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SUPPLEMENTARY METHODS

Detection of Type 1 and Type 17 immunoreactive MMRp CRC samples in TCGA.

Gene expression data were obtained for 176 MSS colorectal cancer participants from the PanCancer Atlas consortium, available at the publication page (https://gdc.cancer.gov/about-data/publications/pancanatlas). Data matrices were standardized, normalized, and batch corrected using the “TCGAbiolinks” package(1). “IFNG”,”RORC”,”IL17A”,”TBX21”,”IL23R” and “IDO1” genes were categorized into “positive” and “negative” based on their expression distribution (local minimum in bimodal distribution, and median in unimodal distribution as cut off value) was used to define "**Type 1**” (Th1 signature= *IFNG+* and/or *TBX21+* and/or *IDO1+*) , " **Type 17**” (Th1/Th17 mixed signature: [*IFNG+* and/or *TBX21+* and/or *IDO1+*] + [*RORC+* and/or *IL17A+* and/or *IL23R+*]) and “Others” MSS CRC (**supplementary table 7**). The number of patients in each immune signature category was calculated and visualized in pie chart (**supplementary Fig.S10**).

References

1. Colaprico A, Silva TC, Olsen C, Garofano L, Cava C, Garolini D, et al. TCGAbiolinks: an R/Bioconductor package for integrative analysis of TCGA data. Nucleic Acids Res. 2016;44:e71.

Table S1. Demographics of patients from clinical trial NCT01876511

Arm[a]	Pt ID	Age	Gender	Race[b]	Ethnicity	Disease	Stage (On study)	Differentiation[c]	Histology	KRAS[d]	BRAF[d]	Somatic mutations
A MSI	1-001	65	Female	AA	non-Hispanic	Colorectal	IV	Poor	adenocarcinoma	Mut	WT	1291
B MSS	1-007	79	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	Mut	WT	67
B MSS	1-010	65	Female	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	Mut	118
B MSS	1-014	71	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	Mut	WT	NA
A MSI	1-015	39	Male	Asian	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	Mut	WT	2345
A MSI	1-016	51	Male	White	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	WT	WT	4025
B MSS	1-017	53	Female	White	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	Mut	WT	70
B MSS	1-018	32	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	NA
A MSI	1-019	64	Female	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	1477
B MSS	1-021	66	Female	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	77
B MSS	1-022	67	Male	AA	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	102
B MSS	1-028	39	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	Mut	WT	65
A MSI	1-031	24	Female	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	Mut	WT	941
B MSS	1-032	50	Female	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	Mut	WT	26
B MSS	1-037	47	Male	White	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	WT	WT	83
B MSS	1-038	61	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	76
B MSS	1-040	70	Male	White	non-Hispanic	Colorectal	IV	Mod	Adenocarcinoma	WT	WT	109
A MSI	1-052	47	Male	AA	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	WT	WT	478
A MSI	1-110	50	Male	White	non-Hispanic	Colorectal	IV	Poor	Adenocarcinoma	WT	Mut	NA

[a], Cohorts of the clinical trial NCT01876511: A, MSI CRC; B, MSS CRC

[b], AA, African-American

[c], Poor, poor differentiation; Mod, moderate differentiation; Well, well differentiated

[d], Mut, mutated; WT, wild type

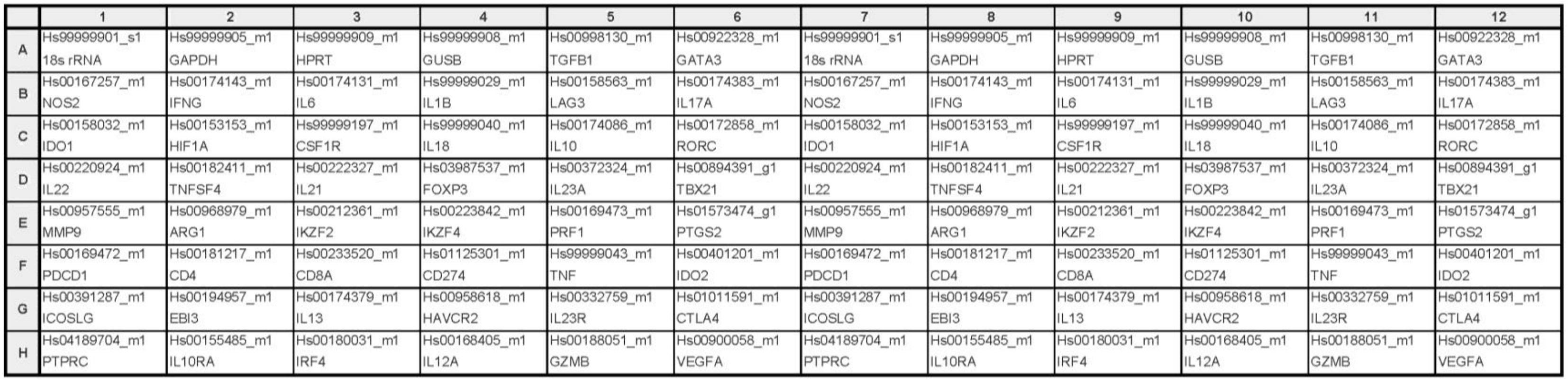
Table S2. Demographics of untreated CRC patients

MMRp CRC (n=25)								
Pt ID	**Age**	**Gender**	**Race**[a]	**Stage**	**Size (mm)**	**Histology**	**Kras**	**BRAF**
3735	64	M	W	3	70	Adenocarcinoma (low grade)	Mutated	WT
3754	72	F	AA	2	30	Adenocarcinoma (high grade)	WT	Mutated
3752	80	F	W	2	25	Adenocarcinoma (low grade)	Mutated	WT
3725	50	F	W	4	80	Adenocarcinoma (low grade)	Mutated	WT
3739	46	F	W	3	54	Adenocarcinoma (low grade)	WT	WT
3743	91	F	W	3	30	Adenocarcinoma (low grade)	Mutated	WT
3749	40	M	W	3	50	Adenocarcinoma (low grade)	WT	WT
3753	55	F	AA	4	47	Adenocarcinoma w/focal mucinous features (low grade)	WT	WT
3756	61	M	W	4	45	Adenocarcinoma (low grade)	WT	WT
3760	35	F	W	2	80	Adenocarcinoma (low grade)	WT	WT
3762	74	M	W	4	54	Adenocarcinoma (low grade)	WT	WT
3763	73	F	W	2	30	Adenocarcinoma (low grade)	WT	WT
3764	65	F	W	4	22	Adenocarcinoma (low grade)	WT	WT
3766	63	F	W	4	55	Adenocarcinoma (low grade)	WT	WT
3768	51	M	AA	4	42	Adenocarcinoma, NOS (low grade)	Mutated	WT
3785	60	F	W	3	40	Adenocarcinoma (low grade)	WT	WT
3789	61	M	O	3	50	Adenocarcinoma (low grade)	WT	WT
3972	85	M	W	3	40	Adenocarcinoma (high grade)	Mutated	WT
3977	45	F	W	1	50	Adenocarcinoma (low grade)	WT	WT
3979	83	F	AA	3	35	Adenocarcinoma (low grade)	Mutated	WT
4039	62	F	A	3	95	Adenocarcinoma (low grade)	Mutated	WT
4042	61	M	W	3	90	Adenocarcinoma (low grade)	Mutated	WT
4074	81	M	A	1	65	Adenocarcinoma (low grade)	N/A	N/A
4084	67	M	AA	2	60	Adenocarcinoma (low grade)	Mutated	N/A
4139	73	F	W	2	80	Adenocarcinoma (low grade)	N/A	N/A

MMRd CRC (n=11)								
Pt ID	**Age**	**Gender**	**Race**	**Stage**	**Size (mm)**	**Histology**	**Kras**	**BRAF**
3774	51	M	O	2	45	Adenocarcinoma w/ medullary features (high grade)	WT	WT
3976	61	F	W	1	20	Adenocarcinoma, focal mucinous features (low grade)	Mutated	WT
3982	68	M	W	2	45	Adenocarcinoma, mucinous features (low grade)	WT	WT
3987	71	F	W	2	50	Adenocarcinoma w/ mucinous features	WT	Mutated
3992	97	F	W	2	45	Adenocarcinoma (high grade)	WT	Mutated
3996	73	F	W	1	20	Adenocarcinoma (low grade)	WT	WT
3997	33	M	O	4	63	Adenocarcinoma (Moderately differentiated)	WT	WT
3998	84	F	W	2	72	Adenocarcinoma (low grade)	Mutated	WT
4089	63	M	W	1	30	Adenocarcinoma (low grade)	N/A	N/A
4095	50	M	W	1	45	Adenocarcinoma (low grade)	N/A	N/A
4100	67	F	W	2	43	Adenocarcinoma (low grade)	N/A	N/A

Table S3. Target genes and specific primer/probes gene expression assay used for Taqman-PCR.

A. taqman IR gene expression assays used for whole tumor tissue



	1	2	3	4	5	6	7	8	9	10	11	12
A	Hs99999901_s1 18s rRNA	Hs99999905_m1 GAPDH	Hs99999909_m1 HPRT	Hs99999908_m1 GUSB	Hs00998130_m1 TGFB1	Hs00922328_m1 GATA3	Hs99999901_s1 18s rRNA	Hs99999905_m1 GAPDH	Hs99999909_m1 HPRT	Hs99999908_m1 GUSB	Hs00998130_m1 TGFB1	Hs00922328_m1 GATA3
B	Hs00167257_m1 NOS2	Hs00174143_m1 IFNG	Hs00174131_m1 IL6	Hs99999029_m1 IL1B	Hs00158563_m1 LAG3	Hs00174383_m1 IL17A	Hs00167257_m1 NOS2	Hs00174143_m1 IFNG	Hs00174131_m1 IL6	Hs99999029_m1 IL1B	Hs00158563_m1 LAG3	Hs00174383_m1 IL17A
C	Hs00158032_m1 IDO1	Hs00153153_m1 HIF1A	Hs99999197_m1 CSF1R	Hs99999040_m1 IL18	Hs00174086_m1 IL10	Hs00172858_m1 RORC	Hs00158032_m1 IDO1	Hs00153153_m1 HIF1A	Hs99999197_m1 CSF1R	Hs99999040_m1 IL18	Hs00174086_m1 IL10	Hs00172858_m1 RORC
D	Hs00220924_m1 IL22	Hs00182411_m1 TNFSF4	Hs00222327_m1 IL21	Hs03987537_m1 FOXP3	Hs00372324_m1 IL23A	Hs00894391_g1 TBX21	Hs00220924_m1 IL22	Hs00182411_m1 TNFSF4	Hs00222327_m1 IL21	Hs03987537_m1 FOXP3	Hs00372324_m1 IL23A	Hs00894391_g1 TBX21
E	Hs00957555_m1 MMP9	Hs00968979_m1 ARG1	Hs00212361_m1 IKZF2	Hs00223842_m1 IKZF4	Hs00169473_m1 PRF1	Hs01573474_g1 PTGS2	Hs00957555_m1 MMP9	Hs00968979_m1 ARG1	Hs00212361_m1 IKZF2	Hs00223842_m1 IKZF4	Hs00169473_m1 PRF1	Hs01573474_g1 PTGS2
F	Hs00169472_m1 PDCD1	Hs00181217_m1 CD4	Hs00233520_m1 CD8A	Hs01125301_m1 CD274	Hs99999043_m1 TNF	Hs00401201_m1 IDO2	Hs00169472_m1 PDCD1	Hs00181217_m1 CD4	Hs00233520_m1 CD8A	Hs01125301_m1 CD274	Hs99999043_m1 TNF	Hs00401201_m1 IDO2
G	Hs00391287_m1 ICOSLG	Hs00194957_m1 EBI3	Hs00174379_m1 IL13	Hs00958618_m1 HAVCR2	Hs00332759_m1 IL23R	Hs01011591_m1 CTLA4	Hs00391287_m1 ICOSLG	Hs00194957_m1 EBI3	Hs00174379_m1 IL13	Hs00958618_m1 HAVCR2	Hs00332759_m1 IL23R	Hs01011591_m1 CTLA4
H	Hs04189704_m1 PTPRC	Hs00155485_m1 IL10RA	Hs00180031_m1 IRF4	Hs00168405_m1 IL12A	Hs00188051_m1 GZMB	Hs00900058_m1 VEGFA	Hs04189704_m1 PTPRC	Hs00155485_m1 IL10RA	Hs00180031_m1 IRF4	Hs00168405_m1 IL12A	Hs00188051_m1 GZMB	Hs00900058_m1 VEGFA

