

Supporting information

Supporting Methods

Next generation sequencing (NGS) analysis (supplementary)

Two hundred nanograms genomic DNA was sonicated into 150–200 bp fragments using a Covaris-M220 (Covaris Inc., Woburn, MA, USA). Libraries were constructed using the DNA fragments ligated with paired-end adaptors (SureSelect XT library preparation kit for HCC) (Agilent Technologies, Santa Clara, CA, USA) or a SureSelect XT HS kit (for mCRC) (Agilent Technologies), purified, and amplified using PCR. The exome regions were enriched from adapter-ligated libraries via hybridization and PCR amplification using biotinylated oligoRNA bait targeting the human exome (SureSelect Human All Exon V6) (Agilent Technologies) and MHC regions.

For RNA-seq, libraries were prepared from 300–800 ng total RNA using a TruSeq™ Stranded mRNA library preparation kit (for HCC) (Illumina, San Diego, CA, USA) or a SureSelect strand-specific RNA library preparation kit (for mCRC) (Agilent Technologies). Sequencing of 101 base-paired-end reads (for the whole-exome DNA library) and 76 base-paired-end reads (for the RNA library) was performed using a NextSeq™ 550 Sequencer (Illumina) according to the manufacturer's protocol. Fastq files were generated from the raw output data of the NextSeq™ 550 using bcl2fastq, followed by a FastQC quality check.²¹

***In vitro* Human PBMC stimulation**

A total of 1×10^8 PBMC were harvested from patients after surgical resection, with for each sampling a quantity of 20~30 ml total blood, performed three to five times at the different time points. For $\gamma\delta$ T cells induction, the CD8 negative fraction was purified from PBMC using MACS beads protocol. The CD8 negative fraction was cultured in 10% AB serum AIM-V medium supplemented with 5 μ M Zoledronate and 1000 IU/ml IL2 for seven days. Cells were then harvested, irradiated with 100 Gy X ray, and 2×10^3 $\gamma\delta$ T cells were used as stimulator. For responder cells, CD8 positive fraction was also purified from PBMC of the same patient. Then, cells were co-cultivated with $\gamma\delta$ T cells, in the presence of each peptide; 100ng/ml HLA-A24 restricted CMV peptide (QYDPVAALF), 100ng/ml HLA-A02 restricted FluM1 peptide (GILGFVFTL), 10 μ g/ml HLA-A02 restricted MART-1 peptide (ELAGIGILTV), and 3 μ g/ml neopeptide. After two days, we added 50 IU/ml IL-2 and 10 ng/ml IL-15, and made the co-culture up to two weeks. After stimulation, cells were harvested, washed and used for ELISpot assay with autologous $\gamma\delta$ T cells as target in the presence of 3 μ g/ml of each neopeptide.

Table S1. Data for 28 patients with hepatocellular carcinoma (HCC) and 18 with metastatic colorectal carcinoma (mCRC) harboring mutations.

Origin	Patient_ID	Total mutation	Non-synonymus	Frame shift	average VAF(%)	MSI Status	SCORE of Top50	SCOREadj of Top50	Haplotype for HLA-Tg	Peptide used for vaccinaiton
HCC	114	93	67	6	38.55	MSS	3.704	3.7412	-	-
HCC	116	158	126	7	36.54	MSS	3.8927	4.7791	-	-
HCC	117	174	137	7	39.06	MSS	3.8105	4.5871	A24/B7	15
HCC	120	64	46	6	32.81	MSS	2.9851	3.2847	-	-
HCC	122	80	62	5	40.1	MSS	3.8125	3.5717	A24	6
HCC	123	65	50	3	24.71	MSS	2.7668	1.5957	A24	3
HCC	125	85	65	6	19.45	MSS	3.2658	3.0461	-	-
HCC	126	115	96	8	15.25	MSS	3.4346	2.6327	A02	14
HCC (combined)	127	166	126	9	37.34	MSS	3.9875	4.7163	B35	12
HCC	128	38	26	2	31.18	MSS	1.3887	0.0859	-	-
HCC	129	174	129	6	40.95	MSS	3.76	4.8427	A24	8
HCC	130	84	64	7	23.13	MSS	3.4359	1.0824	A24	8
HCC	134	111	82	7	32.74	MSS	3.3958	3.0625	B35	10
HCC	135	86	65	6	24.27	MSS	3.4451	3.0294	B35	8
HCC	136	262	97	3	16.05	MSS	3.5552	3.5702	-	-
HCC	137	115	87	6	36.12	MSS	3.759	4.0991	B35	7
HCC	138	86	54	2	44.44	MSS	3.343	2.5629	-	-
HCC	139	213	164	6	33.42	MSS	3.9112	4.7178	-	-
HCC (iCCA)	140	55	39	1	25.34	MSS	2.428	1.1494	-	-
HCC	141	95	76	5	15.15	MSS	3.3963	2.5738	-	-
HCC	142	118	89	8	35.69	MSS	4.0026	4.2009	B7	8
HCC (metastasis)	143	137	101	7	28.14	MSS	3.4037	2.3631	-	-
HCC	144	95	75	4	44.26	MSS	3.744	3.8198	-	-
HCC	145	215	168	8	28.19	MSS	3.9814	4.8403	A02	17
HCC	146	109	84	8	16.79	MSS	3.9888	5.4377	A02	13
HCC (sarcomatoid)	147	234	175	13	37.95	MSS	4.0387	5.3694	B35	9
HCC	148	103	79	6	44.35	MSS	3.784	4.7142	-	-
HCC	150	244	205	17	37.67	MSS	3.922	4.9919	A02	14
mCRC	7	172	110	3	26.06	MSS	3.7779	2.2932	A24	8
mCRC	13	189	135	6	34.66	MSS	3.8822	3.5385	A24	14
mCRC	16	134	93	2	11.51	MSS	3.1832	0.1424	A02/A24/B7	12
mCRC	21	107	77	4	32.51	MSS	3.631	2.8058	A02	5
mCRC	31	148	100	6	35.59	MSS	3.7211	3.6743	A24	13
mCRC	40	157	111	11	17.9	MSS	3.8488	4.5017	B7	10
mCRC	45	161	108	6	35.46	MSS	3.7267	3.4863	A02	12
mCRC	61	200	136	10	20.47	MSS	3.8732	2.2355	A02	10
mCRC	66	104	66	6	7.78	MSS	3.4288	1.3232	A02	14
mCRC (& HCC)	92	123	92	5	29.46	MSS	3.8505	4.6611	-	-
mCRC	103	133	93	6	29.68	MSS	3.4402	3.0434	A02/A24	5
mCRC	154	199	146	5	27.02	MSS	3.7647	4.2422	-	-
mCRC	156	113	75	3	15.15	MSS	3.353	2.7729	A24	-
mCRC	157	166	109	4	30.2	MSS	3.5256	3.8699	-	-
mCRC	159	107	71	2	32.52	MSS	3.5629	3.7201	A24	11
mCRC	164	117	83	4	33.4	MSS	3.7926	4.6262	A24/B7	12
mCRC	171	144	98	7	17.97	MSS	3.7691	3.5967	-	-
mCRC	172	105	62	7	23.95	MSS	3.8443	3.5671	-	-

Microsatellite instability (MSI) status, SCORE/SCOREadj for top 50, mutations, human leukocyte antigen (HLA) type, and number of peptides used are shown.

