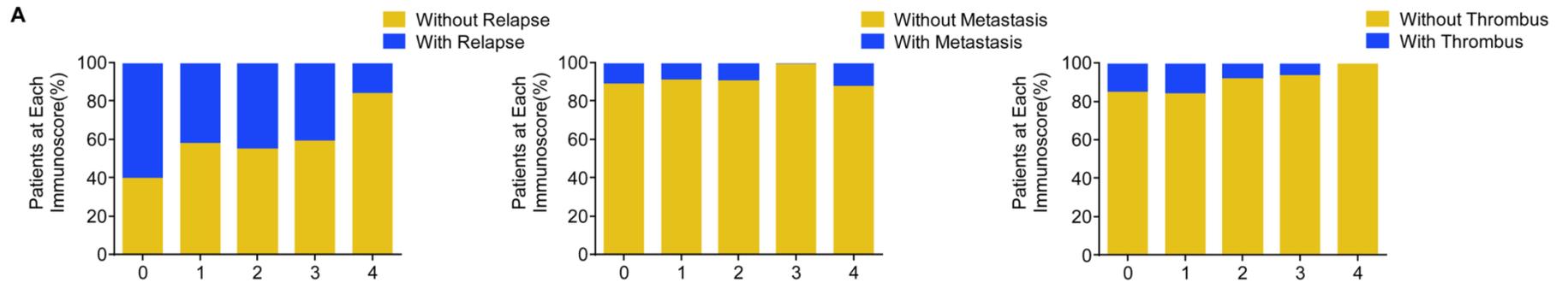

Figure S2. Clinical characteristics of HCC patients with different IS.

# CD8$_{CT}$ is a better indicator than immunoscore to predict the prognosis of HCC
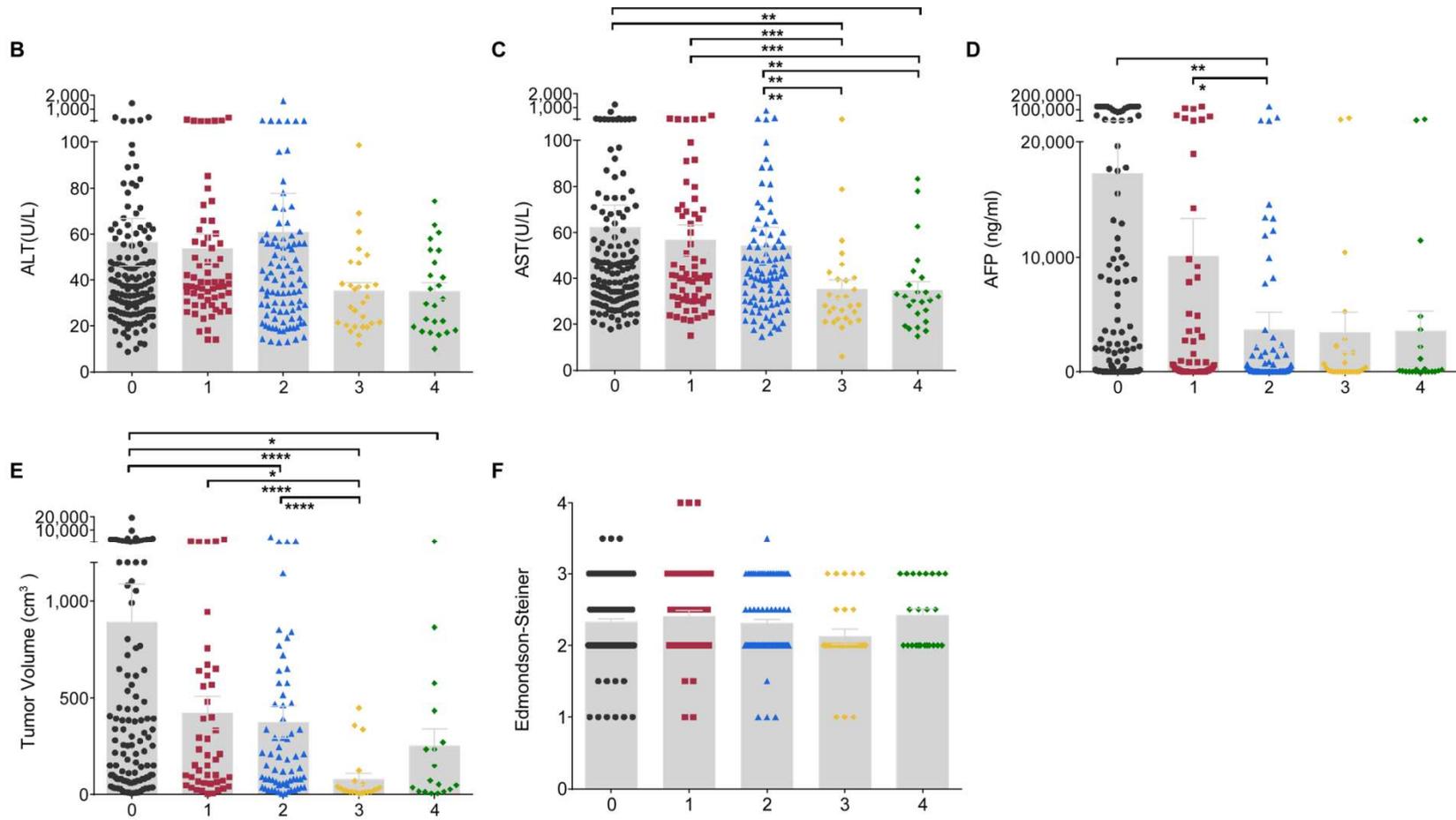
Figure S2. Clinical characteristics of HCC patients with different IS.
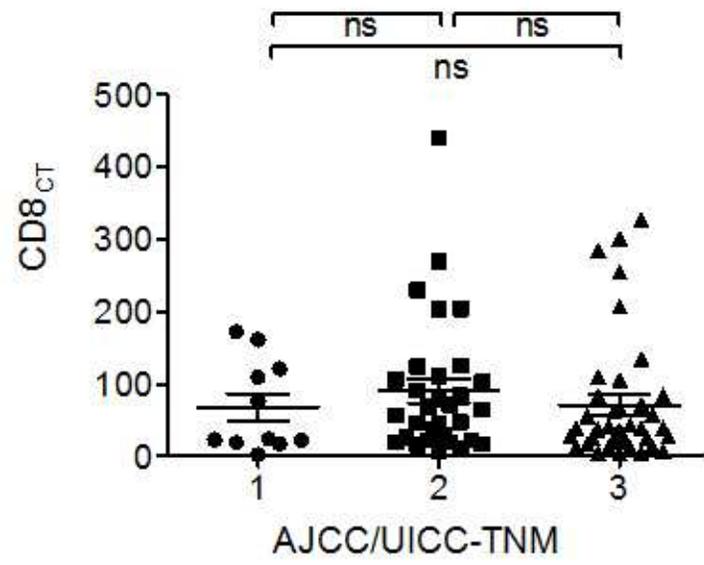
S.Table 1.　Clinicopathological characteristics in each cohort and in the total material