B. taqman IR gene expression assays used for cell sorted CD8+ TIL

	1	2	3	4	5	6	7	8	9	10	11	12
A	Hs99999901_s1 18s rRNA	Hs99999919_m1 CCR4	Hs01013469_m1 CCR7	Hs00386811_m1 CD27	Hs01007422_m1 CD28	Hs01002913_g1 CD40	Hs00961622_m1 IL10	Hs00155485_m1 IL10RA	Hs00989291_m1 IFNG	Hs00902334_m1 IL7R	Hs01081697_m1 IL2RB	Hs00907779_m1 IL2RA
B	Hs00174114_m1 IL2	Hs00174383_m1 IL17A	Hs01003716_m1 IL15	Hs03044418_m1 CTLA4	Hs01550088_m1 PDCD1	Hs00158563_m1 LAG3	Hs00958618_m1 HAVCR2	Hs00359999_m1 ICOS	Hs00155512_m1 TNFRSF9	Hs00921974_m1 TNFSF10	Hs00542477_m1 TNFSF14	Hs00998604_m1 TNFRSF14
C	Hs00174277_m1 TNFRSF8	Hs00174286_m1 TNFSF8	Hs00188346_m1 TNFRSF18	Hs00937194_g1 TNFRSF4	Hs00699198_m1 BTLA	Hs00203436_m1 TBX21	Hs01076122_m1 RORC	Hs00172872_m1 EOMES	Hs00231122_m1 GATA3	Hs01056533_m1 IRF4	Hs00180288_m1 CBLB	Hs00166165_m1 EGR2
D	Hs01548894_m1 CDK2	Hs01565683_g1 CDK4	Hs00176278_m1 DGKA	Hs01632414_s1 DGKZ	Hs00231780_m1 EGR3	Hs00236330_m1 FAS	Hs00174877_m1 LEP	Hs00173587_m1 LGALS3	Hs04188773_g1 LTA	Hs00542678_m1 NFATC1	Hs00905451_m1 NFATC2	Hs00190046_m1 NFATC3
E	Hs00402870_m1 HDAC9	Hs00169473_m1 PRF1	Hs01554355_m1 GZMB	Hs00174151_m1 SELL	Hs00226053_m1 RNF128	Hs00181225_m1 FASLG	Hs01062014_m1 NOTCH1	Hs00959886_g1 PTPN2	Hs00396777_m1 PPP2R2D	Hs01088027_g1 ING4	Hs00395201_m1 ITCH	Hs00235006_m1 ITGA1
F	Hs00869394_s1 ARHGAP5	Hs01058318_m1 ALK	Hs03986111_s1 TOB1	Hs00269995_m1 DTX1	Hs00171072_m1 CCL1	Hs00213192_m1 NRN1	Hs01103582_s1 JUN	Hs01083836_m1 BCL2L11	Hs01113624_g1 TNF	Hs00219575_m1 HLA-DRA	Hs00751481_s1 XCL1	Hs00175568_m1 CD244
G	Hs00705164_s1 SOCS1	Hs00219699_m1 CRTAM	Hs00199894_m1 CD160	Hs00929964_m1 KLRG1	Hs04183523_m1 PTGER2	Hs00963707_g1 GRN	Hs00246266_m1 GNLY	Hs00757930_m1 CXCL13	Hs00215132_m1 LYAR	Hs01555214_g1 TXN	Hs02330328_s1 SOCS3	Hs00183425_m1 SMAD2
H	Hs01091226_g1 DUSP2	Hs00996325_g1 KDM6B	Hs00153451_m1 E2F1	Hs00231667_m1 E2F2	Hs00939627_m1 GUSB	Hs02758991_g1 GAPDH	Hs02800695_m1 HPRT1	Hs00892674_m1 RGS16	Hs01547250_m1 LEF1	Hs01065372_g1 LAT	Hs01058407_m1 CD4	Hs00233520_m1 CD8A

Table S4. Summary statistics of Immune Related gene expression between Responder (R) and Non-Responder (NR) mCRC treated with pembrolizumab, sorted by Wilcoxon-Mann Whitney p-values (Median).

	All Patients N = 19	**Nonresponder** N = 13	**Responder** N = 6	**P-Value***
RORC				
Mean (SD)	0 (3.16)	-1.68 (1.57)	3.64 (2.6)	0.003
Median (Range)	-0.6 (-4.16, 6.28)	-0.86 (-4.16, 0.81)	3.84 (-0.47, 6.28)	**< 0.001**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL23R				
Mean (SD)	0 (2.84)	-1.07 (2.8)	2.31 (0.98)	0.001
Median (Range)	-0.2 (-5.23, 6.56)	-1.63 (-5.23, 6.56)	2.14 (1.31, 3.94)	**0.002**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
LAG3				
Mean (SD)	0 (1.82)	0.76 (1.43)	-1.65 (1.51)	0.009
Median (Range)	0.3 (-3.76, 3.93)	0.51 (-2.14, 3.93)	-1.43 (-3.76, 0.26)	**0.003**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IDO1				
Mean (SD)	0 (4.68)	1.77 (4.4)	-3.84 (2.58)	0.003
Median (Range)	-1.4 (-7.34, 9.47)	-0.16 (-3.36, 9.47)	-3.25 (-7.34, -1.43)	**0.009**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
CD4				
Mean (SD)	0 (1.17)	0.38 (1.13)	-0.83 (0.79)	0.017
Median (Range)	-0.2 (-1.52, 2.9)	0.02 (-1.1, 2.9)	-0.97 (-1.52, 0.64)	**0.017**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
CD274				
Mean (SD)	0 (4.06)	1.27 (4.28)	-2.74 (1.44)	0.008
Median (Range)	-1.2 (-4.9, 9.79)	0.05 (-3.63, 9.79)	-2.48 (-4.9, -1.24)	**0.02**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
ICOSLG				
Mean (SD)	0 (1.41)	-0.3 (1.56)	0.66 (0.78)	0.093
Median (Range)	0 (-2.33, 4.12)	-0.27 (-2.33, 4.12)	0.37 (-0.03, 1.73)	**0.029**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
VEGFA				
Mean (SD)	0 (1)	-0.33 (0.98)	0.71 (0.66)	0.017
Median (Range)	0 (-1.72, 1.8)	-0.21 (-1.72, 1.09)	0.61 (-0.06, 1.8)	**0.046**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
CD8A				
Mean (SD)	0 (1.9)	0.61 (1.62)	-1.32 (1.89)	0.06

Median (Range)	-0.1 (-4.15, 3.08)	0.36 (-1.77, 3.08)	-1.31 (-4.15, 1.28)	**0.058**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
MMP9				
Mean (SD)	0 (1.65)	0.43 (1.79)	-0.94 (0.74)	0.031
Median (Range)	-0.5 (-1.71, 4.03)	-0.28 (-1.64, 4.03)	-1.13 (-1.71, 0.43)	**0.058**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
CTLA4				
Mean (SD)	0 (1.69)	0.43 (1.68)	-0.93 (1.41)	0.094
Median (Range)	-0.2 (-2.58, 3.47)	-0.13 (-1.75, 3.47)	-1.35 (-2.58, 1.23)	**0.106**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
TNF				
Mean (SD)	0 (2.61)	0.52 (2.92)	-1.12 (1.34)	0.112
Median (Range)	-0.7 (-2.48, 9.32)	-0.41 (-1.76, 9.32)	-1.46 (-2.48, 0.67)	**0.127**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IKZF4				
Mean (SD)	0 (0.95)	-0.17 (0.88)	0.36 (1.07)	0.319
Median (Range)	-0.2 (-1.35, 1.78)	-0.45 (-1.35, 1.78)	0.33 (-1.34, 1.73)	**0.152**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL10				
Mean (SD)	0 (3.13)	0.5 (3.09)	-1.08 (3.21)	0.34
Median (Range)	-1 (-3.87, 7.25)	-0.8 (-2.2, 7.25)	-2.12 (-3.87, 4.98)	**0.152**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
PDCD1				
Mean (SD)	0 (4.21)	0.82 (4.28)	-1.77 (3.78)	0.21
Median (Range)	-1 (-5.88, 7.22)	-0.88 (-4.11, 7.22)	-2.95 (-5.88, 4.95)	**0.152**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IRF4				
Mean (SD)	0 (1.79)	0.36 (1.6)	-0.78 (2.08)	0.269
Median (Range)	-0.2 (-2.92, 3.22)	0.14 (-2.24, 3.22)	-1.46 (-2.92, 2.88)	**0.179**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
TNFSF4				
Mean (SD)	0 (4.58)	-0.3 (5.48)	0.65 (1.63)	0.573
Median (Range)	-1.1 (-5.44, 11.2)	-1.68 (-5.44, 11.2)	0.8 (-1.12, 2.44)	**0.179**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL6				
Mean (SD)	0 (4.6)	0.87 (4.61)	-1.88 (4.33)	0.236
Median (Range)	1.4 (-7.25, 5.85)	3.15 (-7.25, 5.85)	-2.51 (-6.24, 4.46)	**0.203**
No. Missing (%)	0 (0)	0 (0)	0 (0)	

