

Supplemental Table 3. Comparison of tumor immune microenvironment features between immune subgroups of 73 MSI-H CRCs

Variable		Immune-low (n = 15)	Immune- intermediate (n = 41)	Immune- high (n = 17)	P-value (overall)	P-value (immune- low vs. others)	P-value (immune- low vs. high)
CD3 ⁺ TIL density at IM area	High (≥ 506 cells/mm ²)	0 (0%)	19 (46%)	17 (100%)	< 0.001	< 0.001	< 0.001
	Low (< 506 cells/mm ²)	15 (100%)	22 (54%)	0 (0%)			
CD3 ⁺ TIL density at CT area	High (≥ 388 cells/mm ²)	0 (0%)	19 (46%)	17 (100%)	< 0.001	< 0.001	< 0.001
	Low (< 388 cells/mm ²)	15 (100%)	22 (54%)	0 (0%)			
CD8 ⁺ TIL density at IM area	High (≥ 249 cells/mm ²)	0 (0%)	20 (49%)	16 (94%)	< 0.001	< 0.001	< 0.001
	Low (< 249 cells/mm ²)	15 (100%)	21 (51%)	1 (6%)			
CD8 ⁺ TIL density at CT area	High (≥ 193 cells/mm ²)	0 (0%)	19 (46%)	17 (100%)	< 0.001	< 0.001	< 0.001
	Low (< 193 cells/mm ²)	15 (100%)	22 (54%)	0 (0%)			
FoxP3 ⁺ TIL density at IM area	High (≥ 49 cells/mm ²)	4 (27%)	20 (49%)	13 (76%)	0.018	0.037	0.005
	Low (< 49 cells/mm ²)	11 (73%)	21 (51%)	4 (24%)			
FoxP3 ⁺ TIL density at CT area	High (≥ 74 cells/mm ²)	3 (20%)	19 (46%)	15 (88%)	0.014	0.008	< 0.001
	Low (< 74 cells/mm ²)	12 (80%)	22 (54%)	2 (12%)			
CD68 ⁺ TAM density at IM area	High (≥ 1181 cells/mm ²)	6 (40%)	18 (44%)	13 (76%)	0.051	0.353	0.036
	Low (< 1181 cells/mm ²)	9 (60%)	23 (56%)	4 (24%)			
CD68 ⁺ TAM density at CT area	High (≥ 502 cells/mm ²)	4 (27%)	18 (44%)	15 (88%)	0.001	0.037	< 0.001
	Low (< 502 cells/mm ²)	11 (73%)	23 (56%)	2 (12%)			
CD163 ⁺ TAM density at IM area	High (≥ 923 cells/mm ²)	5 (33%)	17 (41%)	15 (88%)	0.002	0.132	0.001

	Low (< 923 cells/mm ²)	10 (67%)	24 (59%)	2 (12%)			
CD163 ⁺ TAM density at CT area	High (≥ 302 cells/mm ²)	4 (27%)	18 (44%)	15 (88%)	0.001	0.037	<0.001
	Low (< 302 cells/mm ²)	11 (73%)	23 (56%)	2 (12%)			
TLS activity	Active (maximum diameter of LAs ≥ 1 mm)	0 (0%)	29 (71%)	17 (100%)	< 0.001	< 0.001	< 0.001
	Inactive (maximum diameter of LAs < 1 mm)	15 (100%)	12 (29%)	0 (0%)			
PD-L1 expression in tumor cells	Low (H-score < 19)	14 (93%)	35 (85%)	12 (71%)	0.2	0.438	0.178
	High (H-score ≥ 19)	1 (7%)	6 (15%)	5 (29%)			
PD-L1 expression in immune cells	Low (H-score < 121)	13 (87%)	26 (63%)	3 (18%)	< 0.001	0.01	< 0.001
	High (H-score ≥ 121)	2 (13%)	15 (37%)	14 (82%)			

Abbreviations: MSI-H, microsatellite instability-high; CRCs, colorectal cancers; TIL, tumor-infiltrating lymphocyte; IM, invasive margin; CT, center of tumor; TAM, tumor-associated macrophage; TLS, tertiary lymphoid structure; LAs, lymphoid aggregates; PD-L1, programmed death-ligand 1.

Supplemental Table 4. Comparison of major molecular features between immune subgroups of 73 MSI-H CRCs

Variable		Immune-low (n = 15)	Immune- intermediate (n = 41)	Immune- high (n = 17)	P-value (overall)	P-value (immune- low vs. others)	P-value (immune- low vs. high)
MSI-H subtype ^a	Sporadic <i>MLH1</i> -methylated	5 (33%)	20 (49%)	10 (59%)	0.35	0.204	0.149
	Lynch or Lynch-like	10 (67%)	21 (51%)	7 (41%)			
CIMP	CIMP-high	3 (20%)	15 (37%)	9 (53%)	0.156	0.126	0.055
	CIMP-low/negative	12 (80%)	26 (63%)	8 (47%)			
<i>MLH1</i> promoter methylation	Methylated	5 (33%)	20 (49%)	10 (59%)	0.35	0.204	0.149
	Unmethylated	10 (67%)	21 (51%)	7 (41%)			
<i>KRAS</i> mutation	Absent	4 (27%)	31 (76%)	12 (71%)	0.003	0.001	0.013
	Present	11 (73%)	10 (24%)	5 (29%)			
<i>BRAF</i> mutation	Absent	15 (100%)	36 (92%)	15 (88%)	0.419	0.577	0.486
	Present	0 (0%)	3 (8%)	2 (12%)			
HLA class I IHC expression	Loss or reduced	2 (13%)	6 (15%)	4 (24%)	0.662	1	0.659
	Positive	13 (87%)	35 (85%)	13 (76%)			
B2M IHC expression	Loss or reduced	4 (27%)	16 (39%)	6 (35%)	0.693	0.417	0.712
	Positive	11 (73%)	25 (61%)	11 (65%)			

Abbreviations: MSI-H, microsatellite instability-high; CRCs, colorectal cancers; CIMP, CpG island methylator phenotype; HLA, human leukocyte antigen; B2M, beta-2-microglobulin; IHC, immunohistochemistry.

^aSporadic MSI-H CRC indicates a case showing *MLH1* loss with *MLH1* promoter hypermethylation, whereas Lynch or Lynch-like MSI-H CRC indicates a case showing any DNA mismatch repair (MMR) protein loss without *MLH1* promoter hypermethylation.