

Supplemental Figures and Tables

Figure S1

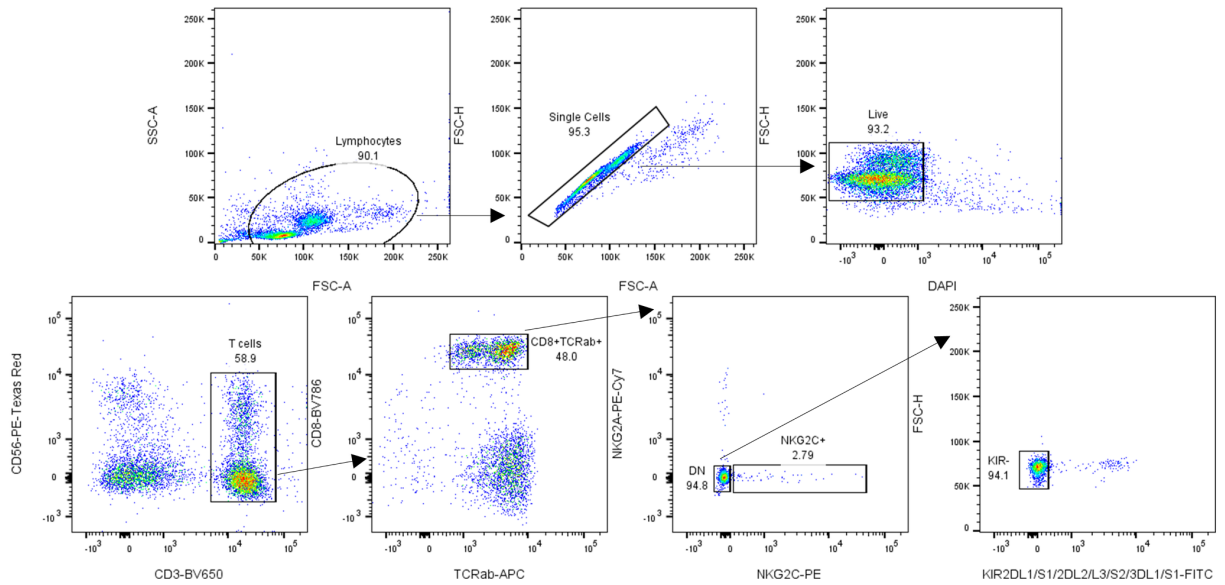


Figure S1. Flow cytometry gating strategy for FACS-sorted NKG2C⁺ T_{KLR} cells and CD8 T_{Conv} cells.

Figure S2

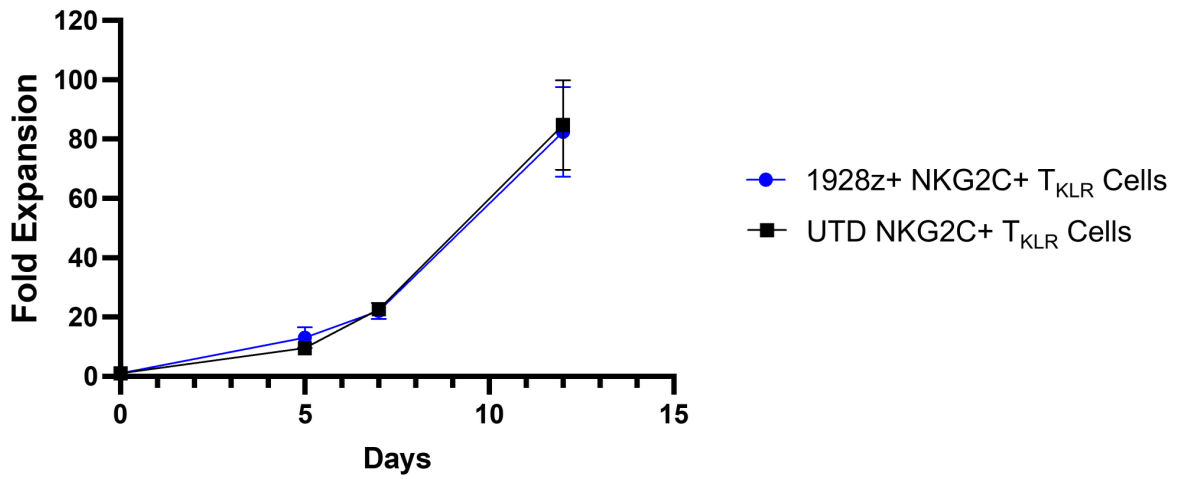


Figure S2. Fold ex vivo expansion of 1928z⁺ NKG2C⁺ T_{KLR} cells and UTD NKG2C⁺ T_{KLR} cells during co-culture with K562 feeder cells. Data are presented as mean values +/- SEM. Statistical analysis was performed by multiple two-tailed unpaired t tests (n=3 independent experiments).

Figure S3

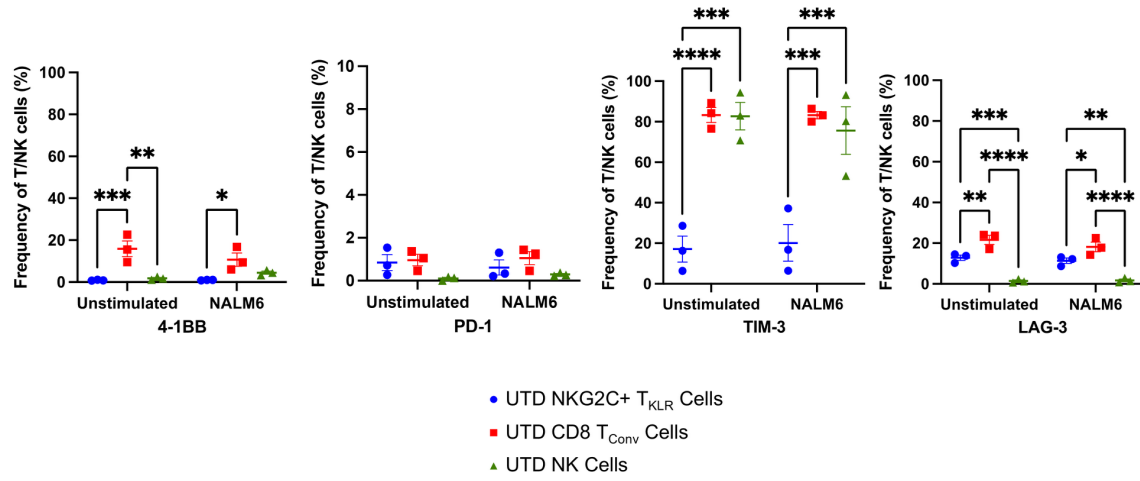


Figure S3. Flow cytometry phenotyping of UTD T and NK cells with or without 8-hour stimulation with CD19⁺ NALM6 target cells. Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by two-way ANOVA (n=3 independent experiments).

Figure S4

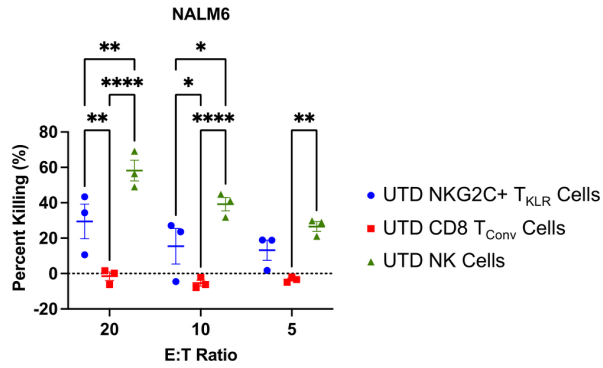


Figure S4. Cytotoxicity of UTD T and NK cells against CD19⁺ NALM6 target cells. Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by two-way ANOVA (n=3 independent experiments).