PRF1				
Mean (SD)	0 (3.98)	0.9 (4.38)	-1.94 (2.06)	0.072
Median (Range)	-0.6 (-5.55, 10.53)	-0.58 (-2.98, 10.53)	-1.55 (-5.55, 0.33)	**0.21**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL23A				
Mean (SD)	0 (4.16)	-0.5 (4.32)	1.07 (3.93)	0.45
Median (Range)	-2.2 (-4.35, 8.42)	-2.31 (-4.35, 8.42)	0.25 (-2.66, 7.03)	**0.282**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IDO2				
Mean (SD)	0 (3.96)	0.77 (4.35)	-1.68 (2.46)	0.138
Median (Range)	-1.2 (-5.31, 7.36)	3.06 (-5.31, 7.36)	-1.61 (-4.76, 1.93)	**0.323**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
PTGS2				
Mean (SD)	0 (3.17)	0.55 (3.38)	-1.2 (2.5)	0.228
Median (Range)	-0.5 (-4.79, 10.38)	-0.48 (-2.75, 10.38)	-0.86 (-4.79, 1.77)	**0.368**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL17A				
Mean (SD)	0 (4.55)	0.64 (4.86)	-1.39 (3.82)	0.344
Median (Range)	-1.3 (-6.25, 7.02)	3.37 (-5.54, 7.02)	-1.46 (-6.25, 4.01)	**0.416**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL22				
Mean (SD)	0 (2.79)	0.46 (2.61)	-0.99 (3.16)	0.355
Median (Range)	0.5 (-7.22, 3.49)	0.55 (-7.22, 3.49)	-0.73 (-5.08, 3.22)	**0.456**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
ARG1				
Mean (SD)	0 (4.64)	0.8 (3.99)	-1.74 (5.84)	0.365
Median (Range)	2.3 (-8.88, 5.46)	2.54 (-6.87, 4.75)	-1.96 (-8.88, 5.46)	**0.51**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL13				
Mean (SD)	0 (3.41)	0.57 (2.94)	-1.23 (4.32)	0.386
Median (Range)	1.1 (-7.19, 4.09)	1.15 (-6.33, 4.09)	-0.13 (-7.19, 3.82)	**0.51**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
GZMB				
Mean (SD)	0 (4.07)	0.39 (4.1)	-0.85 (4.25)	0.565
Median (Range)	-0.6 (-5.8, 8.8)	-0.56 (-4.07, 8.8)	-1 (-5.8, 6.05)	**0.521**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL10RA				

Mean (SD)	0 (1.32)	0.13 (1.43)	-0.29 (1.13)	0.504
Median (Range)	-0.3 (-1.65, 3.28)	-0.25 (-1.65, 3.28)	-0.55 (-1.32, 1.87)	**0.521**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL21				
Mean (SD)	0 (2.85)	0.75 (1.54)	-1.62 (4.36)	0.248
Median (Range)	0.7 (-6.94, 3.37)	0.69 (-2.38, 3.37)	-0.51 (-6.94, 3.1)	**0.568**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
FOXP3				
Mean (SD)	0 (3.71)	-0.23 (3.72)	0.49 (4)	0.718
Median (Range)	-1 (-3.12, 10.34)	-1.01 (-3.11, 10.34)	-0.9 (-3.12, 8.07)	**0.639**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL1B				
Mean (SD)	0 (3.46)	0.38 (4.01)	-0.83 (1.86)	0.379
Median (Range)	-0.6 (-4.49, 12.27)	-0.47 (-4.49, 12.27)	-0.95 (-3.14, 1.42)	**0.639**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
NOS2				
Mean (SD)	0 (2.08)	0.24 (2.03)	-0.53 (2.27)	0.498
Median (Range)	-0.2 (-3.33, 4.87)	-0.16 (-2.56, 4.87)	-0.58 (-3.33, 2.01)	**0.639**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IFNG				
Mean (SD)	0 (5.2)	0.32 (5.47)	-0.7 (4.98)	0.693
Median (Range)	-2.4 (-6.65, 8.16)	-2.39 (-6.14, 8.16)	-1.21 (-6.65, 6.09)	**0.701**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
EBI3				
Mean (SD)	0 (3.43)	0.05 (3.39)	-0.11 (3.84)	0.935
Median (Range)	-0.9 (-3.47, 6.98)	-0.87 (-3.29, 6.84)	-0.69 (-3.47, 6.98)	**0.765**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
TGFB1				
Mean (SD)	0 (1.18)	0.1 (1.3)	-0.21 (0.91)	0.554
Median (Range)	-0.1 (-1.44, 3.32)	-0.1 (-1.44, 3.32)	-0.37 (-1.19, 0.99)	**0.765**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
CSF1R				
Mean (SD)	0 (1.48)	-0.02 (1.65)	0.03 (1.19)	0.941
Median (Range)	-0.3 (-2.65, 3.87)	-0.49 (-2.65, 3.87)	-0.18 (-1.09, 2.09)	**0.831**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
GATA3				
Mean (SD)	0 (1.82)	-0.11 (1.64)	0.23 (2.31)	0.758

Median (Range)	-0.4 (-1.96, 4.03)	-0.18 (-1.96, 3.12)	-0.85 (-1.74, 4.03)	**0.831**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
HIF1A				
Mean (SD)	0 (0.88)	0.05 (0.96)	-0.11 (0.76)	0.707
Median (Range)	-0.1 (-1.23, 1.68)	-0.09 (-1.23, 1.68)	-0.02 (-1.21, 1)	**0.966**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IKZF2				
Mean (SD)	0 (0.89)	-0.06 (0.77)	0.12 (1.17)	0.746
Median (Range)	0 (-1.15, 2.26)	0.03 (-1.1, 1.81)	-0.09 (-1.15, 2.26)	**0.966**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL12A				
Mean (SD)	0 (2.84)	-0.13 (2.81)	0.29 (3.16)	0.787
Median (Range)	-0.3 (-3.24, 7.16)	-0.33 (-3.24, 7.16)	-0.37 (-2.61, 5.76)	**>0.99**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
IL18				
Mean (SD)	0 (1.27)	0.02 (1.52)	-0.05 (0.53)	0.889
Median (Range)	-0.2 (-3.71, 2.12)	-0.04 (-3.71, 2.12)	-0.25 (-0.56, 0.87)	**>0.99**
No. Missing (%)	0 (0)	0 (0)	0 (0)	
TBX21				
Mean (SD)	0 (4.73)	0.09 (4.47)	-0.2 (5.69)	0.915
Median (Range)	-2.5 (-4.24, 11.27)	-2.56 (-4.24, 8.52)	-2.44 (-3.73, 11.27)	**>0.99**
No. Missing (%)	0 (0)	0 (0)	0 (0)	

*P-value based on t-test and Wilcoxon-Mann Whitney test

Table S5. Significant p values ($p<0.05$) of gene expression between newly defined R-type and NR-type MMRp CRC specimens.

	Non-parametric multiple t-test primary R- versus NR-type MMRp CRC[a]						
Genes	**P value**	**Mean R MMRp (n=3)**	**Mean NR MMRp (n=11)**	**Difference**	**SE of difference**	**t ratio**	**df**
LAG3	0.00013952	0.0524318	0.00836918	0.0440626	0.00803292	5.48526	12
CD8A	0.000306537	1.37435	0.148175	1.22618	0.244972	5.00538	12
PTPRC	0.000591193	8.63849	1.44078	7.19771	1.31474	5.47464	8
FOXP3	0.000880331	0.0986468	0.0225226	0.0761241	0.0173394	4.39024	12
CD274	0.00169571	0.0191249	0.00478136	0.0143435	0.00356695	4.02123	12
CTLA4	0.00462478	0.375785	0.0633476	0.312438	0.0900208	3.47072	12
CD4	0.00761172	2.09297	0.368644	1.72433	0.538604	3.20147	12
IRF4	0.00847909	0.129499	0.0239186	0.10558	0.0335884	3.14334	12
IL10RA	0.00860155	1.12714	0.302912	0.824224	0.262858	3.13562	12
GATA3	0.0202629	0.122791	0.0365723	0.0862189	0.0322442	2.67393	12
IDO1	0.0219489	0.22015	0.00997234	0.210178	0.0798955	2.63066	12
IKZF2	0.0232372	0.107724	0.0257105	0.0820137	0.0315469	2.59974	12
PRF1	0.0277546	0.0998818	0.0205513	0.0793305	0.0316923	2.50315	12
HIF1A	0.0432477	5.39949	2.01431	3.38519	1.49818	2.25953	12
EBI3	0.0440331	0.0101568	0.00325257	0.00690419	0.00306915	2.24955	12
TGFB1	0.0483204	1.22629	0.374702	0.851589	0.38746	2.19788	12

[a] no correction for multiple comparisons

▨ T cell checkpoints; 🟩 T cell infiltration; 🟨 T reg

Table S6. Summary statistics for paired differences of gene expression between cell sorted CD8$^+$ PD1^{hi} and CD8$^+$ PD1^{lo} TILs of resected primary CRC. Gene expression in PD1hi and PD1lo were relative to expression in PD1^{neg} CD8$^+$ TILs (ΔΔCt).

	PD1 Low - PD1 High N = 6	**P-Value***
ARHGAP5		
Mean (SD)	0.87 (1.31)	0.164
Median (Range)	0.5 (-0.75, 2.7)	**0.219**
No. Missing (%)	0 (0)	
BCL2L11		
Mean (SD)	0.28 (1.02)	0.531
Median (Range)	-0.1 (-0.57, 2.27)	**>0.99**
No. Missing (%)	0 (0)	
BTLA		
Mean (SD)	-21.56 (51.94)	0.356
Median (Range)	-0.4 (-127.58, 0.4)	**0.438**
No. Missing (%)	0 (0)	
CBLB		
Mean (SD)	0.04 (0.37)	0.8
Median (Range)	0.2 (-0.51, 0.46)	**>0.99**
No. Missing (%)	0 (0)	
CCR4		
Mean (SD)	-0.64 (1.17)	0.236
Median (Range)	-0.4 (-2.41, 0.53)	**0.438**
No. Missing (%)	0 (0)	
CCR7		
Mean (SD)	0.18 (0.27)	0.16
Median (Range)	0.1 (-0.08, 0.69)	**0.156**
No. Missing (%)	0 (0)	
CD160		
Mean (SD)	0.14 (0.49)	0.509
Median (Range)	0 (-0.37, 1.03)	**>0.99**
No. Missing (%)	0 (0)	
CD244		
Mean (SD)	-0.58 (1.3)	0.321
Median (Range)	-0.2 (-2.49, 0.75)	**0.563**
No. Missing (%)	0 (0)	

CD27		
Mean (SD)	-1.79 (2.81)	0.179
Median (Range)	-0.8 (-6.18, 0.98)	**0.438**
No. Missing (%)	0 (0)	
CD28		
Mean (SD)	-0.26 (1.02)	0.563
Median (Range)	0.2 (-1.62, 0.72)	**>0.99**
No. Missing (%)	0 (0)	
CD8A		
Mean (SD)	0.03 (0.3)	0.794
Median (Range)	0 (-0.36, 0.57)	**>0.99**
No. Missing (%)	0 (0)	
CDK2		
Mean (SD)	-0.06 (0.69)	0.827
Median (Range)	-0.2 (-0.69, 1.27)	**0.438**
No. Missing (%)	0 (0)	
CDK4		
Mean (SD)	-0.28 (0.52)	0.25
Median (Range)	-0.4 (-0.84, 0.66)	**0.313**
No. Missing (%)	0 (0)	
CRTAM		
Mean (SD)	-0.26 (1.26)	0.64
Median (Range)	-0.7 (-1.3, 2.14)	**0.438**
No. Missing (%)	0 (0)	
CTLA4		
Mean (SD)	-15.89 (34.77)	0.314
Median (Range)	-2.2 (-86.78, 0.54)	**0.094**
No. Missing (%)	0 (0)	
CXCL13		
Mean (SD)	-35405.57 (76899.48)	0.311
Median (Range)	-838.8 (-191625.87, -0.3)	**0.031**
No. Missing (%)	0 (0)	
DGKA		
Mean (SD)	-0.08 (0.51)	0.715
Median (Range)	0 (-0.77, 0.59)	**>0.99**
No. Missing (%)	0 (0)	
DGKZ		
Mean (SD)	-0.38 (0.71)	0.25