Table S2. Medians and statistical tests of mutation characteristics and SCORE/SCOREadj for patients with hepatocellular carcinoma (HCC) and metastatic colorectal carcinoma (mCRC).

	HCC	mCRC	<i>Mann-Whitny U-test</i>
n	28	18	-
Non-synonymus	83 (26-205)	95.5 (62-146)	<i>ns (p=0.2856)</i>
Frameshift	6 (1-17)	5.5 (2-11)	<i>ns (p=0.1705)</i>
Total mutation	110 (38-244)	139 (104-200)	<i>ns (p=0.0995)</i>
SCORE of Top50	3.724 (1.3887-4.0387)	3.746 (3.353-3.882)	<i>ns (p=0.7807)</i>
SCOREadj of Top50	3.656 (0.086- 5.438)	3.553 (0.142-4.661)	<i>ns (p=0.2986)</i>

Table S3. Predicted neopeptides used for *in vitro* stimulation with patient's PBMC

* = positive wells by paired t-tests; p < 0.05

The results of gray-filled columns showed in Figure S1 as representative pictures.

Origin	Patient_ID	Peptide_ID	Peptide sequences	Length	Score	Haplotype	ELISPOT assay (S-N)	Exp.column No.
HCC	117	117-3-1	HPVGHLSM	9	4.20393	B07:02	11.3	1
HCC	117	117-3-4	MWLKPVSWCW	10	3.41241	A24:02	11.3	2
HCC	117	117-3-7	KHMWLKPVSW	10	2.97003	A24:02	35.7	3
HCC	117	117-3-8	CVYQGEWHR	9	2.95091	A11:01	7	4
HCC	117	117-2-1	ASREVLAVK	9	3.93185	A11:01	-2	5
HCC	117	117-2-2	HYDFPYGSF	9	3.87974	A24:02	9.7	6
HCC	117	117-2-4	LMPTAVPSIF	10	3.71683	A24:02	-10	7
HCC	117	117-2-5	TVTHWKIQK	9	3.70208	A11:01	0	8
HCC	117	117-2-6	VSQTRLLMK	9	3.69574	A11:01	7	9
HCC	117	117-2-7	PFLIKDMTF	9	3.58034	A24:02	42	10
HCC	117	117-2-8	KIRRQLAQL	9	3.54999	B07:02	3.7	11
HCC	117	117-2-9	AQLQGYERK	9	3.4756	A11:01	5.7	12
HCC	117	117-2-11	LYYTGSDNII	10	3.36483	A24:02	39.3	13
HCC	117	117-2-12	RQMATMNT	9	3.36295	A11:01	19.3	14
HCC	117	117-2-13	LYYTGSDNI	9	3.32842	A24:02	22	15
HCC	117	117-2-14	TILNFLRHGK	10	3.2482	A11:01	29.3	16
HCC	117	117-2-15	VVLGVGVAR	9	3.21884	A11:01	35.7	17
HCC	117	117-2-16	LMPTAVPSI	9	3.11319	A24:02	23.7	18
HCC	117	117-2-17	VARLLVNVL	9	2.60684	B07:02	40.7	19
HCC	117	117-2-18	SVASRQFR	9	2.52602	A11:01	12.3	20
HCC	117	117-2-19	GTILNFLRH	9	2.01575	A11:01	12.7	21
HCC	117	117-2-20	TYLGRSLGPV	10	1.92145	A24:02	31.7	22
HCC	117	117-1-1	TYFGTILNF	9	4.20952	A24:02	18	23
HCC	117	117-1-2	TVMKQLMRK	9	4.19503	A11:01	27.7	24
HCC	117	117-1-3	VPRRVIHVV	9	4.1761	B07:02	20.7	25
HCC	117	117-1-4	KPAHTASKL	9	4.149	B07:02	17	26
HCC	117	117-1-5	HPAHHSCPL	9	4.11375	B07:02	18	27
HCC	117	117-1-6	LPQHTFHVI	9	3.9609	B07:02	37	28
HCC	117	117-1-7	RPHPAHSC	9	3.74953	B07:02	4.3	29
HCC	117	117-1-8	AVSQTRLLMK	10	3.74783	A11:01	-9	30
HCC	117	117-1-9	ALHYQQELK	9	3.72368	A11:01	12.3	31
HCC	117	117-1-10	TVIEMTDSK	9	3.67531	A11:01	14.3	32
HCC	117	117-1-11	HLHNYALYY	9	3.63417	A11:01	0.7	33
HCC	117	117-1-12	SITKKKFKK	9	3.57907	A11:01	26.7	34
HCC	117	117-1-13	SHYDFPYGSF	10	3.47561	A24:02	39.3	35
HCC	117	117-1-14	TYFGTILNFL	10	3.45114	A24:02	10.3	36
HCC	117	117-1-15	RTRSVQRKL	9	3.41496	B07:02	2	37
HCC	117	117-1-16	LIPFMPFLIK	10	3.34167	A11:01	1.7	38
HCC	117	117-1-17	VYSFRVKISM	10	3.06573	A24:02	4.3	39
HCC	117	117-1-18	NLRRATTYL	9	3.05065	B07:02	1	40
HCC	117	117-1-19	AVSQTRLLM	9	3.00315	B07:02	2	41
HCC	117	117-1-20	HPAHHSCPLL	10	3.03029	B07:02	9	42
HCC	117	117-1-21	AVEQQREAL	9	3.01959	B07:02	0.7	43
HCC	117	117-1-22	AGACNTVMK	9	2.98346	A11:01	2	44
HCC	117	117-1-23	YPTVKQICKL	10	2.93133	B07:02	531	* 45