Figure S5

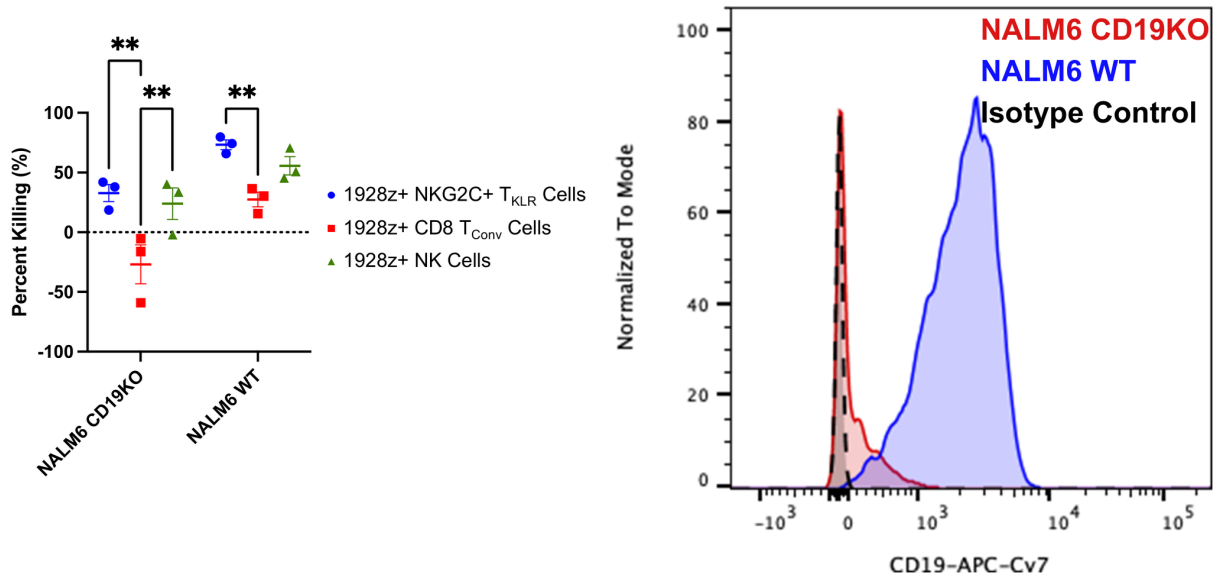


Figure S5. Cytotoxicity of 1928z⁺ T and NK cells against CD19⁺ NALM6 WT and NALM6 CD19KO target cells at a 5:1 E:T ratio. Data are presented as mean values +/- SEM. Statistical analysis was performed by two-way ANOVA (n=3 independent experiments).

Figure S6

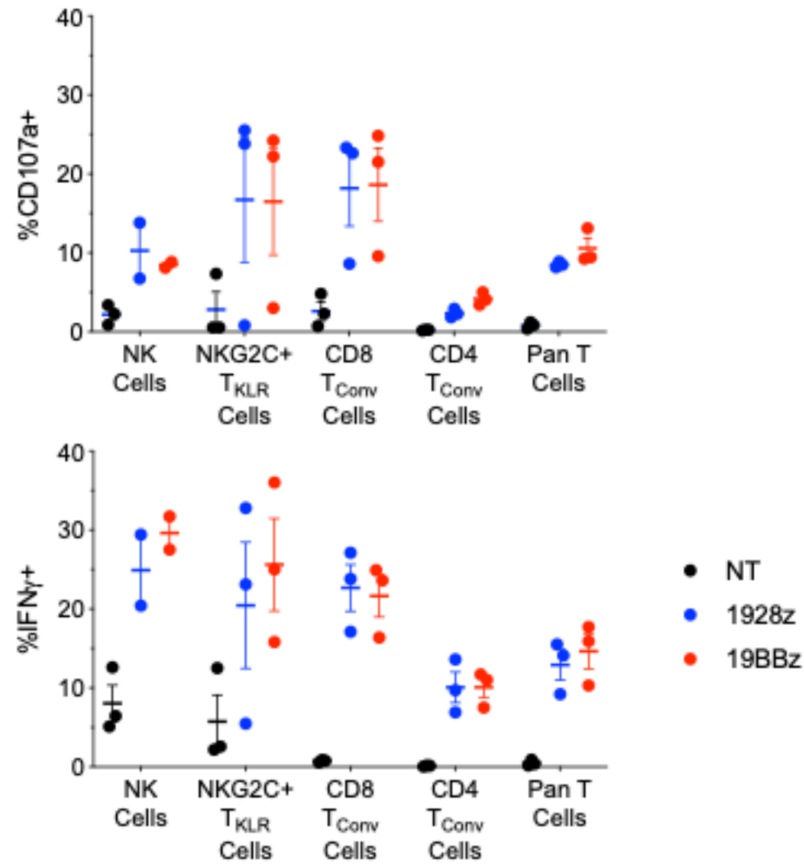
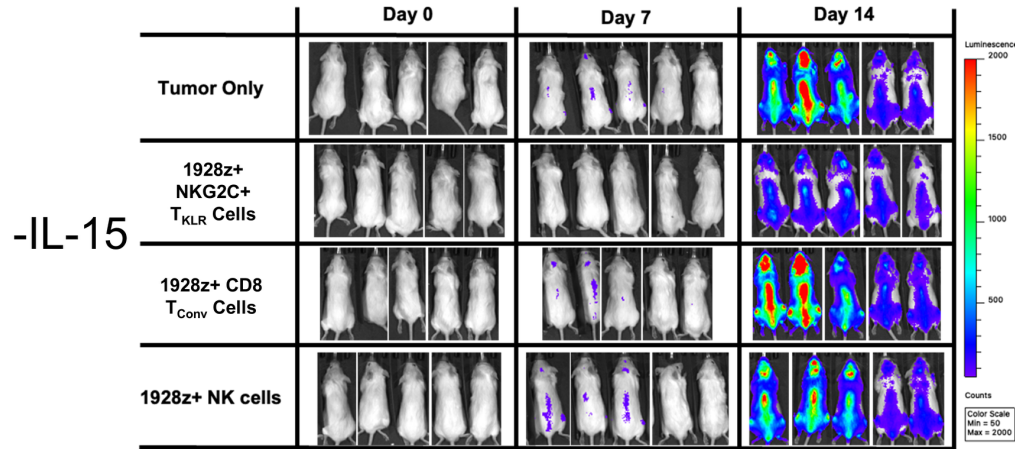


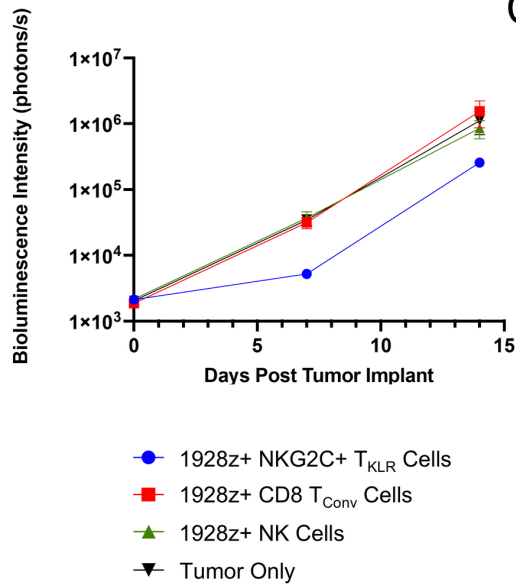
Figure S6. Degranulation (CD107a) and IFN γ production in NK and T cells expressing CD28- or 41BB-based CARs following 4-hour coculture with CD19⁺ NALM6 tumor targets. Data are presented as mean values $\pm$ SEM. Wilcoxon matched-pairs signed rank test performed between 1928z and 19BBz of paired donors for each population, no statistically significant difference found.