Median (Range)	-0.4 (-1.45, 0.64)	**0.438**
No. Missing (%)	0 (0)	
DTX1		
Mean (SD)	0.11 (0.42)	0.547
Median (Range)	0 (-0.41, 0.71)	**0.688**
No. Missing (%)	0 (0)	
DUSP2		
Mean (SD)	0.09 (0.45)	0.651
Median (Range)	0.2 (-0.73, 0.51)	**0.563**
No. Missing (%)	0 (0)	
E2F1		
Mean (SD)	-1.02 (1.32)	0.118
Median (Range)	-0.8 (-3.35, 0.53)	**0.094**
No. Missing (%)	0 (0)	
E2F2		
Mean (SD)	-240.24 (637.01)	0.398
Median (Range)	-0.3 (-1537.94, 99.65)	**0.59**
No. Missing (%)	0 (0)	
EGR2		
Mean (SD)	0.18 (0.55)	0.454
Median (Range)	0.2 (-0.56, 0.91)	**0.563**
No. Missing (%)	0 (0)	
EGR3		
Mean (SD)	0.35 (0.38)	0.074
Median (Range)	0.2 (0, 0.88)	**0.063**
No. Missing (%)	0 (0)	
EOMES		
Mean (SD)	-4.97 (12.31)	0.368
Median (Range)	-0.5 (-29.72, 3.7)	**0.438**
No. Missing (%)	0 (0)	
FAS		
Mean (SD)	0.15 (0.71)	0.633
Median (Range)	0.1 (-0.67, 1.42)	**0.688**
No. Missing (%)	0 (0)	
FASLG		
Mean (SD)	-0.83 (1.14)	0.133
Median (Range)	-0.4 (-2.96, 0.18)	**0.094**
No. Missing (%)	0 (0)	

GATA3		
Mean (SD)	0.12 (0.31)	0.373
Median (Range)	0.2 (-0.24, 0.59)	**0.563**
No. Missing (%)	0 (0)	
GNLY		
Mean (SD)	-0.09 (0.59)	0.736
Median (Range)	0.1 (-0.96, 0.69)	**>0.99**
No. Missing (%)	0 (0)	
GRN		
Mean (SD)	-1.13 (2.32)	0.285
Median (Range)	-0.4 (-5.79, 0.32)	**0.156**
No. Missing (%)	0 (0)	
GZMB		
Mean (SD)	-1.43 (0.85)	0.009
Median (Range)	-1.6 (-2.35, -0.37)	**0.031**
No. Missing (%)	0 (0)	
HAVCR2		
Mean (SD)	-4.28 (6.76)	0.182
Median (Range)	-2.3 (-14.11, 3.46)	**0.219**
No. Missing (%)	0 (0)	
HDAC9		
Mean (SD)	-5.55 (38.04)	0.735
Median (Range)	0.2 (-75.42, 41.61)	**>0.99**
No. Missing (%)	0 (0)	
HLA-DRA		
Mean (SD)	-2.18 (4.66)	0.303
Median (Range)	-0.5 (-8.32, 3.62)	**0.219**
No. Missing (%)	0 (0)	
HPRT1		
Mean (SD)	-0.06 (0.37)	0.708
Median (Range)	-0.1 (-0.6, 0.55)	**0.438**
No. Missing (%)	0 (0)	
ICOS		
Mean (SD)	-1.06 (2.08)	0.265
Median (Range)	-0.2 (-4.96, 0.59)	**0.563**
No. Missing (%)	0 (0)	
IFNG		
Mean (SD)	-0.24 (0.62)	0.387
Median (Range)	0 (-1.16, 0.35)	**0.688**

No. Missing (%)	0 (0)	
IL10		
Mean (SD)	-33.18 (60.44)	0.237
Median (Range)	-1.5 (-148.63, 4.11)	**0.563**
No. Missing (%)	0 (0)	
IL10RA		
Mean (SD)	0.15 (0.36)	0.36
Median (Range)	0.1 (-0.29, 0.61)	**0.563**
No. Missing (%)	0 (0)	
IL15		
Mean (SD)	0.1 (0.17)	0.223
Median (Range)	0.1 (-0.19, 0.3)	**0.219**
No. Missing (%)	0 (0)	
IL17A		
Mean (SD)	1.76 (17.76)	0.818
Median (Range)	-2.3 (-15.22, 36.28)	**0.59**
No. Missing (%)	0 (0)	
IL2		
Mean (SD)	0.1 (0.16)	0.191
Median (Range)	0.1 (-0.05, 0.41)	**0.219**
No. Missing (%)	0 (0)	
IL2RA		
Mean (SD)	0.51 (0.45)	0.038
Median (Range)	0.5 (-0.07, 1.24)	**0.063**
No. Missing (%)	0 (0)	
IL2RB		
Mean (SD)	0.1 (0.26)	0.408
Median (Range)	0.1 (-0.28, 0.48)	**0.438**
No. Missing (%)	0 (0)	
IL7R		
Mean (SD)	0.23 (0.33)	0.142
Median (Range)	0.1 (-0.07, 0.77)	**0.313**
No. Missing (%)	0 (0)	
ING4		
Mean (SD)	0.2 (0.95)	0.628
Median (Range)	-0.1 (-0.6, 1.95)	**>0.99**
No. Missing (%)	0 (0)	
IRF4		

Mean (SD)	0.16 (0.33)	0.3
Median (Range)	0.2 (-0.36, 0.58)	**0.313**
No. Missing (%)	0 (0)	
ITCH		
Mean (SD)	0 (0.28)	0.999
Median (Range)	0 (-0.39, 0.29)	**>0.99**
No. Missing (%)	0 (0)	
ITGA1		
Mean (SD)	0.15 (0.25)	0.183
Median (Range)	0.1 (-0.21, 0.45)	**0.219**
No. Missing (%)	0 (0)	
JUN		
Mean (SD)	0.13 (0.31)	0.371
Median (Range)	0 (-0.15, 0.66)	**0.844**
No. Missing (%)	0 (0)	
KDM6B		
Mean (SD)	0.37 (0.34)	0.043
Median (Range)	0.3 (0.01, 0.95)	**0.031**
No. Missing (%)	0 (0)	
KLRG1		
Mean (SD)	-0.16 (1.3)	0.774
Median (Range)	0.2 (-2.74, 0.96)	**0.438**
No. Missing (%)	0 (0)	
LAG3		
Mean (SD)	-1.04 (1.01)	0.054
Median (Range)	-1.2 (-2.08, 0.59)	**0.094**
No. Missing (%)	0 (0)	
LAT		
Mean (SD)	-0.26 (0.67)	0.393
Median (Range)	0 (-1.52, 0.36)	**0.563**
No. Missing (%)	0 (0)	
LEF1		
Mean (SD)	0.11 (0.39)	0.511
Median (Range)	0.1 (-0.51, 0.67)	**0.313**
No. Missing (%)	0 (0)	
LGALS3		
Mean (SD)	0.02 (0.44)	0.898
Median (Range)	0.1 (-0.57, 0.7)	**0.844**
No. Missing (%)	0 (0)	

LTA		
Mean (SD)	-0.09 (1.03)	0.842
Median (Range)	0 (-1.75, 1.41)	**>0.99**
No. Missing (%)	0 (0)	
LYAR		
Mean (SD)	-0.05 (0.43)	0.804
Median (Range)	-0.1 (-0.53, 0.72)	**0.688**
No. Missing (%)	0 (0)	
NFATC1		
Mean (SD)	-0.06 (0.61)	0.831
Median (Range)	0 (-0.86, 0.66)	**>0.99**
No. Missing (%)	0 (0)	
NFATC2		
Mean (SD)	0.01 (0.79)	0.977
Median (Range)	0 (-1.2, 1.13)	**>0.99**
No. Missing (%)	0 (0)	
NFATC3		
Mean (SD)	0.02 (0.23)	0.816
Median (Range)	0 (-0.26, 0.39)	**>0.99**
No. Missing (%)	0 (0)	
NOTCH1		
Mean (SD)	0.14 (1)	0.753
Median (Range)	-0.2 (-0.73, 2.1)	**0.688**
No. Missing (%)	0 (0)	
PDCD1		
Mean (SD)	-6.57 (8.21)	0.107
Median (Range)	-3.4 (-23.11, -1.96)	**0.031**
No. Missing (%)	0 (0)	
PPP2R2D		
Mean (SD)	0.08 (0.46)	0.687
Median (Range)	0.1 (-0.69, 0.62)	**0.688**
No. Missing (%)	0 (0)	
PRF1		
Mean (SD)	-0.4 (0.65)	0.198
Median (Range)	-0.4 (-1.08, 0.51)	**0.219**
No. Missing (%)	0 (0)	
PTGER2		
Mean (SD)	0.27 (0.3)	0.075

Median (Range)	0.1 (0.05, 0.66)	**0.031**
No. Missing (%)	0 (0)	
PTPN2		
Mean (SD)	0.09 (0.39)	0.575
Median (Range)	0.1 (-0.53, 0.61)	**0.438**
No. Missing (%)	0 (0)	
RGS16		
Mean (SD)	0.35 (0.63)	0.229
Median (Range)	0.4 (-0.58, 1.07)	**0.219**
No. Missing (%)	0 (0)	
RORC		
Mean (SD)	-0.36 (2.13)	0.696
Median (Range)	0.1 (-4.38, 2.02)	**>0.99**
No. Missing (%)	0 (0)	
SELL		
Mean (SD)	0.16 (0.28)	0.227
Median (Range)	0.1 (-0.16, 0.56)	**0.563**
No. Missing (%)	0 (0)	
SMAD2		
Mean (SD)	0.2 (0.38)	0.257
Median (Range)	0.2 (-0.22, 0.75)	**0.563**
No. Missing (%)	0 (0)	
SOCS1		
Mean (SD)	0.2 (0.45)	0.325
Median (Range)	0.1 (-0.3, 1.02)	**0.313**
No. Missing (%)	0 (0)	
SOCS3		
Mean (SD)	0.05 (0.31)	0.72
Median (Range)	0.1 (-0.29, 0.47)	**>0.99**
No. Missing (%)	0 (0)	
TBX21		
Mean (SD)	0.1 (0.58)	0.693
Median (Range)	0 (-0.61, 0.86)	**0.844**
No. Missing (%)	0 (0)	
TNF		
Mean (SD)	0.21 (0.48)	0.324
Median (Range)	0 (-0.22, 0.84)	**0.688**
No. Missing (%)	0 (0)	

TNFRSF14		
Mean (SD)	0.03 (0.35)	0.84
Median (Range)	0.1 (-0.48, 0.43)	**>0.99**
No. Missing (%)	0 (0)	
TNFRSF18		
Mean (SD)	-1.3 (2.4)	0.243
Median (Range)	-0.4 (-4.99, 1.26)	**0.438**
No. Missing (%)	0 (0)	
TNFRSF4		
Mean (SD)	-184.62 (390.98)	0.3
Median (Range)	0.1 (-974.91, 2.01)	**>0.99**
No. Missing (%)	0 (0)	
TNFRSF8		
Mean (SD)	10.21 (22.69)	0.321
Median (Range)	0.8 (0, 56.46)	**0.059**
No. Missing (%)	0 (0)	
TNFRSF9		
Mean (SD)	-0.58 (1.3)	0.323
Median (Range)	-0.2 (-2.46, 1.12)	**0.313**
No. Missing (%)	0 (0)	
TNFSF10		
Mean (SD)	-0.39 (0.87)	0.33
Median (Range)	-0.6 (-1.28, 1.2)	**0.313**
No. Missing (%)	0 (0)	
TNFSF14		
Mean (SD)	0.08 (0.7)	0.778
Median (Range)	0.2 (-1.18, 0.67)	**0.688**
No. Missing (%)	0 (0)	
TNFSF8		
Mean (SD)	-1.81 (3.19)	0.223
Median (Range)	-0.2 (-8.1, -0.06)	**0.031**
No. Missing (%)	0 (0)	
TOB1		
Mean (SD)	-0.09 (0.53)	0.707
Median (Range)	0 (-1.05, 0.42)	**>0.99**
No. Missing (%)	0 (0)	
TXN		
Mean (SD)	0.08 (0.44)	0.67
Median (Range)	0.1 (-0.48, 0.62)	**0.688**

No. Missing (%)	0 (0)	
XCL1		
Mean (SD)	227.11 (553.15)	0.361
Median (Range)	0.8 (-1.1, 1356.2)	**0.438**
No. Missing (%)	0 (0)	

*P-value is based on paired T-test and Wilcoxon signed Rank Test.