Group	Cohort 1	Cohort 2
Number of patients	90	359
Sex (male)	78 (86.7%)	318 (88.6%)
Age (years)	53.5 ± 10.0	50.0 ± 13.7
Time of inclusion	2007-2009	2002-2010
Last follow-up	Sep 2013	Jul 2014
Endpoints	OS	DFS, OS
OS(days)	33.2 ± 24.0	1299.7 ±1974.2
DFS(days)	----	980.0 ±1796.1
Tumor Volume(cm^3)	406.3 ± 661.1	577.8 ±3.9
Metastasis negative	84 (93.3%)	293 (81.6%)
Cirrhosis positive	33 (36.7%)	178 (49.6%)
ALT (U/L)	----	54.0 ±220.6
AST (U/L)	----	55.1 ±438.4
ALB (g/L)	----	42.6 ± 1.3
AFP (ng/ml)	----	10306.4 ± 4021.4
TBIL (μmol/L)	----	16.5 ± 0.9

Overall survival (OS); Disease Free Survival (DFS);　Alanine transaminase (ALT); Aspartate transaminase (AST).

Table 1. Univariate Analysis of DFS and OS Among Patients With Liver Cancer (cohort 2) According to Clinical or Immune Parameters

Parameter	No. of pts (%)	DFS			OS		
		HR	95% CI	P *	HR	95% CI	P *
Clinical parameters							
Gender (Male)	318(88.6)	0.9	0.6 to 1.36	0.6134	0.75	0.48 to 1.18	0.2141
Age (y)		0.93	0.81 to 1.07	0.3328	0.95	0.8 to 1.12	0.5162
<50	175(48.9)	1.0	(reference)	0.1996	1.0	(reference)	0.5675
50-60	99(27.7)	1.11	0.82 to 1.52	0.5041	1.15	0.8 to 1.65	0.4541
60-70	66(18.4)	1.05	0.74 to 1.48	0.7975	0.96	0.62 to 1.47	0.8492
70	18(11.4)	0.5	0.24 to 1.02	0.0574	0.67	0.31 to 1.46	0.3164
Metastasis (Y/N)	34(10.4)	1.62	1.06 to 2.48	0.0264†	1.49	0.89 to 2.51	0.1309
Tumour Thrombus (Y/N)	43(12.3)	2.94	2.03 to 4.26	<0.0001†	4.44	2.96 to 6.67	<0.0001†
Diameter of Tumor≤5cm	144(40.8)	1.0	(reference)		1.0	(reference)	
Diameter of Tumor >5cm	209(59.2)	1.43	1.1 to 1.86	0.0082†	1.69	1.23 to 2.31	0.0013†
HBV (Y/N)	320(91.2)	1.16	0.72 to 1.88	0.5506	0.76	0.46 to 1.26	0.2832
HCV (Y/N)	7(2.0)	1.38	0.57 to 3.36	0.4763	0.43	0.06 to 3.04	0.3938
AFP <100ng/ml	159(55.8)	1.0	(reference)		1.0	(reference)	
AFP ≥100ng/ml	126(44.2)	1.17	0.87 to 1.58	0.3059	1.43	1.00 to 2.04	0.0478

Table 1. Univariate Analysis of DFS and OS Among Patients With Liver Cancer (cohort 2) According to Clinical or Immune Parameters

Parameter	No. of pts (%)	DFS			OS		
		HR	95% CI	P *	HR	95% CI	P *
Clinical parameters							
UICC (TNM) stage		1.28	1.14 to 1.44	<0.0001†	1.33	1.16 to 1.53	<0.0001†
0-I	182(50.1)		(reference)	<0.0001†		(reference)	<0.0001†
II	27(7.4)		1.4 to 3.47	0.0007†		1.23 to 3.69	0.0067†
III	110(30.3)		1.34 to 2.45	0.0001†		1.54 to 3.1	<0.0001†
IV	44(12.1)		1.2 to 2.97	0.0060†		1.03 to 3.16	0.0399†
BCLC stage		1.1	0.92 to 1.31	0.2979	1.15	0.93 to 1.43	0.1874
A	103(28.8)	1.0	(reference)	0.5170	1.0	(reference)	0.3075
B	164(45.8)	1.18	0.86 to 1.62	0.3144	1.33	0.9 to 1.96	0.1536
C	91(25.4)	1.21	0.84 to 1.73	0.3044	1.34	0.86 to 2.08	0.1925