

Supplementary Table 1. Human cancer cell lines

Cell Line	HLA status	MAGE A1 status	Cancer Type
ME275	HLA-A2 ⁺	MAGE-A1 ⁺	Melanoma
A375	HLA-A2 ⁺	MAGE-A1 ⁺	Melanoma
A375F	HLA-A2 ⁺ overexpressed	MAGE-A1 ⁺ overexpressed	Melanoma
SK-MEL37	HLA-A2 ⁺	MAGE-A1 ⁺	Melanoma
PANC-1	HLA-A2 ⁺	PRAME ⁺	Pancreatic cancer
SW-480	HLA-A2 ⁺ overexpressed	WT1 ⁺	colon adenocarcinoma

Supplementary Table 2: Adverse events experienced following CD4+ CD8+ T_{TCR-MA1-CD8αβ} infusion product in a subject with non-small cell lung cancer.

BODY SYSTEM (CTC/CTCAE CATEGORY)	EVENT	# Subjects Affected	Maximal Grade	Maximal Attribution
Cardiac disorders	Sinus tachycardia	1	1	Possible
Gastrointestinal Disorders	Bloating	1	1	Not related
General disorders and administration site conditions	Fatigue	1	2	Possible
Metabolism and nutrition disorders	Hyperglycemia	1	1	Not related
	Hyperphosphatemia	1	1	Unlikely
Musculoskeletal and connective tissue disorders	Generalized muscle weakness	1	2	Not related
Nervous system disorders	Dizziness	1	1	Unlikely
Psychiatric disorders	Insomnia	1	1	Unlikely
Respiratory, thoracic and mediastinal disorders	Hoarseness	1	1	Not related
skin and subcutaneous tissue disorders	Dry skin	1	1	Not related

Supplementary Table 3: Curated genes for costimulatory ligands on patient scRNA seq dataset

Costim	Inhibitory
CD80	CD276
CD86	CD274
CD40	PDCD1LG2
TNFRSF4	LGALS9
ICOSLG	TGFB1
TNFSF9	VTCN1

Supplementary Table 4: CD8 α chain modification vs CD8 β chain modification

	Name	CCR molecule	Transgene ECD	Transgene 1 TM	Transgene 1 ICD	Transgene 2 ECD	Transgene 2 TM	Transgene 2 ICD	CD8 α modified	CD8 β modified
With CD8 α chain modified	C	CD8a*28CD8b*28	CD8a	CD8a	CD28	CD8b	CD8b	CD28	Yes	Yes
	F	CD8a*28mtCD8b*28mt	CD8a	CD8a	CD28 partial mutant	CD8b	CD8b	CD28 partial mutant	Yes	Yes
	N	CD8a_bSTALK_28tm_28IC	CD8a-CD8bstalk	CD28	CD28	x	x	x	Yes	N/A
	O	CD8a_bSTALK_28Tm_28IC_aIC	CD8a-CD8bstalk	CD28	CD28 + CD8a	x	x	x	Yes	N/A
	P	CD8a_bSTALK_aTm_aIC	CD8a-CD8bstalk	CD8a	CD8a	x	x	x	Yes	N/A
	T	CD8-28 full mutant	CD8a	CD8a	CD28 full mutant	CD8b	CD8b	CD28 full mutant	Yes	Yes
With CD8 β chain modified	A	CD8aCD8b6*28	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +CD28	No	Yes
	E	CD8aCD8b*28mt	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b + CD28 full mutant	No	Yes
	G	CD8a_CD8-CD28	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +CD28wt	No	Yes
	H	CD8a_CD8_41bb	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +41BB	No	Yes

Supplementary Table 5: CD8ab chimeric co-receptor constructs

Identifier	Name	Transgene 1 design			Transgene 2 design			CD8a modified	CD8b modified
		extracellular	tm	intracellular	extracellular	tm	intracellular		
CD8/CD28mut	CD8a_ CD8b ECD-CD28mut ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6AA CD8b + CD28 full mutant	No	Yes
CD8/CD28	CD8a_ CD8b ECD-CD28 ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +CD28wt	No	Yes
CD8/41BB	CD8a_ CD8b ECD _41bb ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +41BB	No	Yes
CD8/ICOS	CD8a_ CD8b ECD _ICOS	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +ICOS	No	Yes
CD8/OX40	CD8a_ CD8b ECD _OX40 ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +OX40	No	Yes
CD8/GITR	CD8a_ CD8b ECD _GITR ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +GITR	No	Yes
CD8ab	TCRM0_CD8ab	CD8a	CD8a	CD8a	CD8b	CD8b	CD8b	No	No
TCR-MA1	TCR alone							N/A	N/A
Irr.CD8ab	Irrelevant TCR + CD8ab	CD8a	CD8a	CD8a	CD8b	CD8b	CD8b	No	No
Irr-CD8/CD28	Irrelevant TCR + CD8a_ CD8b-CD28	CD8a	CD8a	CD8a	CD8b	CD8b	6 AA CD8b +CD28wt	No	Yes
Irr.-CD8/CD28mut	Irrelevant TCR + CD8a_ CD8b ECD-CD28mut ICD	CD8a	CD8a	CD8a	CD8b	CD8b	6AA CD8b + CD28 full mutant	No	Yes