Figure S7

A



B



C

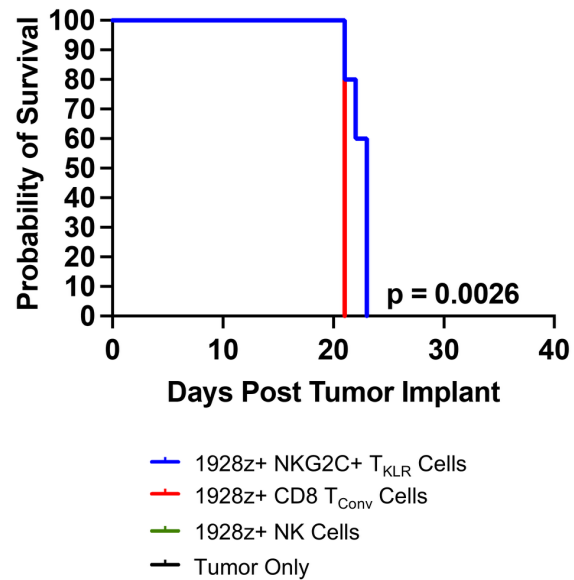


Figure S7. (A) Bioluminescence imaging of mice treated with tumor only, 1928z⁺ NKG2C⁺ T_{KLR} cells, 1928z⁺ CD8 T_{Conv} cells, and 1928z⁺ NK cells without IL-15, measured throughout the course of the study. **(B)** Quantification of bioluminescence imaging of mice treated with tumor only, 1928z⁺ NKG2C⁺ T_{KLR} cells, 1928z⁺ CD8 T_{Conv} cells, and 1928z⁺ NK cells, measured throughout the course of the study; two-way ANOVA performed. **(C)** Kaplan-Meier survival plot of mice

treated with tumor only, 1928z⁺ NKG2C⁺ T_{KLR} cells, 1928z⁺ CD8 T_{Conv} cells, and 1928z⁺ NK cells, measured throughout the course of the study. Data are presented as mean values +/- SEM.

Figure S8

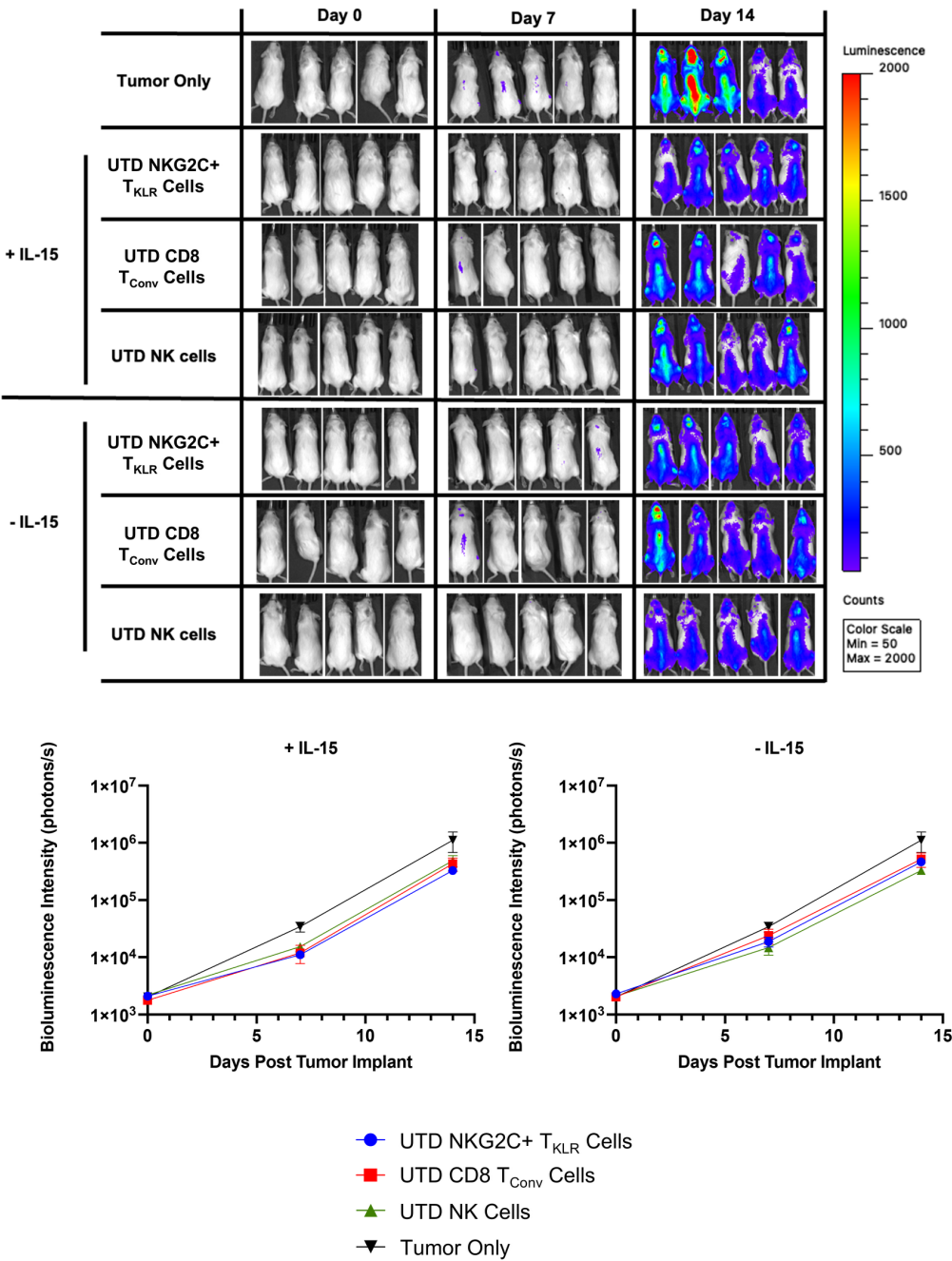


Figure S8. Bioluminescence imaging of mice treated with tumor only, UTD NKG2C⁺ T_{KLR} cells, UTD CD8 T_{Conv} cells, and UTD NK cells, measured throughout the course of the study (left). Quantification of bioluminescence imaging of mice, measured throughout the course of the study

(right). Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by two-way ANOVA (n=5 mice/group). No statistically significant difference found.

Figure S9

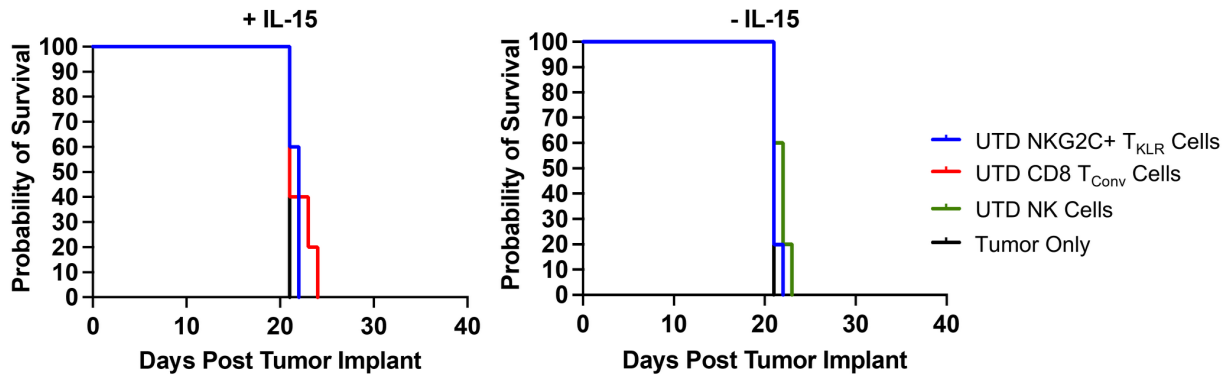


Figure S9. Kaplan-Meier survival plot of tumor only, and UTD T and NK cell groups treated with or without IL-15 co-administration. Statistical analysis was performed by Kaplan-Meier survival analysis (n=5 mice/group). No statistically significant difference found.