HCC	117	117-1-24	VISTLEVQY	9	2.82861	A11:01	1.3		46
HCC	117	117-1-25	MPFLIKDMTF	10	2.76608	B07:02	4		47
HCC	117	117-1-26	DPTYFGTIL	9	2.62689	B07:02	2.3		48
HCC	117	117-1-27	LMRRQMATM	9	2.58648	B07:02	54.3		49
HCC	117	117-1-28	VYWTQEDGVL	10	2.58008	A24:02	6.7		50
HCC	122	122-3-1	LLMMTIASV	9	4.01796	A02:07	136.5	*	1
HCC	122	122-3-2	ILGVGTSFV	9	3.99571	A02:07	8.5		2
HCC	122	122-3-3	GLAGAIPRI	9	3.90801	A02:07	6.0		3
HCC	122	122-3-4	LMTATVTTV	9	3.7686	A02:07	1.5		4
HCC	122	122-3-5	KMMAEEMTV	9	3.69557	A02:07	-5.0		5
HCC	122	122-3-6	VHLPRATL	9	3.11966	A24:02	4.5		6
HCC	122	122-3-7	ALRRGILGV	9	2.89711	A02:07	4.0		7
HCC	122	122-3-8	TMVGGNTKV	9	2.84967	A02:07	3.0		8
HCC	122	122-3-9	IVPPGVLTQV	10	2.57803	A02:07	-3.5		9
HCC	122	122-3-11	TLMTATVTT	9	2.16387	A02:07	25.5	*	10
HCC	122	122-1-1	IFPLKTPAF	9	4.15447	A24:02	3.5		11
HCC	122	122-1-2	GAKDVANSF	9	3.73691	B46:01	-10.5		12
HCC	122	122-1-3	KYERLETMI	9	3.62477	A24:02	-1.0		13
HCC	122	122-1-4	YLDSEIHSGA	10	3.53198	A02:07	10.0		14
HCC	122	122-1-5	YLDSEIHSG	9	2.36197	A02:07	2.0		15
HCC	122	122-1-6	FLHCEDRPV	9	2.15524	A02:07	-4.0		16
HCC	122	122-1-7	FLHCEDRPVV	10	1.9327	A02:07	-7.5		17
HCC	122	122-1-8	IFPLKTPAFA	10	0.15009	A24:02	-4.5		18
HCC	122	122-1-9	MMPNRRNVY	9	0.03588	A24:02	3.5		19
HCC	122	122-1-10	GEVNAPTQF	9	0.00804	A24:02	-3.0		20
HCC	122	122-1-11	PLKTPAFAI	9	0.00794	A24:02	-4.5		21
HCC	122	122-1-12	MAALPKLPH	9	0.00346	B46:01	2.5		22
HCC	122	122-1-13	SYLDSEIHS	9	0.00233	A24:02	3.5		23
HCC	122	122-1-14	KSLGDLQDEY	10	0.00093	B46:01	-4.0		24
HCC	122	122-1-15	KYERLETMIQ	10	0.00087	A24:02	-12.0		25

Table S4. ELISpot assays of all neopeptides used for vaccination and organized by experiment numbers and groups.

* = positive wells by paired t-tests; p < 0.05; experiment conducted in triplicate.

The gray-filled columns showed the peptides used in the results.

Origin	Patient_ID	Peptide_ID	Peptide sequences	Length	Score	Haplotype	ELISPOT assay (spots/well)	Exp.No.-Gp.No.	Exp. No.	Group No.
HCC	126	126-1-01	KLMEGCKRL	9	4.0259	A02	468.67 *	01-01	1	1
HCC	126	126-1-12	ILLKYVVIQA	10	2.2007	A02	4	01-01	1	1
HCC	126	126-1-13	LLGCGAIAI	9	2.4865	A02	7.33	01-01	1	1
HCC	126	126-1-19	YPLEYPTHPL	10	0	A02	5.83	01-01	1	1
HCC	126	126-2-08	FLQILLKYV	9	2.8578	A02	275.67 *	01-01	1	1
HCC	126	126-1-21	LLGCGAIAIA	10	0.6706	A02	715.33 *	01-02	1	2
HCC	126	126-1-26	LQILLKYVV	9	0.2046	A02	48.17 *	01-02	1	2
HCC	126	126-1-27	LLKYVVIQAA	10	0.0003	A02	1.67	01-02	1	2
HCC	126	126-1-29	ILLKYVVIQ	9	0	A02	1	01-02	1	2
HCC	126	126-1-31	KLMEGCKRLQ	10	0	A02	959.83 *	01-02	1	2
HCC	126	126-1-34	QILLKYVVI	9	0.0002	A02	1.5	01-03	1	3
HCC	126	126-1-35	GNYGKVPLA	9	0.0276	A02	0.83	01-03	1	3
HCC	126	126-1-36	PLLGCGAIA	9	0.0006	A02	2	01-03	1	3
HCC	126	126-1-37	TLLDIADPQ	9	0.0001	A02	2	01-03	1	3
HCC	117	117-1-01	TYFGTILNF	9	4.2102	A24	456.17 *	03-01	3	1
HCC	117	117-2-04	LMPTAVPSIF	10	3.6987	A24	19.17	03-01	3	1
HCC	122	122-1-01	IFPLKTPAF	9	4.1628	A24	166.33 *	03-01	3	1
HCC	129	129-2-03	SYLDSGIHF	9	4.2209	A24	802.67 *	03-01	3	1
HCC	130	130-1-01	RYSDPPAYCL	10	4.044	A24	1000 *	03-01	3	1
HCC	117	117-1-14	TYFGTILNFL	10	4.2102	A24	647 *	03-02	3	2
HCC	117	117-2-07	PFLIKDMTF	9	3.8171	A24	23.83 *	03-02	3	2
HCC	123	123-1-01	RFHQESVMI	9	3.4239	A24	1.67	03-02	3	2
HCC	129	129-1-10	AYERRIQRL	9	3.7816	A24	3.17	03-02	3	2
HCC	130	130-2-04	RFLDEDPQRW	10	3.8329	A24	3.83	03-02	3	2
HCC	117	117-1-17	VYSFRVKISM	10	2.9319	A24	29.5 *	03-03	3	3
HCC	117	117-2-16	LMPTAVPSI	9	3.2558	A24	3.83	03-03	3	3
HCC	129	129-1-16	TFPQFRAIQL	10	3.4368	A24	5.67	03-03	3	3
HCC	129	129-1-23	GFYAFLNWF	9	3.4082	A24	370 *	03-03	3	3
HCC	122	122-1-08	IFPLKTPAFA	10	0	A24	6	03-04	3	4
HCC	122	122-1-10	GEVNAPTQF	9	3.9846	A24	4.83	03-04	3	4
HCC	129	129-1-41	SHGFYAFLNW	10	0.1037	A24	453.67 *	03-04	3	4
HCC	130	130-1-20	RYSDPPAYC	9	1.9038	A24	5.67	03-04	3	4
HCC	122	122-1-11	PLKTPAFAI	9	0.0012	A24	29.33	07-01	7	1
HCC	122	122-1-13	SYLDSEIHS	9	0.0002	A24	12.5	07-01	7	1
HCC	123	123-1-11	VSQPVRGLI	9	2.2926	A24	23.83	07-01	7	1
HCC	123	123-1-12	RFHQESVMIA	10	0	A24	18.17	07-01	7	1
HCC	130	130-1-61	LLQGKLGILL	9	2.7557	A24	19.67	07-01	7	1
HCC	129	129-3-19	VYSVRITAV	9	3.6791	A24	10.67	07-02	7	2
HCC	129	129-3-22	KYTGVSQSLNW	10	3.1373	A24	419 *	07-02	7	2
HCC	130	130-3-05	VWWCPMSRL	9	3.4684	A24	102.5 *	07-02	7	2
HCC	130	130-3-07	EFWMTETLF	9	3.3364	A24	143.5 *	07-02	7	2
HCC	122	122-3-06	VHLPRATL	9	3.4139	A24	12 *	07-03	7	3
HCC	129	129-3-29	LLPSTASTNW	10	1.9682	A24	5.5	07-03	7	3
HCC	130	130-3-13	LHLPQFHSL	9	3.2497	A24	5	07-03	7	3
HCC	130	130-3-17	PQLLHLPQF	9	3.0789	A24	6.67 *	07-03	7	3
HCC	145	145-1-10	SLAQYLISV	9	4.2364	A02	10	13-01	13	1
HCC	146	146-1-02	ALMKFSWAM	9	4.1541	A02	14.83	13-01	13	1
HCC	146	146-1-15	ALNPDAVQL	9	4.1446	A02	86.17 *	13-01	13	1
HCC	146	146-1-16	SLWCTLHFM	9	3.6784	A02	632.17 *	13-01	13	1
HCC	150	150-1-13	TLTDAGTMAL	10	3.8052	A02	18	13-01	13	1
HCC	145	145-1-21	ILVSPLVCL	9	NA	A02	1000 *	13-02	13	2
HCC	145	145-1-23	RLVLGGVLV	9	3.9776	A02	131.67 *	13-02	13	2
HCC	145	145-1-29	LLRDADSVTL	10	3.685	A02	4	13-02	13	2
HCC	145	145-2-20	GLLGAMAFAL	10	3.6398	A02	1000 *	13-02	13	2
HCC	150	150-1-35	YLNDSGIHSGA	10	3.4288	A02	1000 *	13-02	13	2
HCC	145	145-1-30	MMFAGLLGA	9	3.1443	A02	1000 *	13-03	13	3
HCC	146	146-1-37	FLSLTHTKA	9	3.7193	A02	14.5 *	13-03	13	3
HCC	150	150-1-66	YLNDSGIHSG	9	2.4643	A02	11.5	13-03	13	3
HCC	150	150-2-86	RLDHIIDAL	10	2.3379	A02	5.17	13-03	13	3
HCC	146	146-1-48	FLSLTHTKAV	10	2.2712	A02	8.17	16-01	16	1
HCC	150	150-1-95	TLTDAGTMA	9	2.6987	A02	5.67	16-01	16	1
HCC	150	150-2-89	RLDHIIDDA	9	2.3789	A02	9.17	16-01	16	1
HCC	145	145-1-64	YLISVRLSS	9	1.1965	A02	8.33	16-01	16	1
HCC	146	146-3-07	FLTNNSEV	9	4.1154	A02	446.33 *	16-02	16	2
HCC	146	146-3-11	GLFCELRHV	9	3.9684	A02	774.5 *	16-02	16	2
HCC	146	146-3-17	LLMTPGSHL	9	3.8184	A02	8.08	16-02	16	2
HCC	150	150-3-06	TVIDSTITI	9	3.8999	A02	1000 *	16-02	16	2
HCC	145	145-3-47	GLGQASEPTV	10	2.6244	A02	13.83	16-03	16	3
HCC	146	146-3-26	YQASMLVFL	9	3.0708	A02	833.67 *	16-03	16	3
HCC	146	146-3-44	LMTPGSHLPL	10	2.6018	A02	10.17	16-03	16	3
HCC	150	150-3-21	RLQLQSPAL	9	3.6926	A02	12.5	16-03	16	3
HCC	145	145-1-48	KLSDQTHST	9	2.7882	A02	71.83 *	19-01	19	1
HCC	145	145-1-50	MMFAGLLGAM	10	2.2099	A02	609.5 *	19-01	19	1