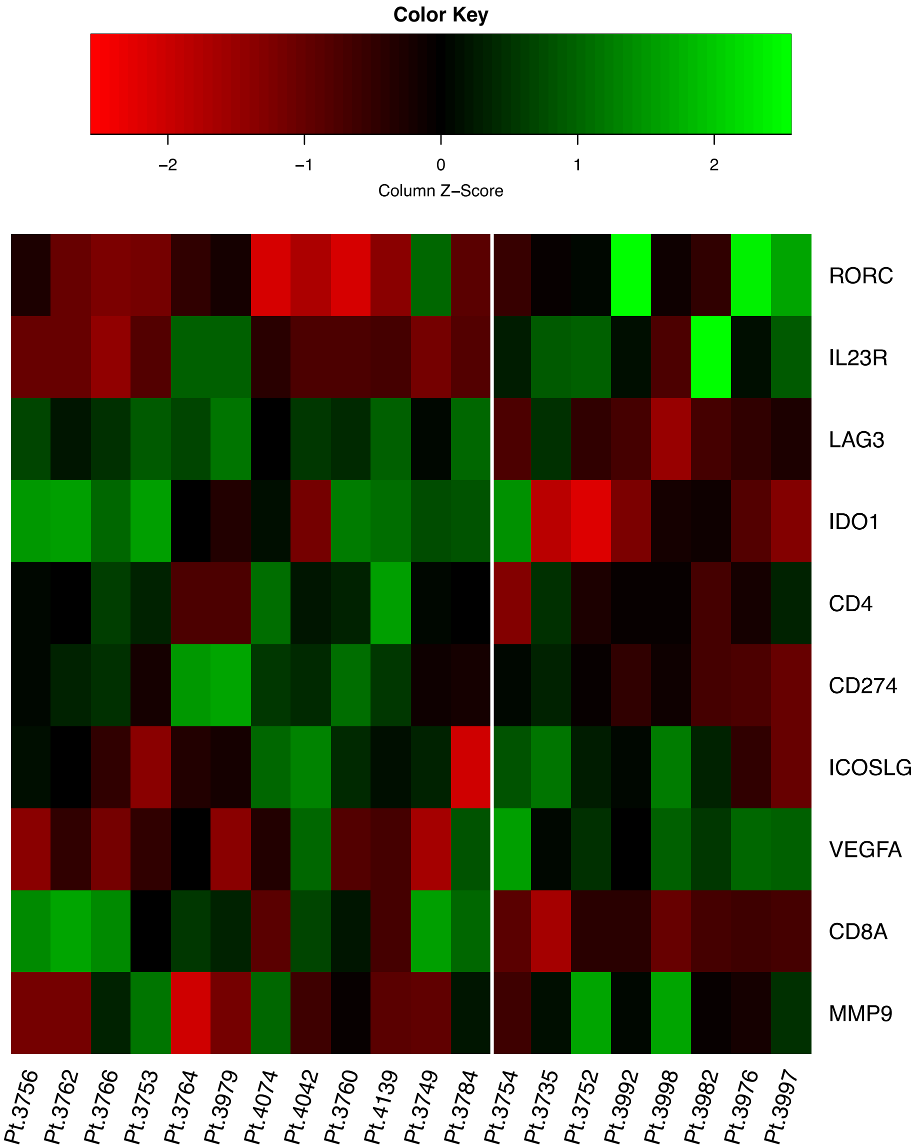
Supplementary Table S7. Th1 and mixed Th1/Th17 immune signatures in MSS samples extracted from TCGA database

Gene marker pattern	Number of patients	Proportion	Immune signature group
IFNG+IDO1-TBX21-IL23R-RORC-IL17A+	16	0.090909091	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R+RORC+IL17A+	16	0.090909091	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R-RORC+IL17A-	6	0.034090909	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R+RORC-IL17A+	5	0.028409091	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R-RORC+IL17A+	4	0.022727273	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R-RORC-IL17A+	4	0.022727273	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R+RORC+IL17A-	3	0.017045455	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R+RORC+IL17A+	3	0.017045455	Th1/Th17 mixed signature
IFNG+IDO1-TBX21-IL23R+RORC-IL17A-	2	0.011363636	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R-RORC+IL17A+	2	0.011363636	Th1/Th17 mixed signature
IFNG+IDO1+TBX21+IL23R+RORC-IL17A+	2	0.011363636	Th1/Th17 mixed signature
IFNG+IDO1+TBX21+IL23R+RORC+IL17A-	2	0.011363636	Th1/Th17 mixed signature
IFNG-IDO1-TBX21+IL23R+RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG-IDO1+TBX21-IL23R-RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG-IDO1+TBX21-IL23R+RORC+IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1-TBX21+IL23R-RORC-IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1-TBX21+IL23R-RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1-TBX21+IL23R-RORC+IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1-TBX21+IL23R+RORC+IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R-RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R+RORC-IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R+RORC-IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R+RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21+IL23R-RORC-IL17A+	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21+IL23R-RORC+IL17A-	1	0.005681818	Th1/Th17 mixed signature
IFNG+IDO1+TBX21-IL23R-RORC-IL17A-	5	0.028409091	Th1 signature
IFNG+IDO1-TBX21-IL23R-RORC-IL17A-	4	0.022727273	Th1 signature
IFNG+IDO1-TBX21+IL23R-RORC-IL17A-	3	0.017045455	Th1 signature
IFNG+IDO1+TBX21+IL23R-RORC-IL17A-	3	0.017045455	Th1 signature
IFNG-IDO1-TBX21+IL23R-RORC-IL17A-	1	0.005681818	Th1 signature
IFNG-IDO1+TBX21-IL23R-RORC-IL17A-	1	0.005681818	Th1 signature
IFNG-IDO1-TBX21-IL23R-RORC-IL17A-	16	0.090909091	Others
IFNG-IDO1-TBX21-IL23R-RORC+IL17A-	13	0.073863636	Others
IFNG-IDO1-TBX21-IL23R-RORC+IL17A+	12	0.068181818	Others
IFNG-IDO1-TBX21-IL23R+RORC-IL17A-	11	0.0625	Others
IFNG-IDO1-TBX21-IL23R+RORC+IL17A-	9	0.051136364	Others
IFNG-IDO1-TBX21-IL23R+RORC+IL17A+	9	0.051136364	Others
IFNG-IDO1-TBX21-IL23R+RORC-IL17A+	6	0.034090909	Others
IFNG-IDO1-TBX21-IL23R-RORC-IL17A+	5	0.028409091	Others

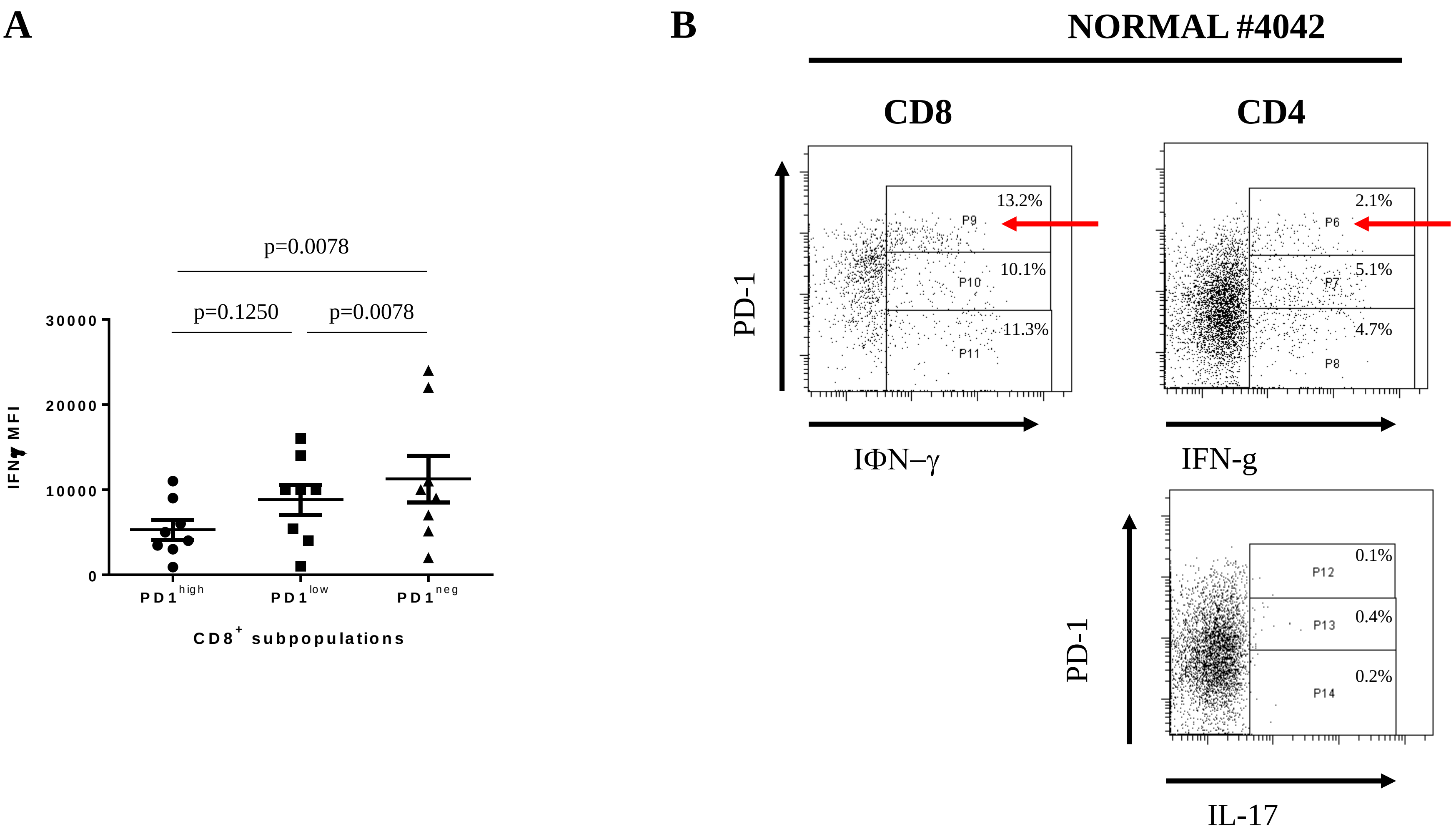
A Gaussian Mixture Model Training (Trial Patients)