Figure S10

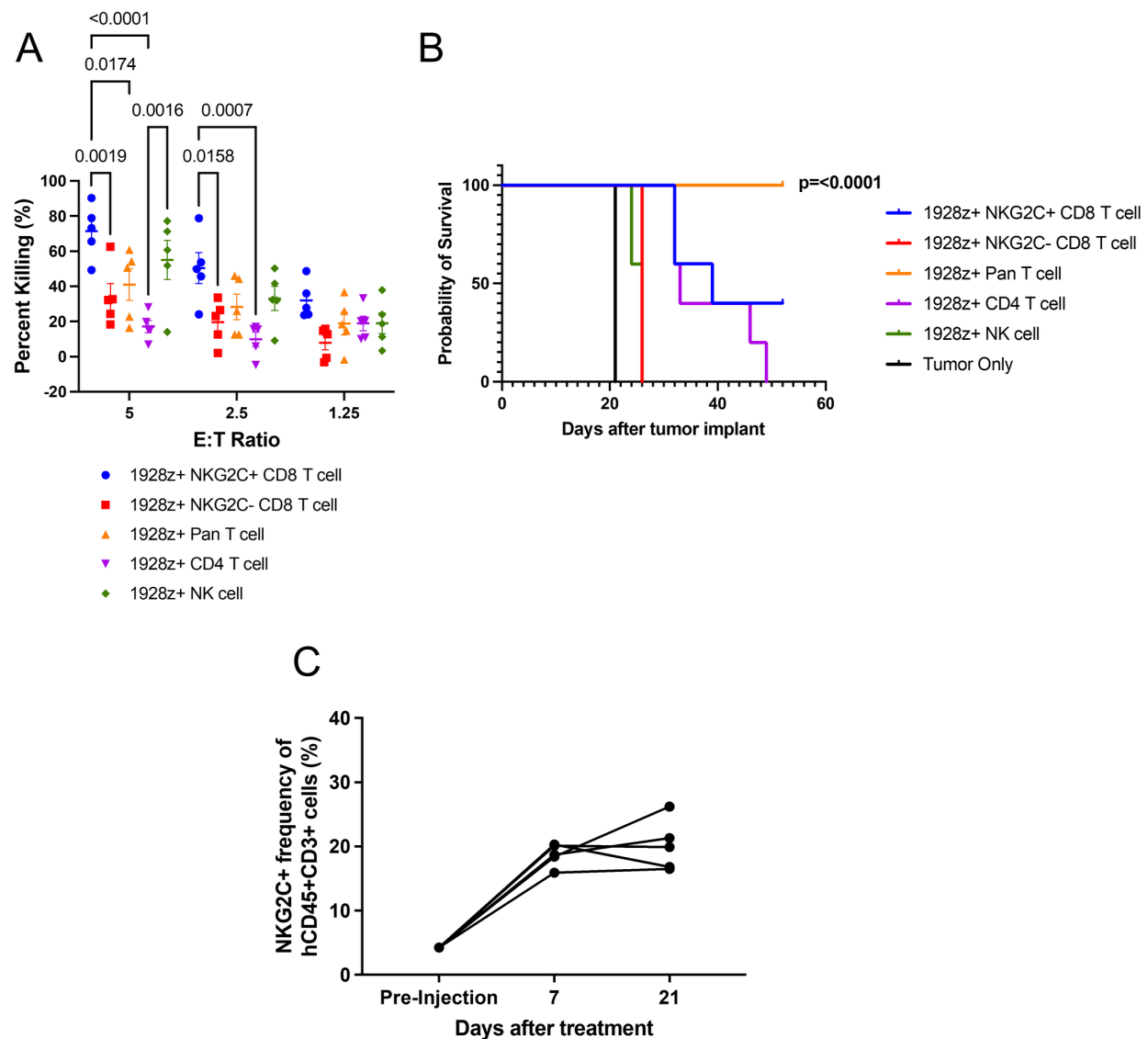


Figure S10. (A) Cytotoxicity of CAR⁺ T and NK cells against CD19⁺ NALM6 target cells. Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by two-way ANOVA. (B) Kaplan-Meier survival plot of NALM6 tumor-bearing mice treated with CAR⁺ T or NK cells (n=5 mice/group). (C) Frequency of NKG2C⁺ cells among human T cells in the 1928z⁺ Pan T pre-infusion product and in circulation afterward.

Figure S11

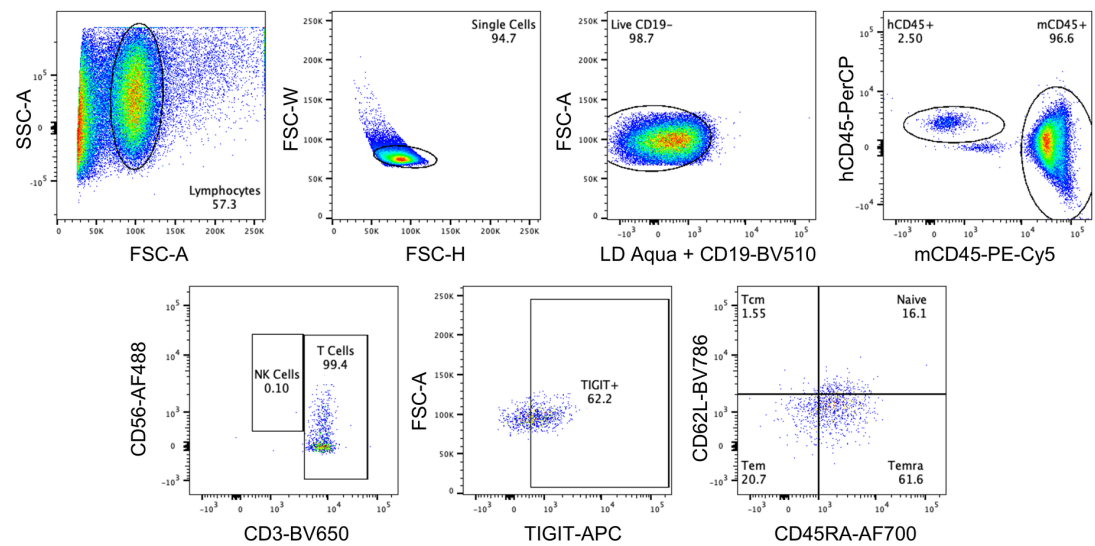


Figure S11. Flow cytometry gating strategy of T and NK cells collected from NALM6 or A375 tumor-bearing mice.