HCC	145	145-1-85	SLETVCLLLA	10	0.4845	A02	29.17		19-01	19	1
HCC	145	145-2-54	QVSNVQMFL	9	2.2049	A02	769	*	19-01	19	1
HCC	145	145-2-84	LLGAMAFALA	10	0.4268	A02	134.67	*	19-01	19	1
HCC	145	145-1-87	QLCLVQVSNV	10	0.5384	A02	83.67	*	19-02	19	2
HCC	145	145-1-88	RIARNTQFI	9	0.8102	A02	4		19-02	19	2
HCC	145	145-1-99	EMNYITAVI	9	0.0992	A02	738.83	*	19-02	19	2
HCC	145	145-2-66	HMMMFAGLL	9	1.4221	A02	68.33	*	19-02	19	2
HCC	146	146-1-78	AVIGGVDSGL	10	2.5595	A02	6		19-02	19	2
HCC	146	146-1-83	ATVDVEHSHV	10	1.9703	A02	4.17		19-03	19	3
HCC	146	146-1-99	RSALVLFFI	9	0.3641	A02	74.17	*	19-03	19	3
HCC	150	150-2-116	SLYYPLSPGT	10	0.9321	A02	5.83		19-03	19	3
HCC	150	150-1-124	KLRSLLCVGL	10	1.1442	A02	11.17		19-04	19	4
HCC	150	150-1-134	LLFSISGGT	9	0.7371	A02	9.5		19-04	19	4
HCC	150	150-1-139	NLCKIQFHNH	10	0.3591	A02	9.83		19-04	19	4
HCC	150	150-2-125	IQAKLRLLEV	10	0.4479	A02	36.33	*	19-04	19	4
HCC	150	150-2-162	SLYYPLSPG	9	0.0267	A02	11.67		19-04	19	4
mCRC	7	007-1-11	NYVNPDKHW	10	3.8774	A24	18	*	27-01	27	1
mCRC	7	007-1-22	SWFKNAEEDL	10	1.6129	A24	7.83		27-01	27	1
mCRC	7	007-1-23	IWKQESKNI	9	1.9407	A24	10.67		27-01	27	1
mCRC	7	007-1-38	AYLHTQHYI	9	4.0746	A24	772.67	*	27-01	27	1
mCRC	7	007-2-36	YFPMLLNHL	9	4.0127	A24	535.17	*	27-01	27	1
mCRC	13	013-2-25	TWHKIDILI	10	2.5255	A24	24		27-01	27	1
mCRC	7	007-1-19	YVNPDKHW	9	1.4701	A24	2.33		27-02	27	2
mCRC	7	007-1-40	RFHGALNVDL	10	0.5216	A24	3.17		27-02	27	2
mCRC	7	007-2-33	RYFPMLLNHL	10	4.0727	A24	1000	*	27-02	27	2
mCRC	13	013-2-04	SSPAEFFEF	9	4.0087	A24	135.83	*	27-02	27	2
mCRC	13	013-2-11	KSLPGVVTW	9	3.2217	A24	647.83	*	27-02	27	2
mCRC	13	013-2-13	TWHKIDIL	9	2.9999	A24	2.5		27-02	27	2
mCRC	13	013-1-30	EFLDLIMSTL	10	2.5926	A24	672.5	*	27-03	27	3
mCRC	13	013-1-47	DFQSITYGF	9	3.5719	A24	94.67	*	27-03	27	3
mCRC	13	013-1-58	KYFKHSELA	9	1.9397	A24	5.83		27-03	27	3
mCRC	13	013-2-120	YIDSPYYSW	9	3.6003	A24	791.33	*	27-03	27	3
mCRC	13	013-2-69	LSSPAEFFEF	10	1.9208	A24	714.83	*	27-03	27	3
mCRC	31	031-1-02	YISNDIPYTF	10	3.7417	A24	6.83		27-03	27	3
mCRC	13	013-1-124	FLDLIMSTL	9	0.5546	A24	2.67		27-04	27	4
mCRC	13	013-1-96	AFMLHMKEM	9	1.1342	A24	9.17	*	27-04	27	4
mCRC	13	013-2-107	YSWQGMSPTF	10	3.7985	A24	12.17	*	27-04	27	4
mCRC	13	013-2-113	GYIDSPYYSW	10	4.1954	A24	562.67	*	27-04	27	4
mCRC	16	016-1-07	AYFQMDVQV	9	3.7168	A24	27.83	*	27-04	27	4
mCRC	16	016-1-17	YWCSIAFYQM	10	1.5302	A24	1000	*	27-04	27	4
mCRC	13	013-2-111	SWQGMSPTF	9	4.1879	A24	255.5	*	28-01	28	1
mCRC	31	031-1-06	YYISNDIPY	9	3.7691	A24	3.33		28-01	28	1
mCRC	31	031-1-15	HWLNDEVISF	10	3.8392	A24	10.17		28-01	28	1
mCRC	31	031-1-30	KYPVPFSTF	10	4.2339	A24	1000	*	28-01	28	1
mCRC	31	031-1-42	FHVGFYGTKF	10	2.6816	A24	13.17		28-01	28	1
mCRC	31	031-2-19	RMKFTWPLF	9	3.2634	A24	703.83	*	28-01	28	1
mCRC	31	031-1-37	MFGTYFHVGF	10	3.371	A24	1000	*	28-02	28	2
mCRC	31	031-1-41	YYISNDIPY	10	2.2518	A24	8.17		28-02	28	2
mCRC	31	031-1-61	KYPVPFSTF	9	2.1995	A24	433.17	*	28-02	28	2
mCRC	31	031-1-74	LLQFQEAUF	9	0.4731	A24	7		28-02	28	2
mCRC	31	031-2-18	QLYPGWSAW	9	3.3138	A24	106.67	*	28-02	28	2
mCRC	103	103-2-59	FFVFSFFIM	9	1.5058	A24	520.5	*	28-02	28	2
mCRC	31	031-1-81	YYETGIIRC	9	0.9593	A24	42.33	*	28-03	28	3
mCRC	66	066-3-12	SWMPPPSQSV	10	3.6347	A24	21.67		28-03	28	3
mCRC	66	066-3-14	AYAPATSWM	9	3.9156	A24	20.67		28-03	28	3
mCRC	66	066-3-29	VFILSARVVL	10	1.7723	A24	9.17		28-03	28	3
mCRC	103	103-1-36	FFIMLPLSL	9	2.6366	A24	538.17	*	28-03	28	3
mCRC	159	159-1-04	KFFDDPVLL	9	4.1333	A24	33.5	*	28-03	28	3
mCRC	31	031-1-72	TYFHVGFYF	9	1.1752	A24	11.17		29-01	29	1
mCRC	66	066-3-20	MPPPSQSVF	9	2.0439	A24	58.33	*	29-01	29	1
mCRC	66	066-3-31	KIKDLMNTLF	10	1.2787	A24	2		29-01	29	1
mCRC	103	103-1-06	SFLFHLINI	9	2.6579	A24	52	*	29-01	29	1
mCRC	103	103-1-67	SFFIMLPLSL	10	1.0984	A24	111.67	*	29-01	29	1
mCRC	159	159-1-39	VMPDEEIERI	10	1.762	A24	1.67		29-01	29	1
mCRC	66	066-3-11	WMPPPSQSVF	10	3.8642	A24	82	*	29-02	29	2
mCRC	103	103-1-40	QMSFLFHLI	9	1.3944	A24	1000	*	29-02	29	2
mCRC	159	159-1-36	KYACHQKDL	9	2.439	A24	2		29-02	29	2
mCRC	159	159-1-38	YYDVMPEEIE	10	2.3462	A24	14	*	29-02	29	2
mCRC	159	159-1-69	VFLIKPEDY	9	0.9145	A24	0.83		29-02	29	2
mCRC	159	159-2-16	NYFSRVIHV	9	4.0264	A24	3.5		29-02	29	2
mCRC	159	159-1-03	FFDDPVLLEL	10	3.245	A24	4.17		29-03	29	3
mCRC	159	159-1-22	LFPNQLHGM	9	3.363	A24	1.5		29-03	29	3
mCRC	159	159-1-25	KYACHQKDLF	10	2.8868	A24	283.83	*	29-03	29	3
mCRC	159	159-1-59	GTLAPFTI	9	1.3537	A24	2		29-03	29	3
mCRC	159	159-1-69	PVLDLLELPEL	9	0.3686	A24	2		29-03	29	3
mCRC	16	016-1-01	TLRGHLAEI	9	3.2459	A02	13.67		30-01	30	1
mCRC	16	016-1-08	QLVKTCPVQL	10	1.601	A02	12.5		30-01	30	1