Patients	Response Status	Predicted Response Status
1	Nonresponder	Responder
10	Nonresponder	Nonresponder
22	Nonresponder	Nonresponder
40	Nonresponder	Nonresponder
14	Nonresponder	Nonresponder
17	Nonresponder	Nonresponder
18	Nonresponder	Nonresponder
21	Nonresponder	Nonresponder
28	Nonresponder	Nonresponder
32	Nonresponder	Nonresponder
37	Nonresponder	Nonresponder
38	Nonresponder	Nonresponder
7	Nonresponder	Nonresponder
110	Responder	Responder
16	Responder	Responder
19	Responder	Responder
52	Responder	Responder
31	Responder	Nonresponder
15	Responder	Nonresponder

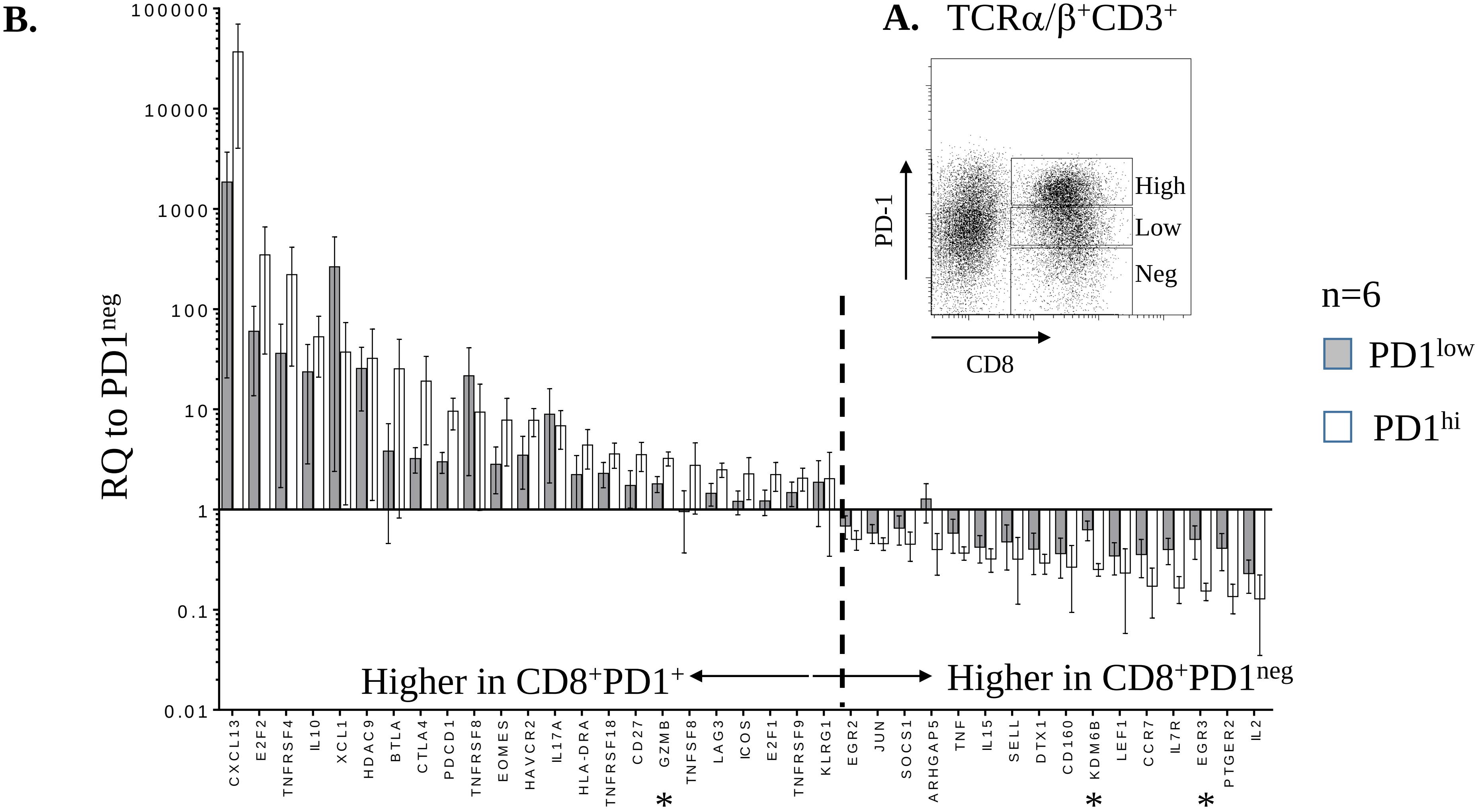
B Heat map of Gene Expression Levels (Primary CRC Cohort)



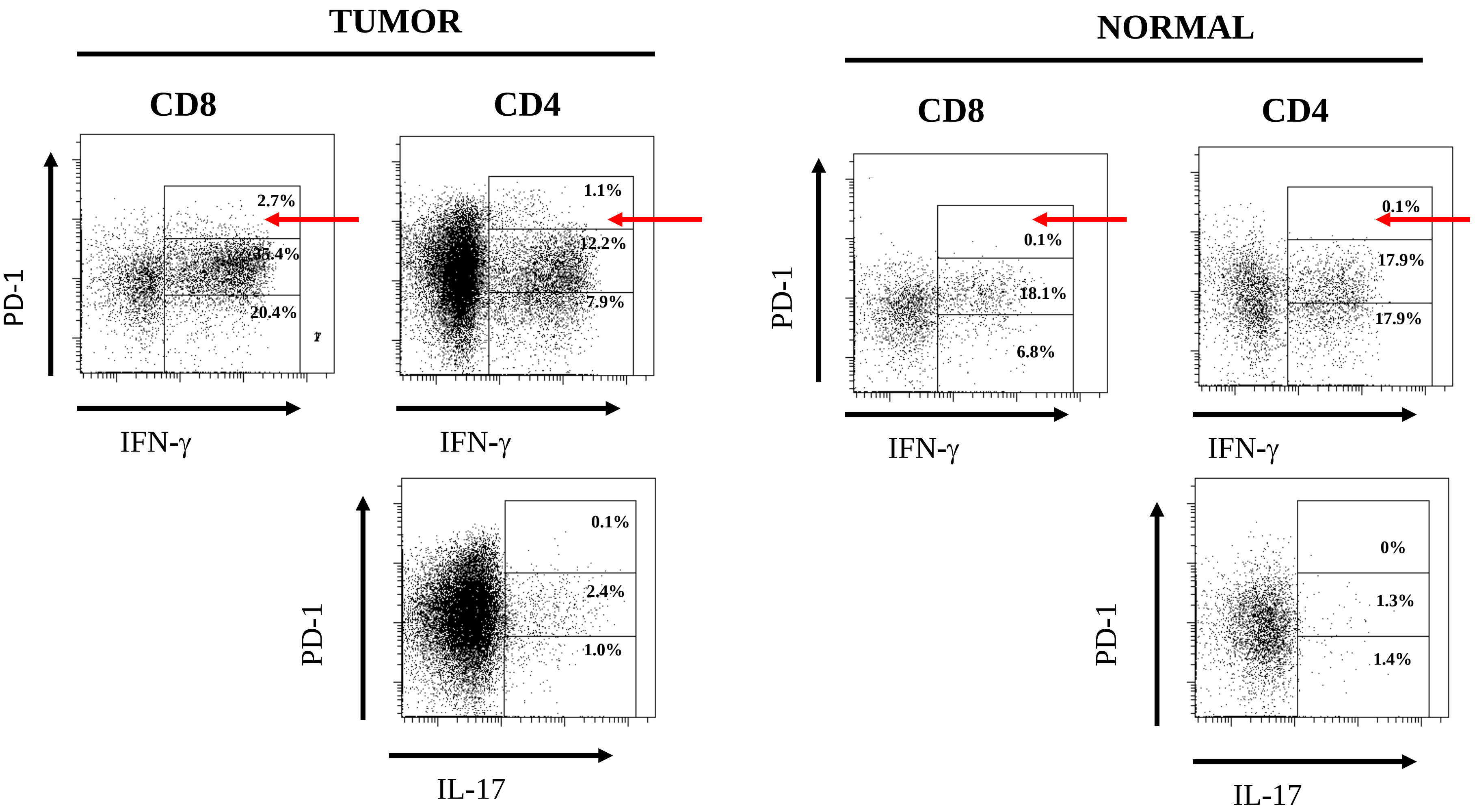
Supplementary figure S1. Model of T*i*ME in primary colon tumor of mCRC patients according to their clinical response to pembrolizumab. A, Gaussian mixture model using the top 10 genes based on p-values were used to model the response status (Responder / Nonresponder) of those patients in the anti-PD1 (pembrolizumab) trial cohort. The model predictions for this training dataset are shown. **B**, Heat map of the gene expression levels of the top 10 genes for patients in the primary CRC cohort (scaling is done within a patient). Patients to the right of the white line are predicted responders; those to the left are predicted nonresponders.