Figure S12

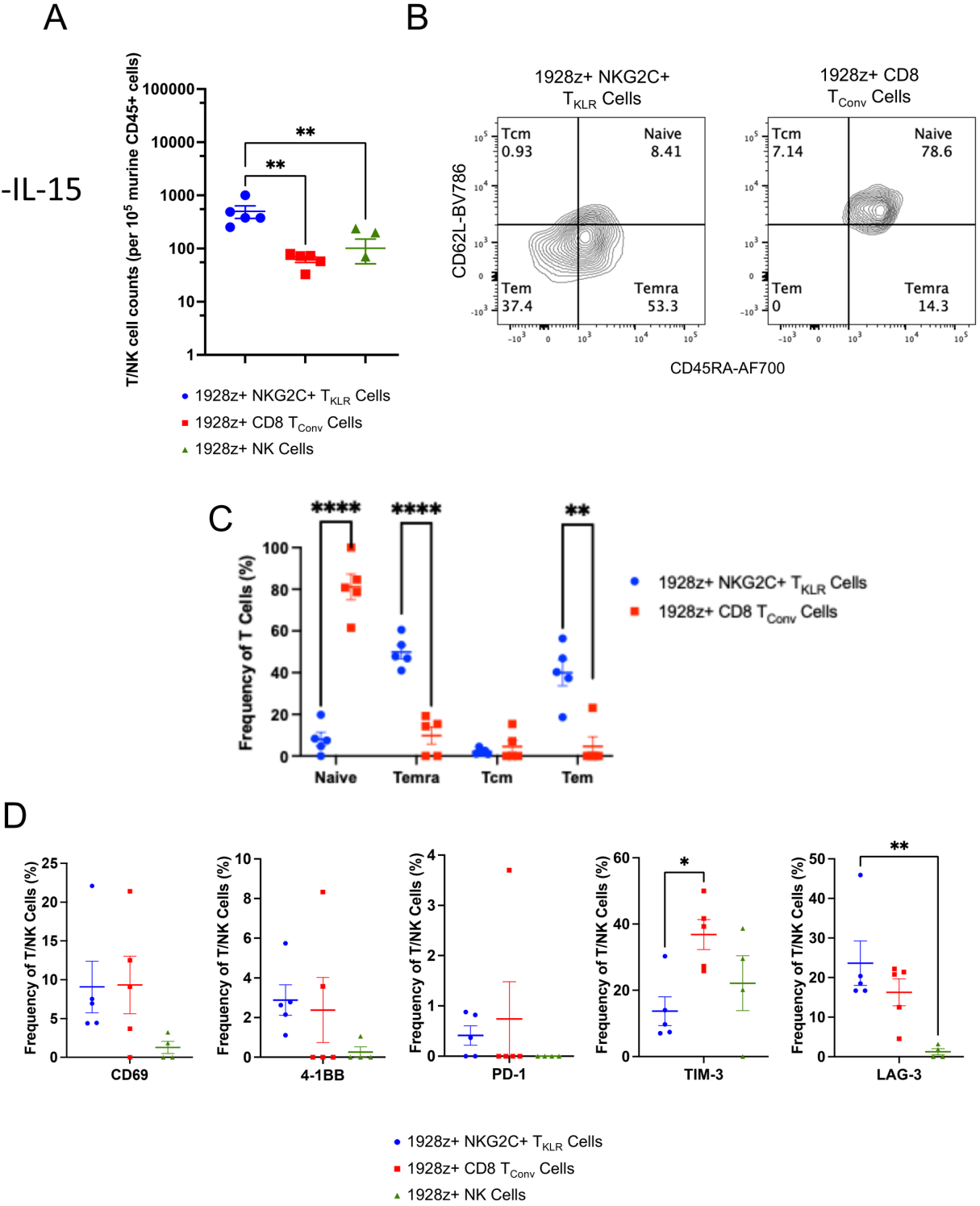


Figure S12. (A) Quantification of circulating CAR⁺ NKG2C⁺ T_{KLR} cells, CAR⁺ CD8 T_{Conv} cells, and CAR⁺ NK cells without IL-15 at day 7 post-infusion, measured via flow cytometry; one-way ANOVA performed. (B) Representative contour plots of circulating naïve, T_{EMRA}, T_{EM}, and T_{CM} populations among CAR⁺ T cells at day 7 post-infusion, measured via CD45RA/CD62L flow cytometry staining (C) Quantification of circulating naïve, T_{Eff}, T_{EM}, and T_{CM} populations among CAR⁺ T cells at day 7 post-infusion, measured via CD45RA/CD62L flow cytometry staining; two-tailed unpaired t-test performed. (D) Activation and exhaustion marker phenotype of circulating CAR⁺ T and NK cells at day 7 post-infusion, measured via flow cytometry; one-way ANOVA performed. Data are presented as mean values +/- SEM (n=5 mice/group).

Figure S13

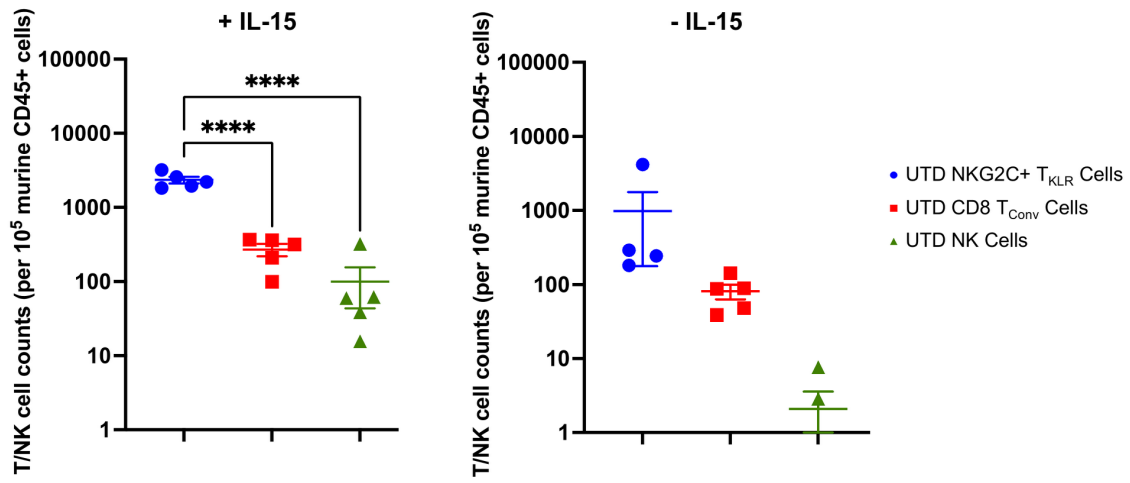


Figure S13. Quantification of circulating UTD NKG2C⁺ T_{KLR} cells, UTD CD8 T_{Conv} cells, and UTD NK cells at day 7 post-infusion, measured via flow cytometry. Data are presented as mean values +/- SEM. Statistical analysis was performed by one-way ANOVA (n=5 mice/group).

Figure S14

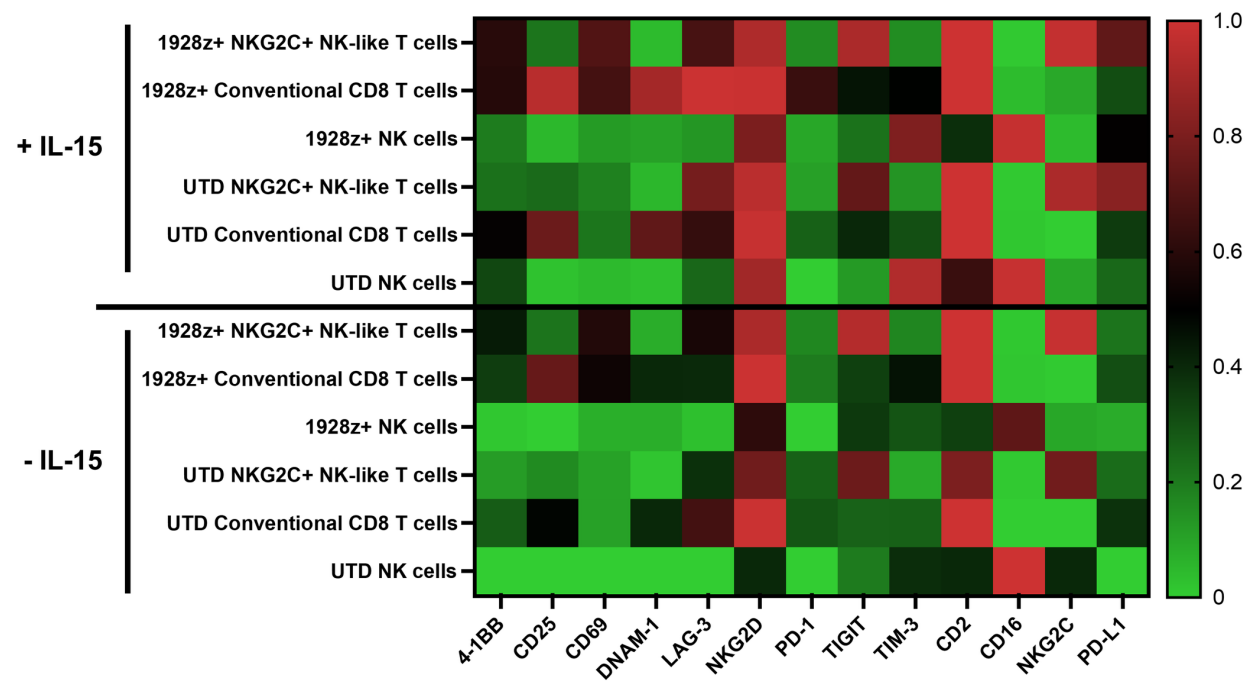


Figure S14. Heatmap of relative frequency of surface marker expression measured on T and NK cells collected from NALM6 tumor-bearing mice at day 7 post-infusion (n=5 mice/group).