mCRC	16	016-1-10	SIAYFQMDV	9	2.9131	A02	42.17	*	30-01	30	1
mCRC	16	016-1-11	KLVVGAAGV	10	2.5737	A02	26		30-01	30	1
mCRC	16	016-1-14	SLPSTTPEEL	10	2.9847	A02	16.17		30-01	30	1
mCRC	21	021-1-17	SVIKISVEPV	10	2.2267	A02	1023.17	*	30-01	30	1
mCRC	16	016-2-13	LVVVGAAGV	9	2.1795	A02	28.33		30-02	30	2
mCRC	16	016-2-16	FQMDVQVGET	10	1.571	A02	22.67		30-02	30	2
mCRC	21	021-1-22	IMHQDIVLL	9	4.1257	A02	55.83	*	30-02	30	2
mCRC	21	021-1-35	VIKISVEPV	9	0.8408	A02	867.67	*	30-02	30	2
mCRC	21	021-1-38	FTEELKYVI	9	0.7493	A02	15.17		30-02	30	2
mCRC	45	045-1-24	GLFCFLGSI	9	3.0188	A02	858.5	*	30-02	30	2
mCRC	21	021-1-43	TTSVIKISV	9	0.6132	A02	5.83		30-03	30	3
mCRC	45	045-1-08	MLQQFEDTL	9	3.7284	A02	5.17		30-03	30	3
mCRC	45	045-1-28	YMFAWTYVTL	10	3.8452	A02	994.5	*	30-03	30	3
mCRC	45	045-1-44	GLFCFLGSIV	10	2.0551	A02	660.17	*	30-03	30	3
mCRC	45	045-2-06	SMHGMPFSA	9	3.4632	A02	21.5	*	30-03	30	3
mCRC	45	045-2-76	HMELWMEQA	9	0.8821	A02	23		30-03	30	3
mCRC	45	045-1-16	KSEGLFCFL	10	1.9088	A02	22.33		30-04	30	4
mCRC	45	045-1-51	MLQQFEDTLV	10	2.8647	A02	13.83		30-04	30	4
mCRC	45	045-1-52	YMFAWTYVT	9	2.9908	A02	1000	*	30-04	30	4
mCRC	45	045-1-98	FLVKTGYTFV	10	3.7888	A02	105.33	*	30-04	30	4
mCRC	45	045-2-68	SMHGMPFSAM	10	1.7747	A02	18.5		30-04	30	4
mCRC	61	061-1-24	AFVEEIFEA	9	3.1629	A02	25.5		30-04	30	4
mCRC	45	045-1-86	FLVKTGYTF	9	3.4661	A02	99	*	31-01	31	1
mCRC	61	061-1-02	FAIGFSYPV	9	3.6921	A02	1000	*	31-01	31	1
mCRC	61	061-1-04	FVEEIFEAV	9	4.0742	A02	28		31-01	31	1
mCRC	61	061-1-40	HLSFFIHGC	9	1.5994	A02	400.33	*	31-01	31	1
mCRC	61	061-2-05	KLEENLAKV	9	4.2273	A02	24.33		31-01	31	1
mCRC	61	061-2-41	MTIEELLTC	9	1.0624	A02	273.67	*	31-01	31	1
mCRC	61	061-1-37	FAIGFSYPVV	10	1.1675	A02	1000	*	31-02	31	2
mCRC	61	061-1-45	AFVEEIFEAV	10	1.4174	A02	13		31-02	31	2
mCRC	61	061-2-33	KLEENLAKVF	10	1.5886	A02	25.67	*	31-02	31	2
mCRC	66	066-1-02	RLTNVVRQV	9	3.9341	A02	39.17	*	31-02	31	2
mCRC	66	066-3-10	KIKDLMNTL	9	3.281	A02	11.17		31-02	31	2
mCRC	66	066-3-19	TLFAAYAPA	9	3.3766	A02	31.67	*	31-02	31	2
mCRC	61	061-1-56	RAFVEEIFEA	10	1.0358	A02	34.83		31-03	31	3
mCRC	66	066-1-04	FQPGDNLEV	9	3.7892	A02	8.5		31-03	31	3
mCRC	66	066-3-06	WMPPPSQSV	9	3.9605	A02	612.83	*	31-03	31	3
mCRC	66	066-3-13	FILSARVVL	9	3.221	A02	467.83	*	31-03	31	3
mCRC	66	066-3-21	ILCTASSSV	9	3.2959	A02	237	*	31-03	31	3
mCRC	66	066-3-22	DLMNTLFAA	9	2.7962	A02	7.17		31-03	31	3
HCC	117	117-1-20	MTDSKISSM	9	2.9797	B7	25.8		32-01	32	1
HCC	117	117-1-37	NLRRATTYL	9	3.0579	B7	19.5		32-01	32	1
HCC	117	117-1-82	LMRRQMATM	9	2.3625	B7	158.3	*	32-01	32	1
HCC	117	117-2-02	VARLLNVNL	9	3.8312	B7	193.8	*	32-01	32	1
HCC	117	117-2-03	VPRRVIHVV	9	4.1911	B7	1000	*	32-01	32	1
HCC	117	117-2-18	LPYQQLSTT	9	1.969	B7	26.5		32-01	32	1
HCC	117	117-2-42	LPRWSGFRST	10	3.1211	B7	1006.5	*	32-01	32	1
HCC	117	117-2-51	IPLACRIRY	9	2.6257	B7	62		32-01	32	1
HCC	142	142-2-32	VPAARLHGPL	10	3.9717	B7	1035.3	*	32-01	32	1
mCRC	164	164-1-28	GPGPSPVTEA	10	3.0102	B7	26.8		32-01	32	1
mCRC	16	016-1-03	LVKTCVQQL	9	2.5968	B7	5.8		32-02	32	2
mCRC	16	016-1-19	VIDGESKVM	9	1.0529	B7	12.3		32-02	32	2
mCRC	16	016-2-09	LPSTTPEEL	9	4.0796	B7	24	*	32-02	32	2
HCC	142	142-2-10	SVLPEGAAL	9	3.8929	B7	9.7		32-02	32	2
HCC	142	142-2-21	LLRRQHRAL	9	3.4542	B7	10		32-02	32	2
HCC	142	142-2-36	GPAARVGQA	9	3.6354	B7	15.5		32-02	32	2
HCC	142	142-2-40	HAAKGLVPA	10	3.4835	B7	905	*	32-02	32	2
HCC	142	142-2-49	CPEQGHPAA	9	3.0154	B7	8		32-02	32	2
HCC	142	142-2-51	RAREGPAARV	10	2.8584	B7	6.7		32-02	32	2
HCC	142	142-2-55	IPETPGKENV	10	2.9099	B7	6		32-02	32	2
mCRC	40	040-1-46	GADGVGKSAL	10	1.9883	B7	59.7		32-03	32	3
mCRC	40	040-1-51	APHIHRWTL	10	4.0601	B7	975.2	*	32-03	32	3
mCRC	40	040-2-06	KPSSPGISF	9	4.2233	B7	306.3	*	32-03	32	3
mCRC	40	040-2-20	SPGISFGSSI	10	3.0946	B7	61.7		32-03	32	3
mCRC	40	040-2-22	LPYDDCLEA	9	3.0423	B7	31.7		32-03	32	3
mCRC	40	040-2-33	TPLSGLTDHL	10	3.8011	B7	42.5		32-03	32	3
mCRC	40	040-2-47	LSHVPRGHL	9	2.0405	B7	52.8		32-03	32	3
mCRC	40	040-2-72	FPCGAVFEEI	10	0.6698	B7	40.8		32-03	32	3
mCRC	164	164-1-07	SPPDPWGCL	9	4.0047	B7	156.5	*	32-03	32	3
mCRC	164	164-1-08	STKPGTLSA	9	3.0477	B7	35		32-03	32	3
mCRC	164	164-1-14	GPSPVTEAL	9	4.2147	B7	889.5	*	32-03	32	3
mCRC	40	040-1-52	APHIHRWTI	9	4.0204	B7	1079.5	*	32-04	32	4
mCRC	40	040-2-73	LPVGRGCTK	9	0.6341	B7	10		32-04	32	4
HCC	117	117-1-77	VPRRVIHVVS	10	2.0246	B7	284.2	*	32-04	32	4
mCRC	164	164-1-12	KPGTLSAPF	9	3.9494	B7	306.5	*	32-04	32	4
mCRC	164	164-1-18	EPNYFSRVI	9	3.4519	B7	10.8		32-04	32	4
mCRC	164	164-1-26	NPSLRENYF	9	3.1239	B7	22		32-04	32	4