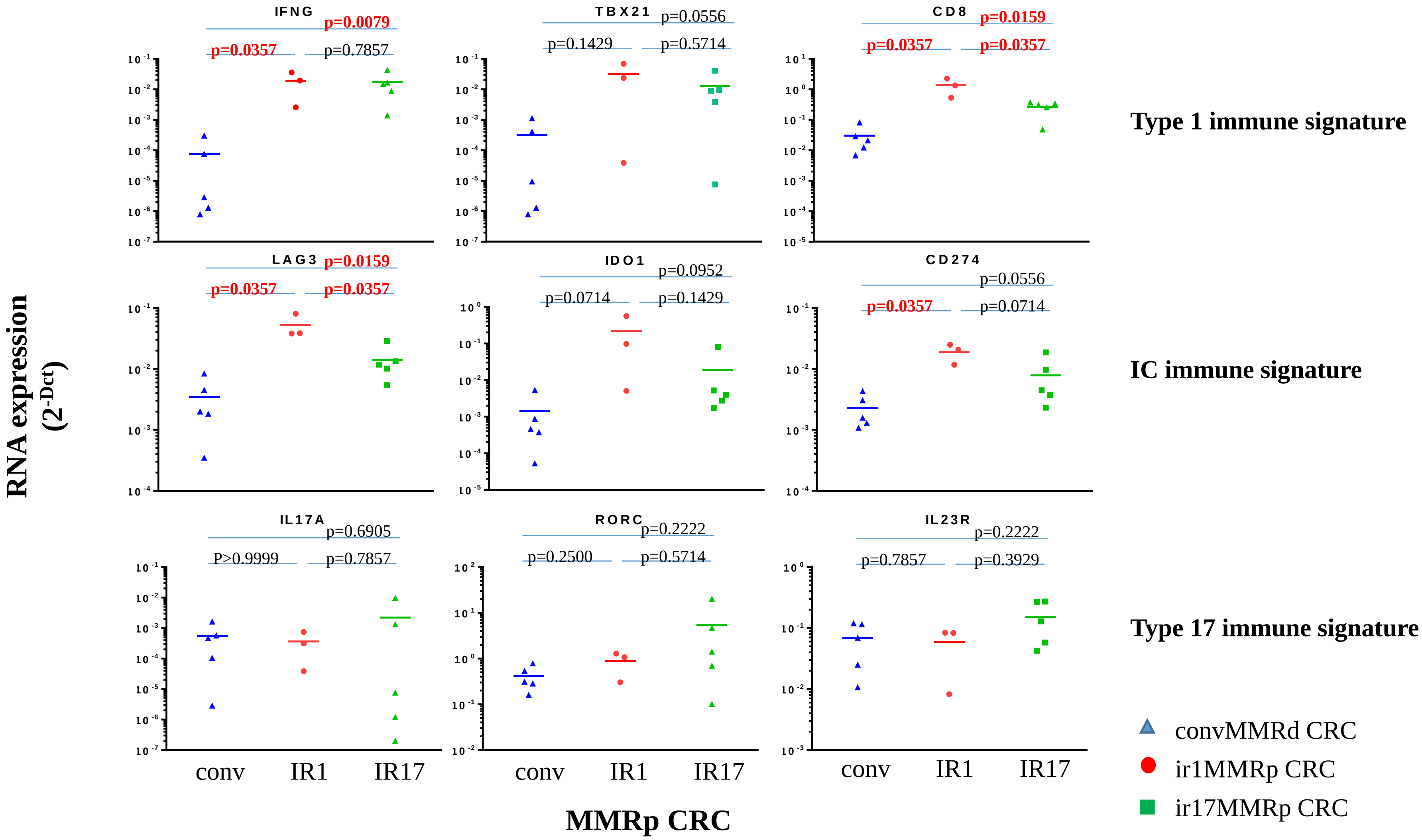
Supplementary figure S2. IFNγ-producing PD1^{hi} CD8^{+} T cells are not detected in the distal normal colon tissue. A, Mean of fluorescence of IFNγ production in PD1^{neg}, PD1^{lo} and PD1^{hi} gates (see **fig.2&S3**). Comparison of IFNγ expression in CD8+ T cells according to the PD-1 expression (Neg, low and high). IFNγ expression level is characterized by the mean of fluorescence (MFI) in the gate of interest. Means of IFNγ MFI were compared between PD1^{neg}, PD1^{Lo} and PD1^{hi} CD8^{+} TILs. The graph shows the p values of Wilcoxon matched pairs signed rank test to compare the means. $p<0.05$ are significant. MFI are compared between groups using Wilcoxon matched-pairs signed rank test. **B**, flow cytometry analysis of T cells isolated from patient 4042 normal tissue distal from the tumors analyzed in **fig.2A**. Gates show the IFNγ production in PD1^{neg}, PD1^{Lo} and PD1^{hi} T cells. Red arrows point out the low proportion of IFNγ^{+}PD1^{hi} populations.



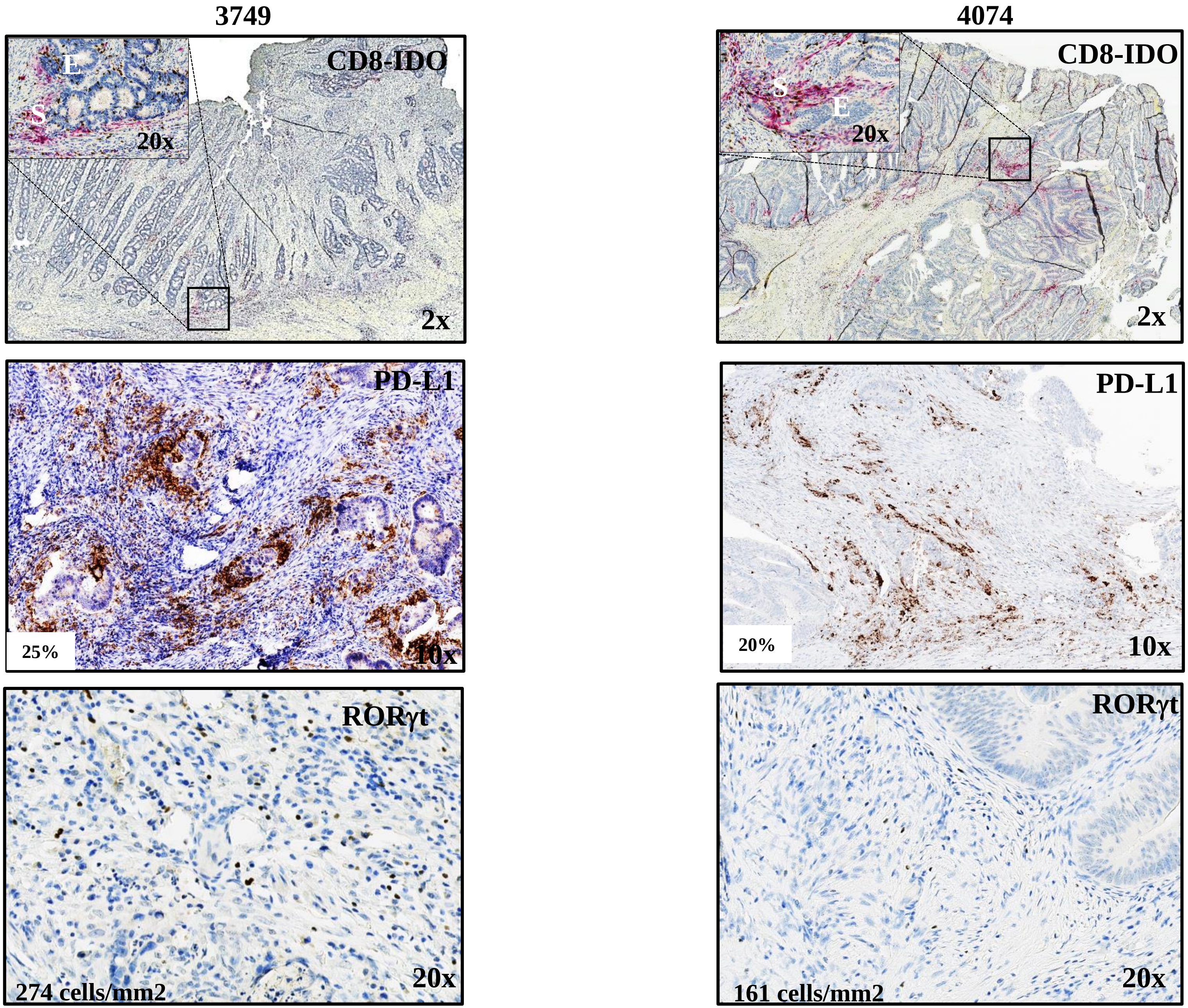
Supplementary figure S3. Cell-sorted PD1^{hi}CD8^{+} TIL in CRC are exhausted T cells. **A**, Gating strategy in CD8^{+}TCRα/β^{+} to dissociate PD^{neg}, PD-1^{lo} and PD-1^{hi} is shown. **B**, Expression of inflammation-related genes was assessed in PD1^{hi} (open bars) and PD1^{lo} (shaded bars) versus PD1^{neg} CD8^{+} TIL cell sorted from 6 CRC freshly resected. The graph represent RQ=2$^{-\Delta\Delta Ct}$. $\Delta\Delta Ct=\Delta Ct-\Delta Ct_{PD1neg}$ and ΔCt represent the CT of the gene of interest normalized by CT$_{GUSB}$ and Ct$_{GAPDH}$. The graph represents all the genes out of 92 selected genes that reached a 2 fold (lower or higher) difference between PD1^{hi} and PD1^{neg}. Bars below the X axis represent genes which are downregulated compared to specimen-matched PD-1^{neg} CD8 cells. Bars above the X axis represent genes which are upregulated compared to specimen-matched PD-1^{neg} CD8 cells. Y axis displays log scale. p-value is based on paired T-test and Wilcoxon signed-rank test to compare means between PD1^{lo} and PD1^{hi} populations**.** *, $p<0.05$ is significant.



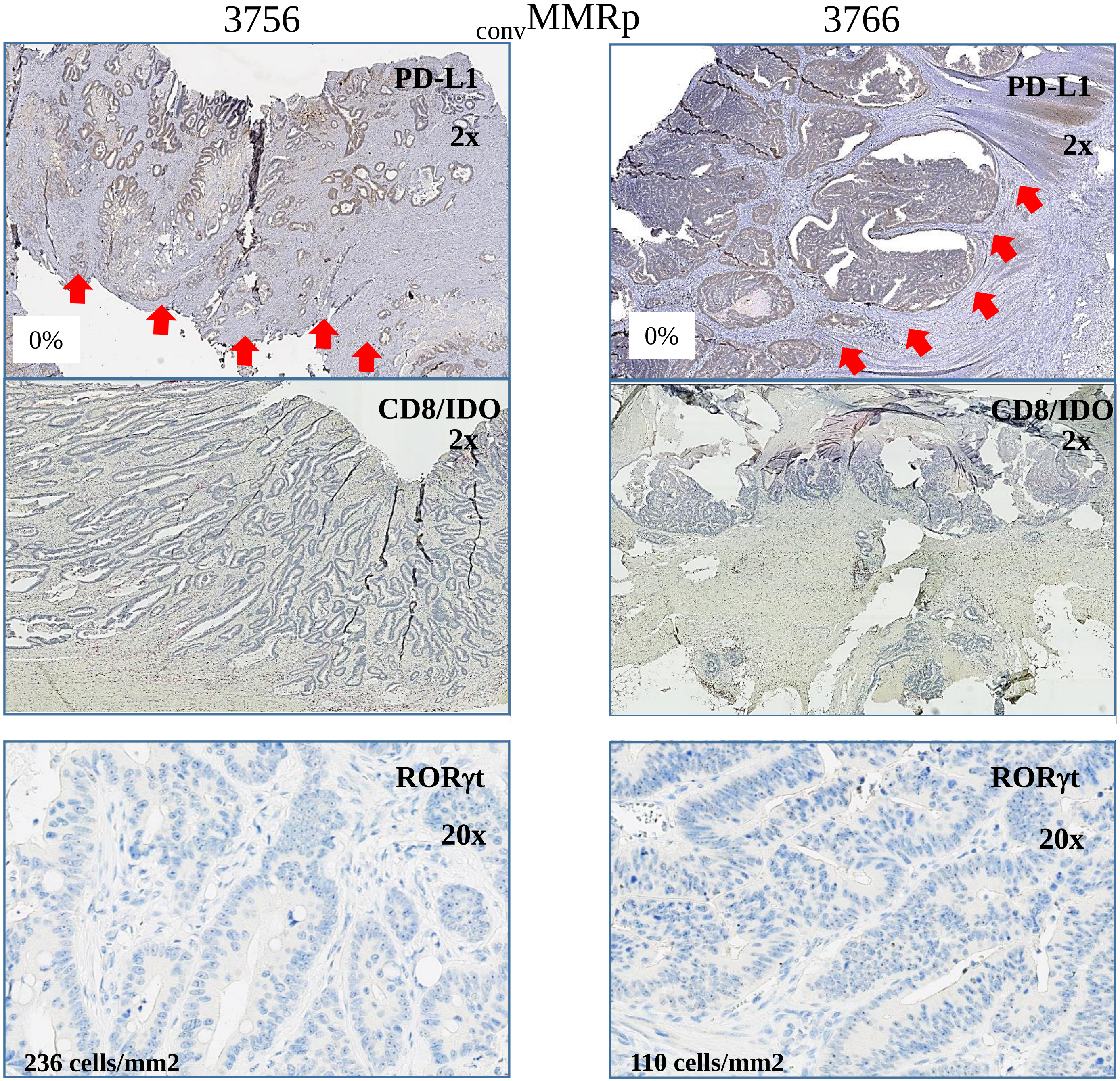
Supplementary figure S4. MSI high MMRd CRC 3784 TILs include only few IFNγ $^{+}$ PD1^{hi} CD4^{+} or CD8^{+} T cells. MFC analysis of tumor (**A**) and normal (**B**) colon-infiltrating T cells. In upper row, graphs show the IFNγ production in PD1^{neg} , PD1^{lo} , PD1^{hi} CD8^{+} T cells (left dot plot), and CD4^{+} T cells (right dot plot). In lower row, graphs showed IL17 production in PD1^{neg} , PD1^{lo} , PD1^{hi} CD4^{+} T cells. Red arrows indicate the IFNγ in PD1^{hi}