Figure S15

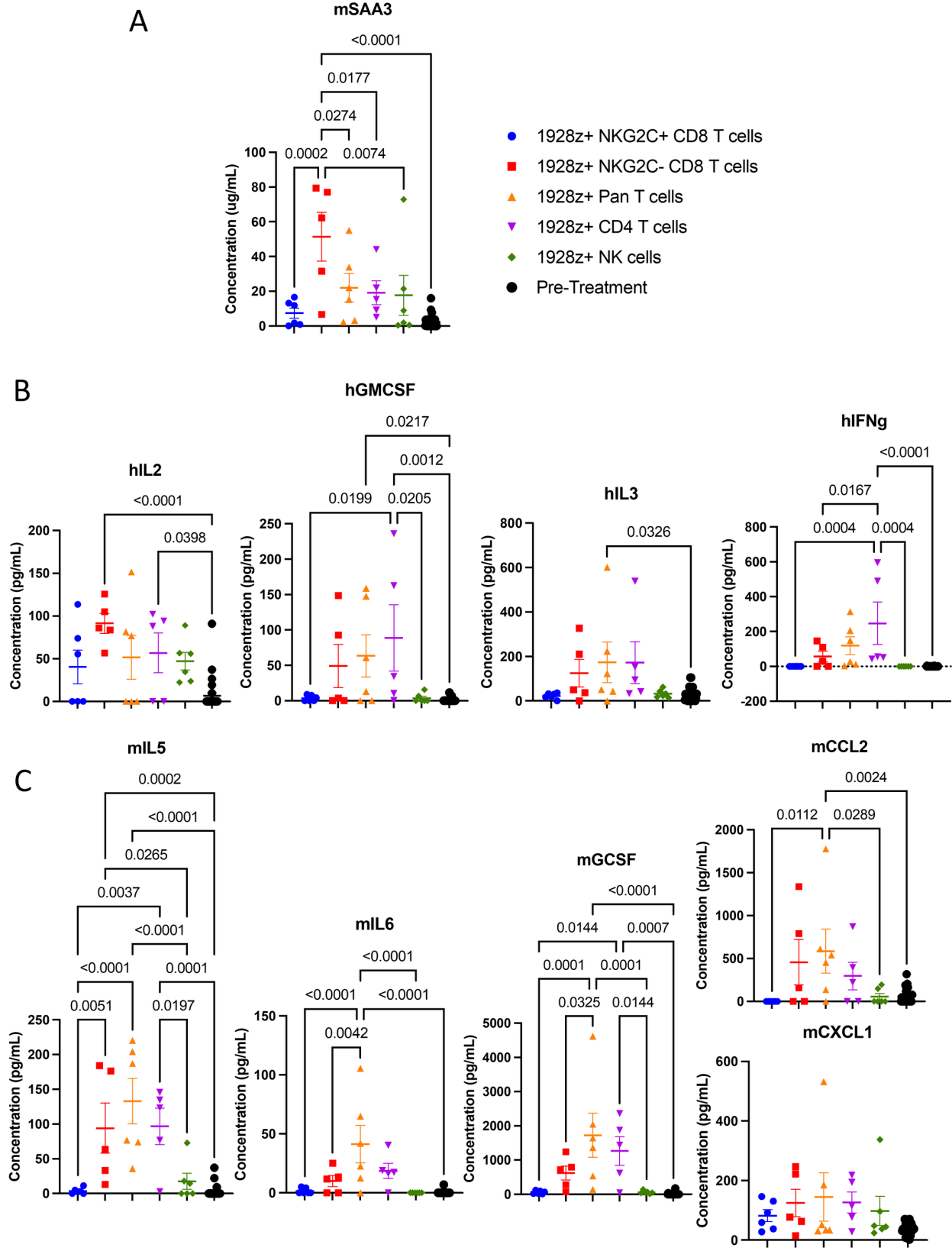


Figure S15. Serum levels of **(A)** murine SAA-3, **(B)** human cytokines, and **(C)** murine cytokines from mice treated with CAR⁺ T and NK cells measured 4 hours pre- and 24 hours post-CAR treatment (n=5 mice for CD8 T_{Conv} group, 6 mice for all other treatment groups). Data are presented as mean values +/- SEM. Statistical analysis was performed by one-way ANOVA.

Figure S16

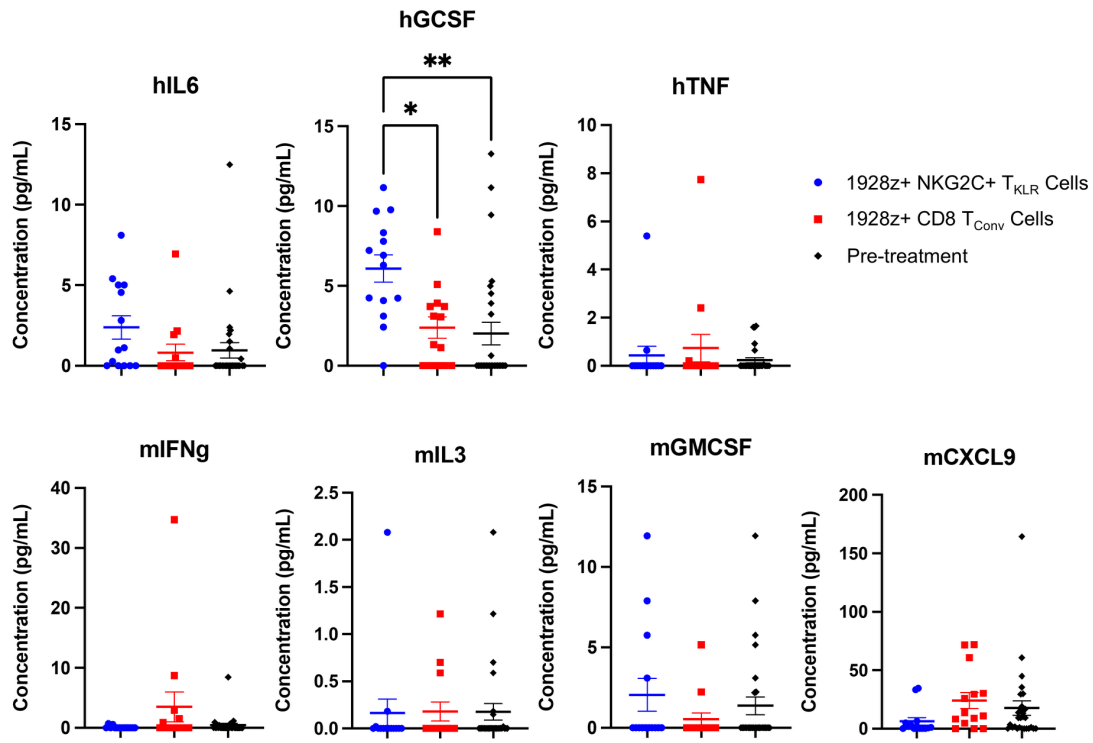


Figure S16. Serum levels of human and murine cytokines of mice treated with CAR⁺ NKG2C⁺ T_{KLR} cells and CAR⁺ CD8 T_{Conv} cells, measured at 4 hours pre- and 24 hours post-CAR infusion. Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by one-way ANOVA (n=14 mice/group).

