Supplementary Table 1. Flow cytometry antibodies

Target	Fluorophore	Clone	Isotype	Source	Identifier
7-AAD	–	–	–	BD Biosciences	559925
Annexin V	PE	–	–	BD Biosciences	556421
Cell Proliferation Dye eFluor™ 670	–	–	–	ThermoFisher Scientific	65-0840-85
Streptavidin	PE	–	–	ThermoFisher Scientific	12-4317-87
Biotin-SP goat anti-mouse IgG, F(ab')$_2$	–	Polyclonal	–	Jackson ImmunoResearch	115-065-072
CD1a	BV421	HI149	Mouse IgG1, κ	BD Biosciences	563938
CD1a	APC	HI149	Mouse IgG1, κ	BD Biosciences	559775
CD2	PE	S5.2	Mouse IgG2a	BD Biosciences	347405
CD3	V450	UCHT1	Mouse IgG1, κ	BD Biosciences	560365
CD3	FITC	UCHT1	Mouse IgG1, κ	BD Biosciences	555332
CD3	PerCP	SK7	Mouse IgG1, κ	BD Biosciences	345766
CD3	PE-Cy7	UCHT1	Mouse IgG1, κ	BD Biosciences	563423
CD3	APC	UCHT1	Mouse IgG1, κ	BD Biosciences	555335
CD4	PE	SK3	Mouse IgG1, κ	BD Biosciences	555347
CD4	PerCP-Cy5.5	SK3	Mouse IgG1, κ	BD Biosciences	332772
CD7	FITC	M-T701	Mouse IgG1, κ	BD Biosciences	555360
CD8	BV510	SK1	Mouse IgG1, κ	BioLegend	563919
CD8	APC-Cy7	SK1	Mouse IgG1, κ	BioLegend	344714
CD25	APC	M-A251	Mouse IgG1, κ	BD Biosciences	555434
CD34	APC	581	Mouse IgG1, κ	BD Biosciences	555824
CD38	APC	HIT2	Mouse IgG1, κ	BD Biosciences	555462
CD45	APC-H7	2D1	Mouse IgG1, κ	BD Biosciences	560178
CD45 RA	V500	HI100	Mouse IgG2b, κ	BD Biosciences	561640
CD69	PE	L78	Mouse IgG1, κ	BD Biosciences	341652
CD197 (CCR7)	BV421	150503	Mouse IgG2a	BD Biosciences	562555
Fas	APC	DX2	Mouse IgG1	BD Biosciences	558814
His	Unconjugated	Penta·His	Mouse IgG1	QIAGEN	34660
His	APC	GG11-8F3.5.1	Mouse IgG1	Miltenyi Biotec	130-119-782
HLA-ABC	BV510	G45-2.6	Mouse IgG1, κ	BD Biosciences	740172
TCR alpha/beta	PE-Cy7	IP26	Mouse IgG1, κ	ThermoFisher Scientific	25-9986-42

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Supplementary Figure Legends

Supplementary Figure 1. Functionality of secreted CD1a-TCE. **(A)** Western blot detection of secreted CD1a-TCE in the conditioned media from transfected HEK293T^{WT} cells. Conditioned media from non-transfected cells (NT) and blinatumomab (Blina) were used as negative and positive controls, respectively. One representative experiment is shown. **(B)** Binding assays of conditioned media from NT- or CD1a-TCE-transfected HEK293T^{WT} cells to K562 cells, primary peripheral blood lymphocytes and MOLT4 cells. Specific binding was detected using anti-His-tag mAb and analyzed by flow cytometry. **(C)** T cell activation assay. Freshly isolated T lymphocytes and CD1a-negative (K562) or CD1a-positive (MOLT4) cells were co-cultured at a 1:1 E:T ratio for 24 hours in the presence of conditioned media from NT, CD1a-CAR- and CD1a-TCE-transfected HEK293T^{WT} cells, and CD69 expression was analyzed by flow cytometry. The inset numbers in **B** and **C** represent the percentage of cells staining positive for the indicated marker.

Supplementary Figure 2. Cells surface expression profiles of CD3 and CD1a from all cell types used in this study. The numbers represent the percentage of cells staining positive for the indicated marker.

Supplementary Figure 3. Comparative in vitro study of engineered CD1a-STAb and CD1a-CAR T cells (cont.). **(A)** CD1a expression on HEK293T^{WT} and HEK293T^{CD1a} cells. **(B)** Cell viability kinetics over time of both cell lines cultured alone. **(C)** Real-time cell cytotoxicity kinetics of HEK293T^{WT} cells co-cultured with activated NT, CD1a-CAR or CD1a-STAb T cells at different E:T ratios (5:1 and 1:1). Cell index values were determined over 80 hours with measurements taken at 15 min intervals after addition of effector cells to target cells. Results from duplicates are shown. **(D)** Cytokine secretion assays from NT, CD1a-CAR or CD1a-STAb T cells co-cultured 24 hours with NALM6, MOLT4 or coT-ALL patient primary cells in a 1:1 E:T ratio. Statistical significance was calculated by a two-way ANOVA test corrected with a Tukey's multiple comparisons test. **(E,F)** Direct contact **(E)** and non-contacting **(F)** bystander T cell cytotoxicity. **(E)** Decreasing numbers of activated effector T (AT) cells (NT, CD1a-CAR or CD1a-STAb) were co-cultured with $5x10^4$ K562^{Luc} target cells and increasing numbers of NT T cells from the same donor (bystander T cells), resulting in the indicated AT:T ratios but maintaining a constant 2:1 Effector (AT+bystander):Target ratio. **(F)**. $5x10^4$ K562^{Luc} cells and $1x10^5$ bystander T cells were plated in the bottom well and decreasing numbers of AT cells (NT, CD1a-CAR or CD1a-STAb) in the upper well; ND, not determined. After 48 hours, the percentage of specific cytotoxicity was calculated by adding D-luciferin to detect bioluminescence **(E,F)**. **(G)** Representative experiment of coT-ALL leukemia escape from immune pressure after 4 days. MOLT4 cells were co-cultured with NT, CD1a-CAR or CD1a-STAb T cells at the indicated E:T ratios, and the expression of CD3 and CD1a was analyzed by flow cytometry. Inset numbers represent the percentage of cells staining positive for the indicated marker. **(H)** Percentage of CD1a-CAR T cells, CD1a-STAb T cells and non-transduced T cells (Non-CAR T and Non-STAb T) within CD1a^{-}CD3^{+} cells from immune escape assays after 4, 7 and 11 days of co-culture with MOLT4 cells at the indicated E:T ratios.

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Supplementary Figure 4. MOLT4 cells express functional Fas receptor (CD95) in the membrane. **(A)** Representative FACS analysis showing cell surface expression of Fas receptor in MOLT4 cells. The inset number represents the percentage of cells staining positive. **(B)** MOLT4 cytotoxicity after 24-hour incubation with the activating anti-Fas clone CH11 with or without the neutralizing anti-Fas clone ZB4. Statistical significance was calculated by a one-way ANOVA test corrected with a Tukey's multiple comparisons test.