mCRC	164	164-1-27	PGPSPVTEAL	10	3.0817	B7	17.2	32-04	32	4	
mCRC	164	164-1-29	EPVLDLLPEL	10	2.7248	B7	11.7	32-04	32	4	
mCRC	164	164-1-30	FPNQLHGMDM	10	2.8955	B7	10.8	32-04	32	4	
mCRC	164	164-1-31	EPGCETTGL	9	2.7334	B7	22.7	32-04	32	4	
mCRC	164	164-2-19	LASTKPGTL	9	3.1167	B7	10	32-04	32	4	
HCC	127	127-1-08	EPHSGLLLL	9	3.9941	B35	125.5	*	39-01	39	1
HCC	127	127-1-21	QPTVRITQL	9	3.4689	B35	13.67		39-01	39	1
HCC	127	127-1-26	VALETLRAW	9	3.11	B35	23.83		39-01	39	1
HCC	127	127-1-31	DAQQLIRLL	9	2.577	B35	10.5		39-01	39	1
HCC	127	127-1-32	QFFPEAVSF	9	3.4305	B35	227.17	*	39-01	39	1
HCC	127	127-1-38	GPGLALHCM	9	2.8512	B35	259.83	*	39-01	39	1
HCC	127	127-1-54	TVDTIDSCM	9	2.0908	B35	10.83		39-01	39	1
HCC	127	127-1-61	ECTLSGVKY	9	2.2491	B35	9.17		39-01	39	1
HCC	127	127-2-19	HPTININGY	9	4.2068	B35	1000	*	39-01	39	1
HCC	134	134-1-15	FLWSAQCSF	9	2.9219	B35	1000	*	39-01	39	1
HCC	134	134-2-23	FEYPNTISFF	10	2.1503	B35	1000	*	39-01	39	1
HCC	134	134-2-65	LPLGLRDRA	9	0.4133	B35	11.83		39-01	39	1
HCC	127	127-1-42	PPFVALETL	9	2.4248	B35	3.5		39-02	39	2
HCC	127	127-2-39	SAANAGNSF	9	4.0742	B35	5.5		39-02	39	2
HCC	127	127-2-68	SPHSLDIREF	10	3.8775	B35	20.5	*	39-02	39	2
HCC	134	134-1-08	MSYDHAITVF	10	3.8107	B35	98.17	*	39-02	39	2
HCC	134	134-1-32	LACPEAQLL	9	1.8304	B35	72.67	*	39-02	39	2
HCC	134	134-1-53	WESLRFSEY	9	1.0262	B35	50.33	*	39-02	39	2
HCC	134	134-2-02	YPNTISFFL	9	4.1572	B35	1000	*	39-02	39	2
HCC	134	134-2-03	LPEALPLGL	9	3.9684	B35	313	*	39-02	39	2
HCC	134	134-2-30	TPVCPKMSL	9	3.8391	B35	348.5	*	39-02	39	2
HCC	134	134-2-70	WSAQCSFGF	9	0.6423	B35	3.17		39-02	39	2
HCC	135	135-1-04	TPVINYDLVY	10	4.1419	B35	1000	*	39-02	39	2
HCC	135	135-2-22	APGRRPLTM	9	3.4247	B35	225	*	39-02	39	2
HCC	135	135-1-15	YLEPEKQVM	9	2.8446	B35	15.33		39-03	39	3
HCC	135	135-1-25	IASVVENTI	9	1.8838	B35	8.5		39-03	39	3
HCC	135	135-2-18	LAHIPYQEL	9	3.0349	B35	40.83	*	39-03	39	3
HCC	135	135-2-21	MAAPGRRPL	9	3.4966	B35	6.67		39-03	39	3
HCC	135	135-2-28	HPRPSFWTS	9	2.9885	B35	18.33		39-03	39	3
HCC	137	137-1-25	VPKKGALDL	9	3.2099	B35	8.83		39-03	39	3
HCC	137	137-1-35	TPYEPGGQSV	10	3.3991	B35	5.33		39-03	39	3
HCC	137	137-2-09	SAGGRTDSF	9	3.5512	B35	4.33		39-03	39	3
HCC	137	137-2-19	SPNVNLVY	9	4.2013	B35	799.33	*	39-03	39	3
HCC	137	137-2-31	SPWAPGPAS	9	3.3139	B35	94.17	*	39-03	39	3
HCC	137	137-2-32	LPQATGETFF	10	3.4709	B35	635.5	*	39-03	39	3
HCC	135	135-2-20	RPLTMLPQL	9	3.1544	B35	7		39-04	39	4
HCC	137	137-2-12	HSPNVNLVY	10	3.9769	B35	1000	*	39-04	39	4
HCC	147	147-1-06	SGLTAISTF	9	3.0492	B35	3		39-04	39	4
HCC	147	147-1-07	HIKDAPEEF	9	3.7203	B35	24.83	*	39-04	39	4
HCC	147	147-1-135	CVESLIAVF	9	1.879	B35	5		39-04	39	4
HCC	147	147-1-146	YDYDYGEDF	9	2.5957	B35	6.33		39-04	39	4
HCC	147	147-2-11	SPEPAGRRVY	10	3.7632	B35	583.17	*	39-04	39	4
HCC	147	147-2-132	LPALCAHPH	9	2.1979	B35	1000	*	39-04	39	4
HCC	147	147-2-156	WPAGELHAK	9	2.5284	B35	1.83		39-04	39	4
HCC	147	147-2-17	PPLKSVAEF	9	3.7176	B35	23.83		39-04	39	4
HCC	147	147-2-34	TAALPASII	9	4.1558	B35	1000	*	39-04	39	4