Figure S17

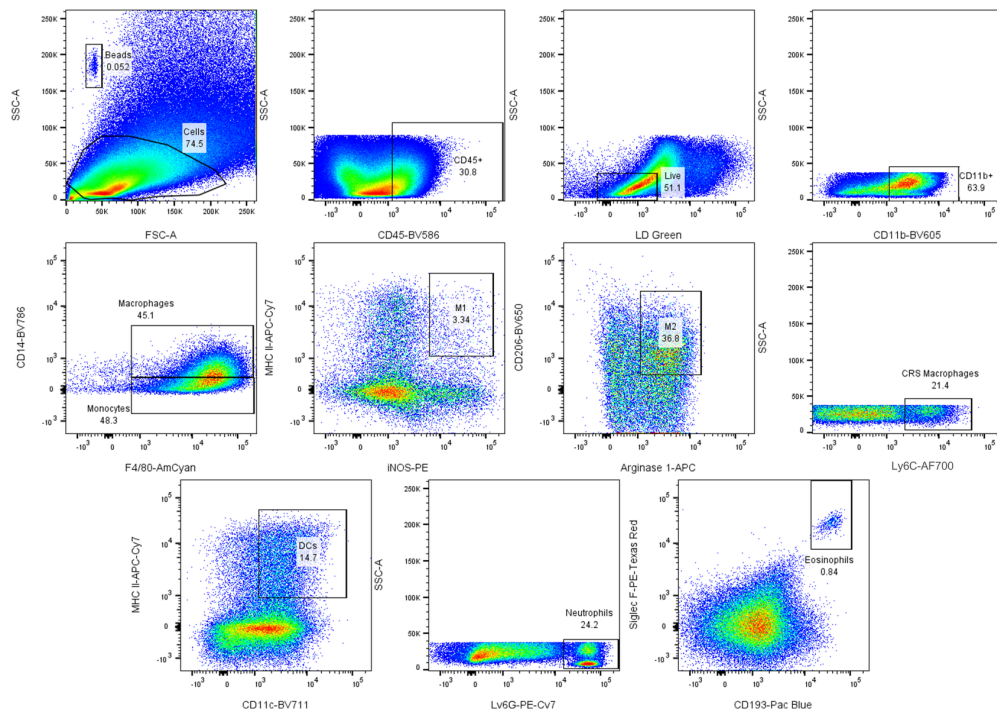


Figure S17. Flow cytometry gating strategy for murine myeloid subsets measured in bone marrow, blood, spleen, and peritoneal cavity of Raji tumor-bearing mice.

Figure S18

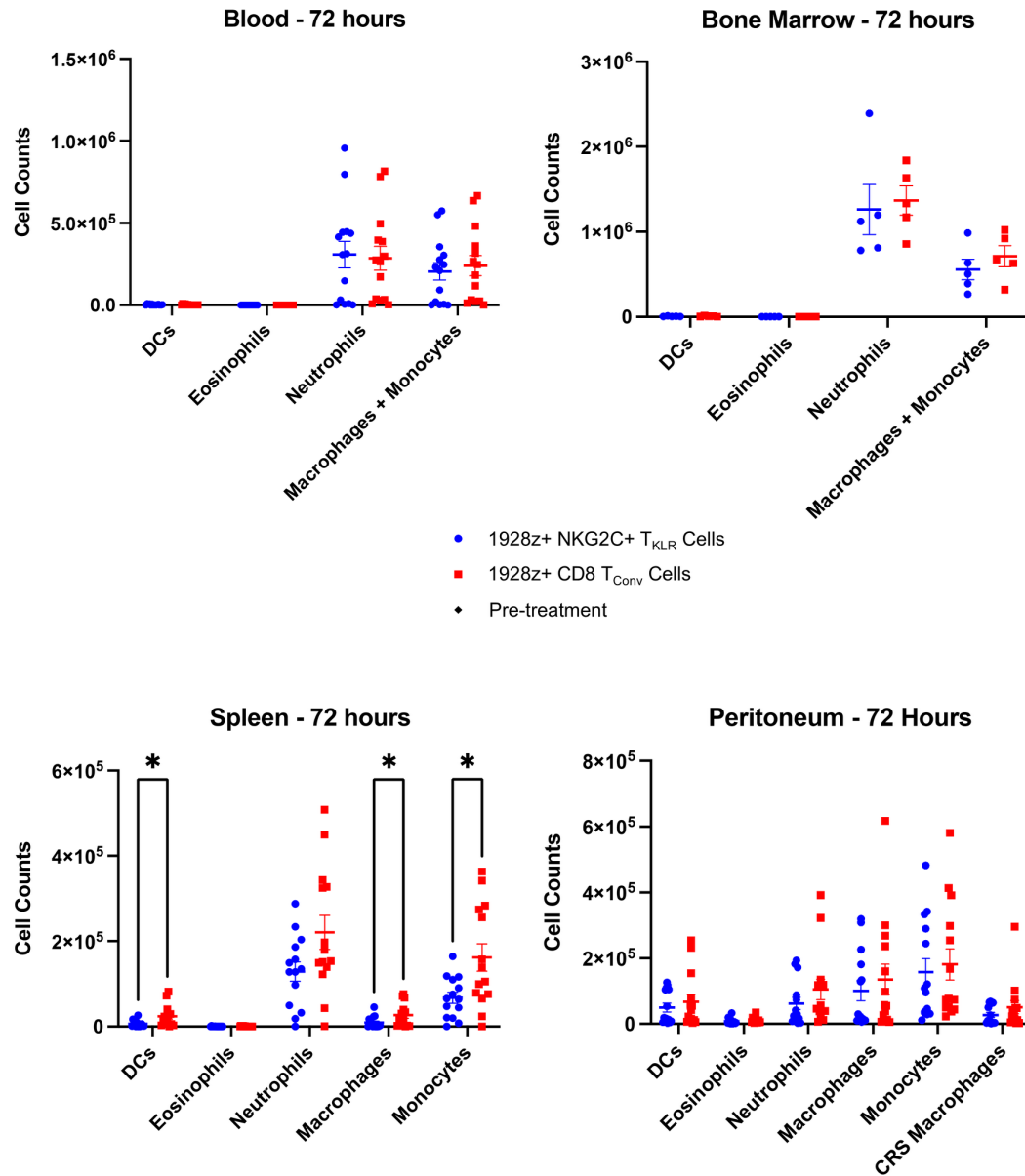


Figure S18. Quantification of murine myeloid subsets measured in bone marrow (BM), blood, spleen, and peritoneal cavity of Raji tumor-bearing mice 72 hours post T cell infusion. CRS macrophages are defined by expression of Ly6C. Data are presented as mean values $\pm$ SEM. Statistical analysis was performed by multiple two-tailed unpaired t-tests (n=14 mice/group).

Figure S19

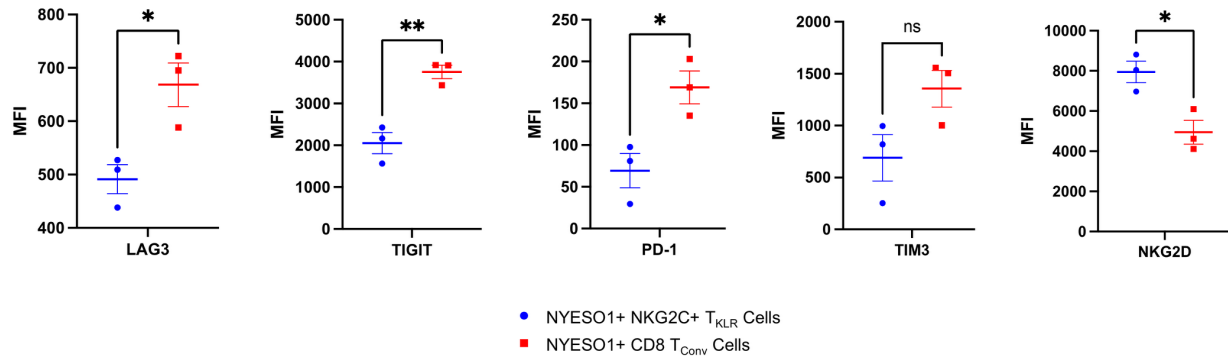


Figure S19. Median fluorescence intensity (MFI) of activating and exhaustion markers on tumor infiltrating TCR-engineered NKG2C⁺ T_{KLR} cells and CD8 T_{Conv} cells, measured at day 17 post-infusion. Data are presented as mean values ± SEM. Statistical analysis was performed by two-tailed unpaired t-test (n=3 mice/group).