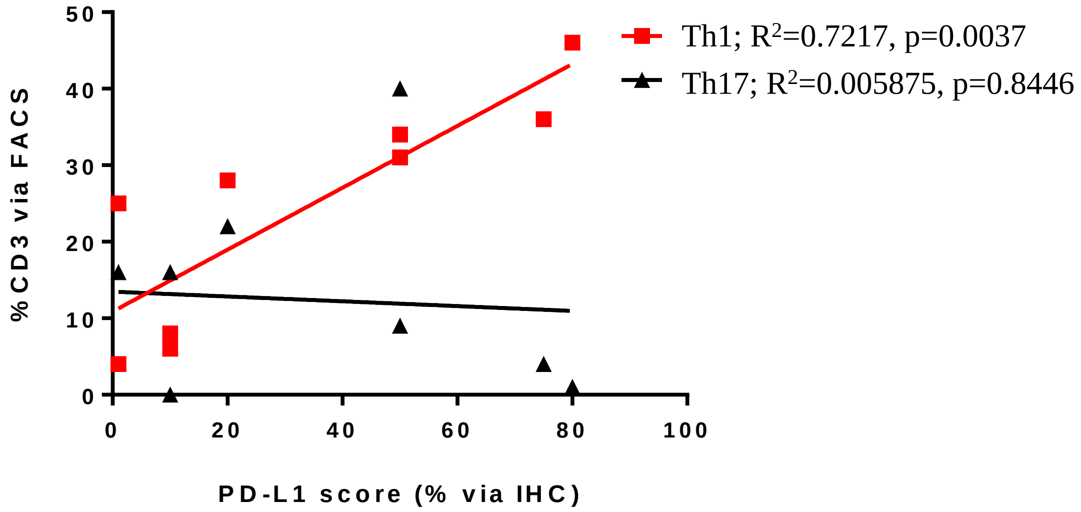
Supplementary figure S5. Gene expression analysis in conventional MMRp (convMMRp; blue triangles), Type 1 immunoreactive MMRp (ir1MMRp; red circles) and type 17 immunoreactive MMRp (ir17MMRp; green squares) colon tumors. Type 1 immunity- (top raw), immune checkpoint- (IC; middle raw)- and type17 immunity- (bottom raw)-associated genes were tested for their expression in FFPE tumor tissue using taqman qRT-PCR. Means of expression were compared between groups using individual Mann-Whitney tests. p<0.05 are significant.



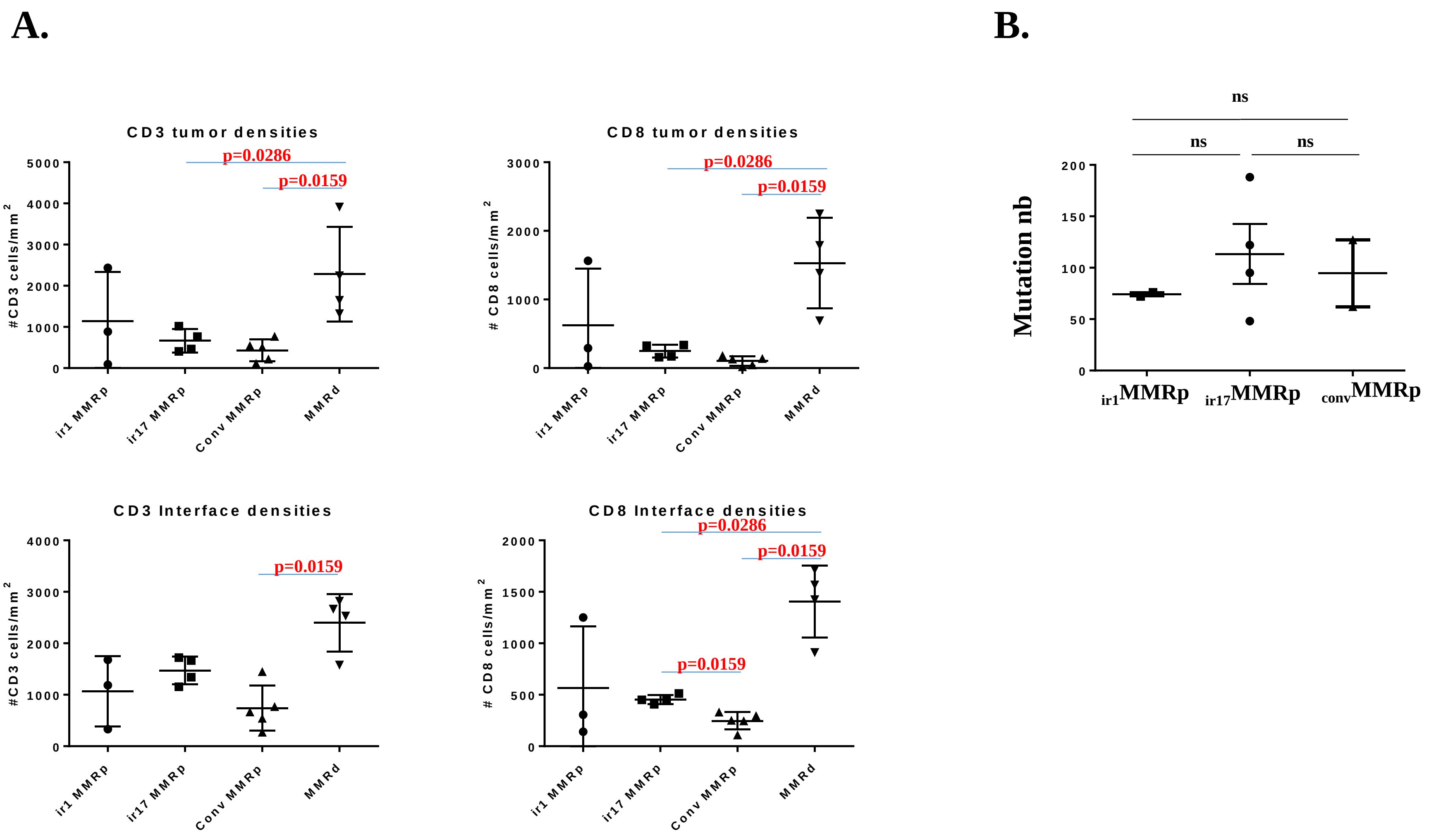
Supplementary Figure S6. IHC staining on type 17 immunoreactive MMRp CRC. Dual staining IDO1(red)/CD8(brown) (Top), PD-L1 (middle) and RORγT (bottom) in representative type 17 immunoreactive MMRp resected primary CRC (3749 and 4074). S, stroma; E, epithelial cells. RORγT cells densities expressed as number of cells/mm2 in tumor invasive front.



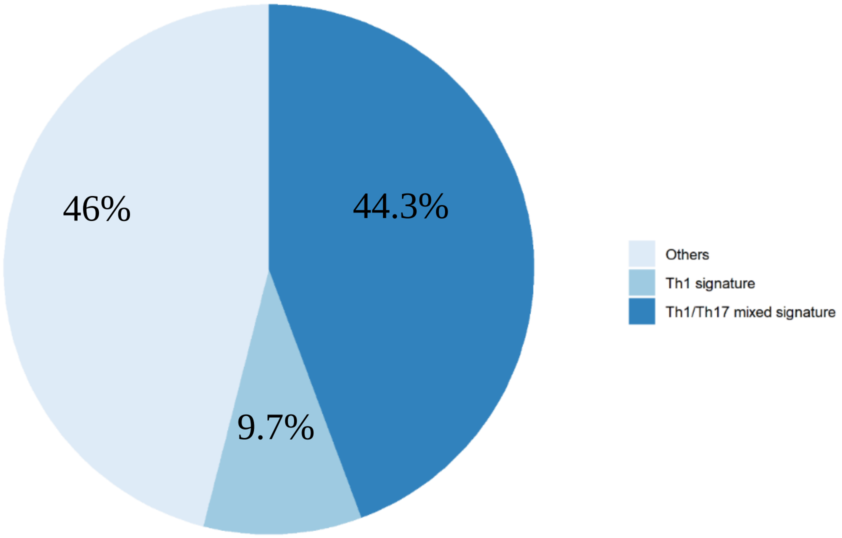
Supplementary Figure S7. IHC staining on conventional MMRp CRC. PD-L1 (Top), dual staining IDO1(red)/CD8(brown) (middle), and RORgT (bottom) stainings in representative conventional MMRp resected primary CRC (3756 and 3766). Red arrows indicate the invasive front. RORgT cells densities expressed as number of cells/mm2 in annotated tumor invasive front.



Supplementary figure S8. Correlation between the PD-L1 expression in IF of primary colon tumors and the proportion of Th1 and Th17 in TILs. For each colon tumor specimen, TIL were freshly isolated for intracellular cytokine and flow cytometry analysis as well as FFPE tissue sections were obtained for PD-L1 IHC and expression scoring. $CD4^{+}IFN\gamma^{+}$ (Th1, red squares) and $CD4^{+}IL17^{+}$ (Th17, black triangles) T cells infiltration are expressed as % of the $CD3^{+}$ T cells. PD-L1 was blindly scored independently by two pathologists. The graph shows the linear regression with R^2 coefficient and p values. Significant p values <0.05



Supplementary Figure S9. CD3 and CD8 cells quantification by IHC and tumor mutation numbers in untreated resected primary CRC lesions. A, CD3 and CD8 digital image quantification in resected primary colon tumors of untreated CRC patients segregated by their MMR status and immunoreactivity, $_{\text{Ir1}}$MMRp, $_{\text{ir17}}$MMRp, $_{\text{conv}}$MMRp and MMRd CRC specimens. Scoring was performed independently by two pathologists. Mann-Whitney test, $p<0.05$ are considered significant. **B**, mutations number in resected primary colon tumor of untreated CRC patients segregated by their immunoreactivity, $_{\text{Ir1}}$MMRp, $_{\text{ir17}}$MMRp, and $_{\text{conv}}$MMRp specimens and for which exomic sequencing and somatic mutations were characterized (unknown in $_{\text{ir1}}$MMRp #3754, MMRp #3756, 3764, 3766).



Supplementary Figure S10. Detection of Type 1 and Type 17 immunoreactive MMRp CRC samples in TCGA database. 176 MSS (or MMRp) CRC samples in the TCGA database have been tested for the detection of a Th1 (representative of Type 1 irMMRp CRC), mixed Th1/Th17 (representative of Type 17 irMMRp CRC) immune signatures based on the analysis of the expression of the genes *IFNG/TBX21/IDO1* (Th1) and *IL17A/RORC/IL23R* (Th17). The details of the gene expression are provided in **Supplementary Table S7** .