Table S5. Sequences of long peptides (Lp) and short peptides shown in Figure 4.

Long peptides		Including short peptides	
Peptide_ID	Peptide sequences	Peptide_ID	Peptide sequences
129-Long-9	Peptide: EHSLQME AYERRIQRL EQEKLELSRKL Mouse: ENSLQIEAYERRIRRL EQEKLELSRKL	129-1-10	AYERRIQRL
129-Long-29	Peptide: PIKEWLNSCDL KYT-GVQSLNWT KIMKT Mouse: PIKEWLNSCDLKYTEGVQSLNWT KIMKT	129-3-22	KYT-GVQSLNW
129-Long-18	Peptide: DL KYT-GVQSLNWT KIMKTIVDDPEGFF Mouse: DLKYTEGVQSLNWT KIMKTIVDDPEGFF	129-3-22	KYT-GVQSLNW
129-Long-14	Peptide: VFEVGP GDSP TFPQFRAIQL EPAEPGQ Mouse: VFEVGA GDSP TFPRFRAIQL EP TESGO	129-1-16	TFPQFRAIQL
117-Long-15	Peptide: VAKLEPPLIPF MPFLIKDMTF THEGNK Mouse: AAKLEPPLIPF MPFLIKDMTF THEGNK	117-2-07	PFLIKDMTF
117-Long-21	Peptide: GAYLIDRDPT TYFGTILNFL RHGKLVLD Mouse: GAYLIDRDPT TYFGPILNFL RHGKLVLD	117-1-01 117-1-14	TYFGTILNF TYFGTILNFL

Bold: short neopeptides; red: mutations vs. wild type; blue: mutations between human and mouse.

Figure S1. Reactivity of patient autologous CD8⁺ T cells against predicted neopeptides.