Figure S20

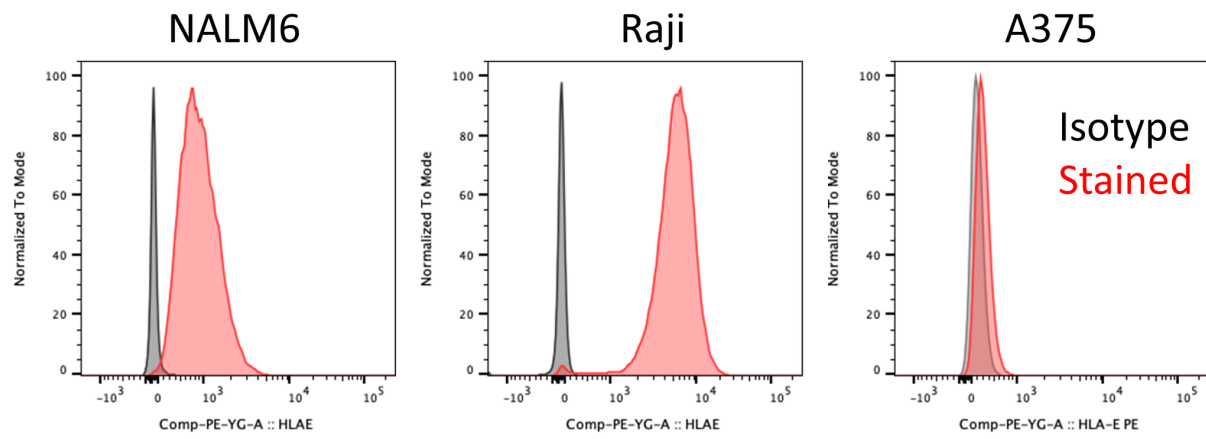


Figure S20. Isotype (gray) and HLA-E (red) staining of NALM6, Raji, and A375 target cell lines.

Table S1. Flow cytometry phenotyping antibodies and reagents.

Antibody Marker/Reagent	Fluorescent Conjugate	Clone	Catalog Number	Manufacturer
TIGIT	AF647	A15153G	372724	Biolegend
DNAM-1	AF700	102511	FAB666N-100UG	R&D Systems
PD-1	APC-Fire750	EH12.2H7	329954	Biolegend
NKG2D	BV421	1D11	568106	BD Biosciences
hCD19	BV510	SJ25C1	363020	Biolegend
TIM-3	BV605	F38-2E2	345017	Biolegend
CD3	BV650	UCHT1	563852	BD Biosciences
LAG-3	BV711	11C3C65	369320	Biolegend
CD25	BV785	M-A251	356140	Biolegend
CD69	PE	FN50	310906	Biolegend
mCD45	PE-Cy5	30-F11	103110	Biolegend
4-1BB	PE-Cy7	4B4-1	309818	Biolegend
CD56	AF488	B159	557699	BD Biosciences
hCD45	PerCP	2D1	347464	BD Biosciences
NKG2C	APC	REA205	130-117-398	Miltenyi
CD45RA	AF700	HI100	304120	Biolegend
CD16	APC-Cy7	3G8	557758	BD Biosciences
CCR7	BV421	G043H7	353208	Biolegend
CD2	BV711	RPA-2.10	300232	Biolegend
CD62L	BV785	DREG-56	304830	Biolegend
PD-L1	PE-Cy7	29E.2A3	329718	Biolegend
TCR Vb13.1	PE	REA560	130-131-910	Miltenyi
CD107a	BV786	H4A3	563869	BD Biosciences
IFN γ	AF700	B27	557995	BD Biosciences
CD56	ECD	N901	A82943	Beckman Coulter
TCRab	APC	IP26	306718	Biolegend
CD8	BV786	RPA-T8	563823	BD Biosciences
NKG2A	PE-Vio770	REA110	130-113-567	Miltenyi
NKG2C	PE	134591	FAB138P	R&D Systems
NKG2C	PE	REA205	130-119-776	Miltenyi
KIR2DL1/S1	FITC	REA284	130-118-961	Miltenyi
KIR2DL2/L3/S2	FITC	CH-L	559784	BD Biosciences
KIR3DL1/S1	FITC	REA168	130-104-836	Miltenyi
NKG2C	Biotin	REA205	130-120-448	Miltenyi
HLA-E	PE	3D12	342604	Biolegend
mCD45	BV570	30-F11	103135	BioLegend
mCD11b	BV605	M1/70	101237	Biolegend
mF4/80	BV510	BM8	123135	Biolegend
mCD14	BV785	Sa14-2	123337	Biolegend
mLy6C	AF700	HK1.4	128023	BioLegend

mCD193	BV421	J073E5	144517	Biolegend
mCD206	BV650	C068C2	141723	Biolegend
mCD11c	BV711	N418	117349	Biolegend
iNOS	PE	W16030C	696805	Biolegend
mSiglec F	PE Dazzle 594	S17007L	155529	Biolegend
mLy6G	PE-Cy7	1A8	127617	BioLegend
mMHC II	APC-Fire 750	M5/114.15.2	107651	Biolegend
mArginase-1	APC	A1exF5	17-3697-80	Thermo Scientific
LD Green			L34970	Thermo Scientific
LD Aqua			L34966	Thermo Scientific
DAPI			564907	BD Biosciences
Human FC Blocking Reagent			130-059-901	Miltenyi
Anti-Mouse FC Block			553142	BD Biosciences
CBA Mouse Mig Flex Set			558341	BD Biosciences
CBA Mouse GM- CSF Flex Set			558347	BD Biosciences
CBA Mouse G- CSF Flex Set			560152	BD Biosciences
CBA Mouse MCP-1 Flex Set			558342	BD Biosciences
CBA Mouse IL-6 Flex Set			558301	BD Biosciences
CBA Mouse IL-3 Flex Set			558346	BD Biosciences
CBA Mouse IL-5 Flex Set			558302	BD Biosciences
CBA Mouse IFN- γ Flex Set			558296	BD Biosciences
KC/CXCL1 Mouse ELISA Kit			EMCXCL1	Thermo Scientific
Mouse SAA-3 ELISA			EZMSAA3-12K	Millipore Sigma
IL-15/IL-15R Complex Mouse Uncoated ELISA Kit with Plates			88-7215-22	Thermo Scientific
CBA Human GM- CSF Flex Set			558335	BD Biosciences
CBA Human G- CSF Flex Set			558326	BD Biosciences
CBA Human IL-6 Flex Set			558276	BD Biosciences
CBA Human TNF Flex Set			558273	BD Biosciences

CBA Human IL-3 Flex Set			558355	BD Biosciences
CBA Human IL-2 Flex Set			558270	BD Biosciences
CBA Human IFN- γ Flex Set			558269	BD Biosciences
CBA Human Soluble Protein Master Buffer Kit			558264	BD Biosciences
CBA Mouse/Rat Soluble Protein Master Buffer Kit			558266	BD Biosciences
NK Cell Isolation Kit, Human			130-092-657	Miltenyi
Anti-Biotin UltraPure Microbeads			130-105-637	Miltenyi
CD8 microbeads, Human			130-045-201	Miltenyi