Supplementary Table 6: CD8-based costimulatory molecule library

Identifier	Transgene 1	Transgene 2			
		Extrac- ellular	tm	intracellular	Intracellular domain sequence
CD8/CD28mut	CD8a	CD8b	CD8b	6 AA CD8b +CD28GG mut	HLCCRRRSKRSRGGHSDAMNMTARRAGPTRKHYQAYAAPRDFAAYRS
CD8/CD28	CD8a	CD8b	CD8b	6 AA CD8b +CD28wt	HLCCRRRSKRSRLLHSDYMNMTPRRPGPTRKHYQPYAPPRDFAAYRS
CD8/41BB	CD8a	CD8b	CD8b	6AA CD8b +4- 1BB	HLCCRRKRGRKLLYIFKQPFMRPVQTTQEEDGCSCRFPEEEEGGCEL
CD8/ICOS	CD8a	CD8b	CD8b	6AA CD8b +ICOS	HLCCRRKYSSSVHDPNGEYMFMRVNTAKKSRLTDVT
CD8/OX40	CD8a	CD8b	CD8b	6AA CD8b +OX40	HLCCRRALYLLRRDQRLPPDAHKKPPGGGSFRTPIQEEQADAHSTLAKI
CD8/GITR	CD8a	CD8b	CD8b	6AA CD8b +GITR	HLCCRRQLGLHIWQLRSQCMWPRETQLLLEVPSTEDARSCQPPEERGERSAEEKGRLGDL WV
CD8ab	CD8a	CD8b	CD8b	CD8b	HLCCRRRRARLRFMKQFYK

Supplementary Table 7: CD28 motif mutation variation

no.	CD28 Chimeric co-receptor	Di-leucine motif	PI3K and GRB2/GADS binding site	ITK and GADS binding site	LCK binding site
A0	CD8αβ-CD28 wt (TCR _{MA1-CD8/CD28})	LL	YMNM	PRRP	PYAP
A1	CD8αβ-CD28 m1-1	GG	YMNM	PRRP	PYAP
A2	CD8αβ-CD28 m1-2	LL	AMNM	PRRP	PYAP
A3	CD8αβ-CD28 m1-3	LL	YMNM	ARRA	PYAP
A4	CD8αβ-CD28 m1-4	LL	YMNM	PRRP	AYAA
A5	CD8αβ-CD28 m2-1	GG	AMNM	PRRP	PYAP
A6	CD8αβ-CD28 m2-2	GG	YMNM	ARRA	PYAP
A7	CD8αβ-CD28 m2-3	GG	YMNM	PRRP	AYAA
E0	CD8αβ-CD28 m2-4	LL	AMNM	ARRA	PYAP
E1	CD8αβ-CD28 m2-5	LL	AMNM	PRRP	AYAA
E2	CD8αβ-CD28 m2-6	LL	YMNM	ARRA	AYAA
E3	CD8αβ-CD28 m3-1	GG	AMNM	ARRA	PYAP
E4	CD8αβ-CD28 m3-2	GG	AMNM	PRRP	AYAA
E5	CD8αβ-CD28 m3-3	GG	YMNM	ARRA	AYAA
E6	CD8αβ-CD28 m3-4	LL	AMNM	ARRA	AYAA
E7	CD8αβ-CD28 mut (TCR _{MA1-CD8/CD28mut})	GG	AMNM	ARRA	AYAA

Supplementary Table 8: Proliferation and Exhaustion gene list for bulk RNA seq dataset

Proliferation	Exhaustion
MKI67	PDCD1
PCNA*	LAG3
MCM2*	HAVCR2
MCM3	CTLA4*
MCM4	TIGIT*
MCM5*	CD160
MCM6*	BTLA
MCM7	ENTPD1
CCND1	TOX
CCND2	PDCD2
CCND3	
CDK1	
CDK2	
CDK4	
CDK6	
CDC6	
CDC45	
TOP2A	
AURKA	
AURKB	
TUBB	

* significantly differentially expressed