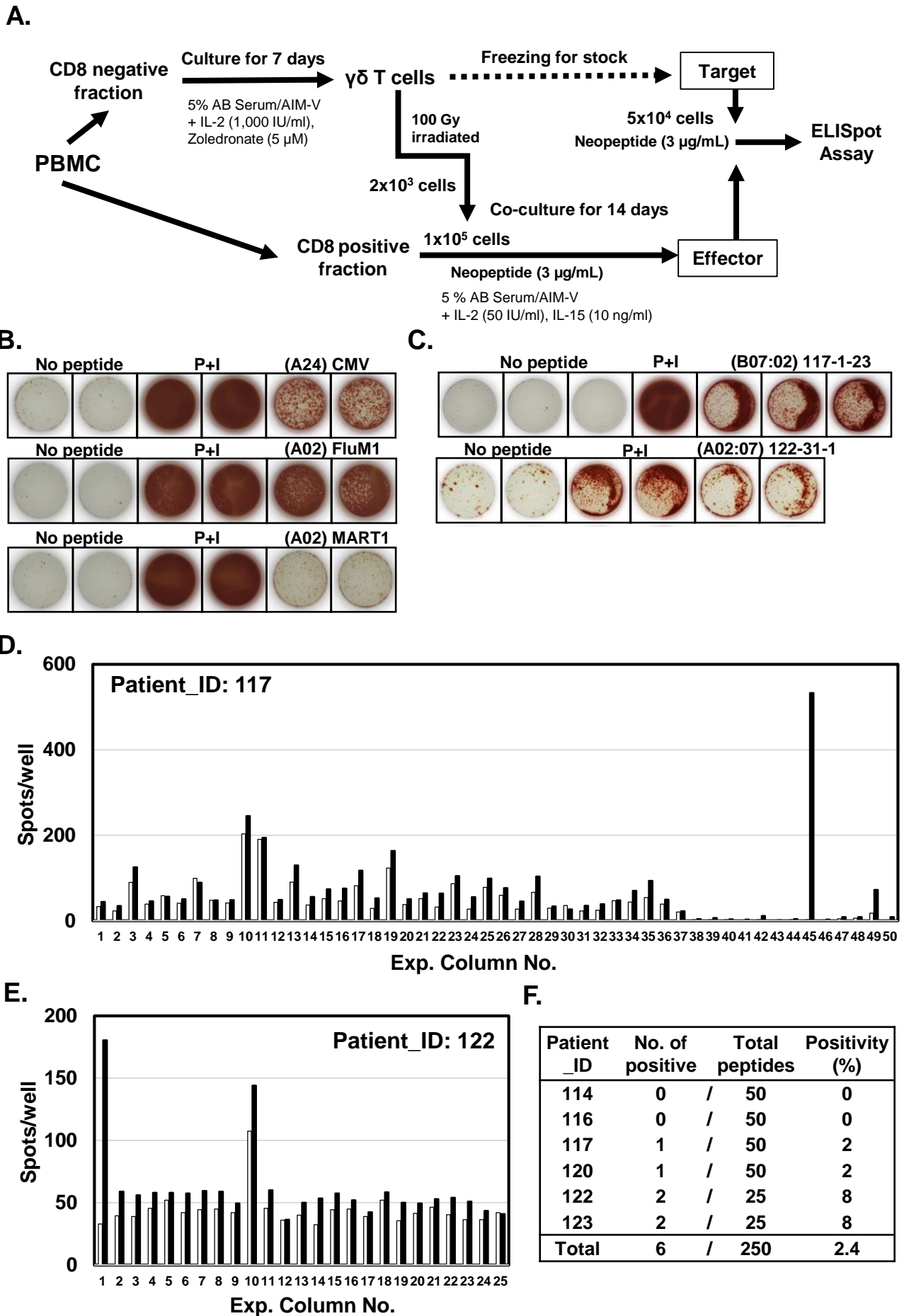
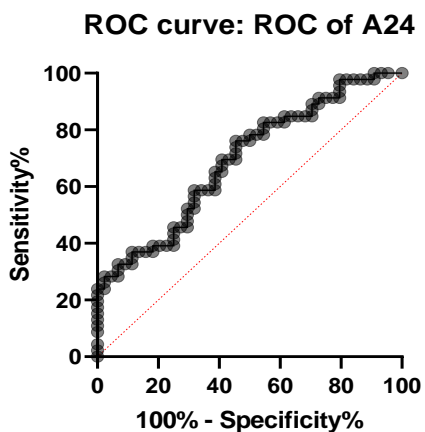
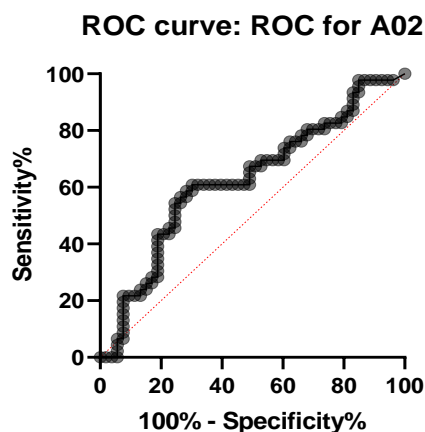


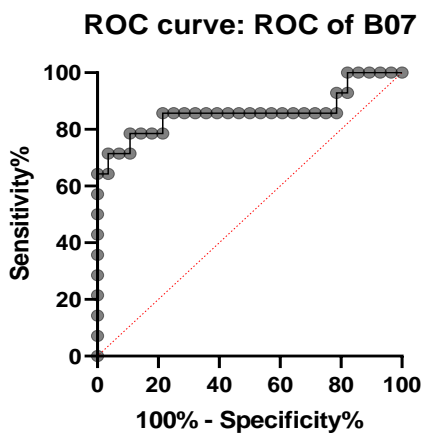
Figure S2. Receiver operating characteristic (ROC) curves demonstrating sensitivity of each prediction of specificity for each HLA haplotype.



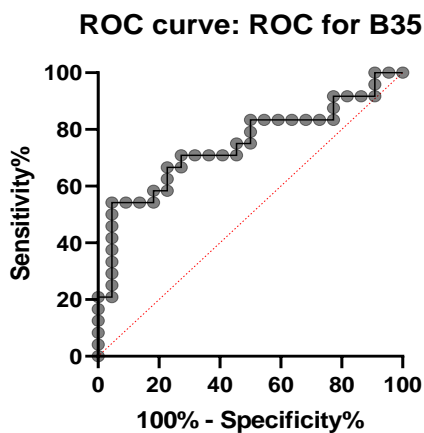
Area	0.6922
Std. Error	0.05494
95% confidence interval	0.5845 to 0.7999
P value	0.0017



Area	0.6259
Std. Error	0.05704
95% confidence interval	0.5141 to 0.7377
P value	0.0313



Area	0.8597
Std. Error	0.07645
95% confidence interval	0.7099 to 1.000
P value	0.0002



Area	0.7462
Std. Error	0.07402
95% confidence interval	0.6011 to 0.8913
P value	0.0043