



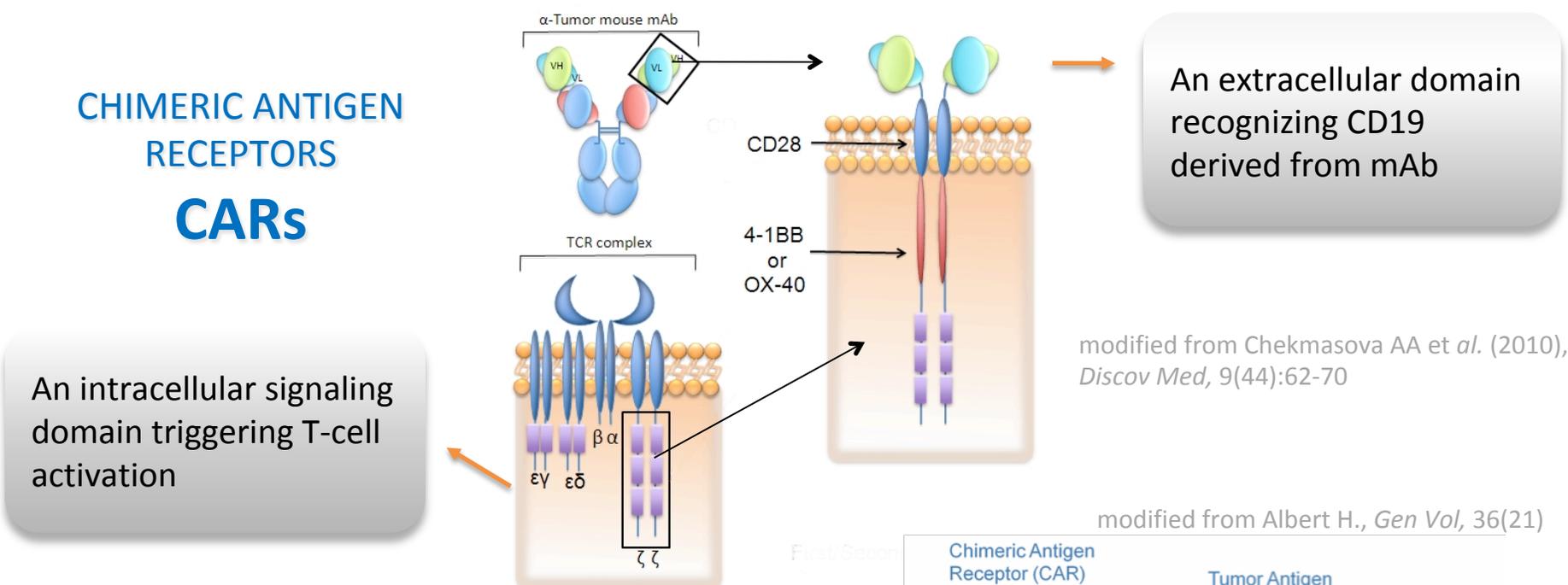
No child should die of cancer

Emerging use of CAR-T cells in leukemia and solid tumors

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What is a CAR-T?

CHIMERIC ANTIGEN RECEPTORS CARs

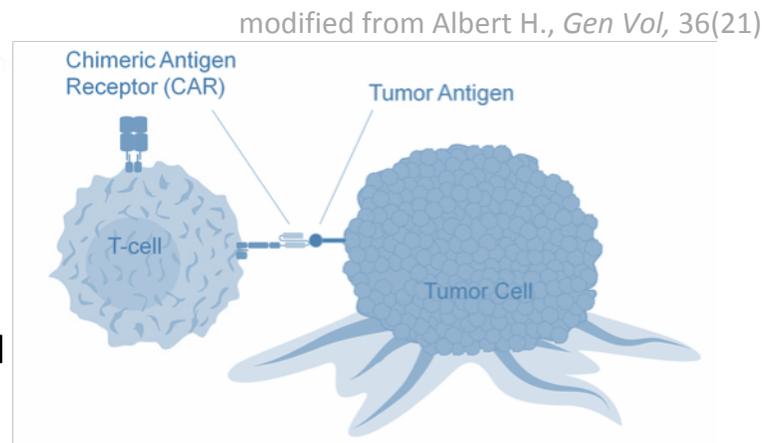


An intracellular signaling domain triggering T-cell activation

An extracellular domain recognizing CD19 derived from mAb

Modified CAR-T cells for ALL:

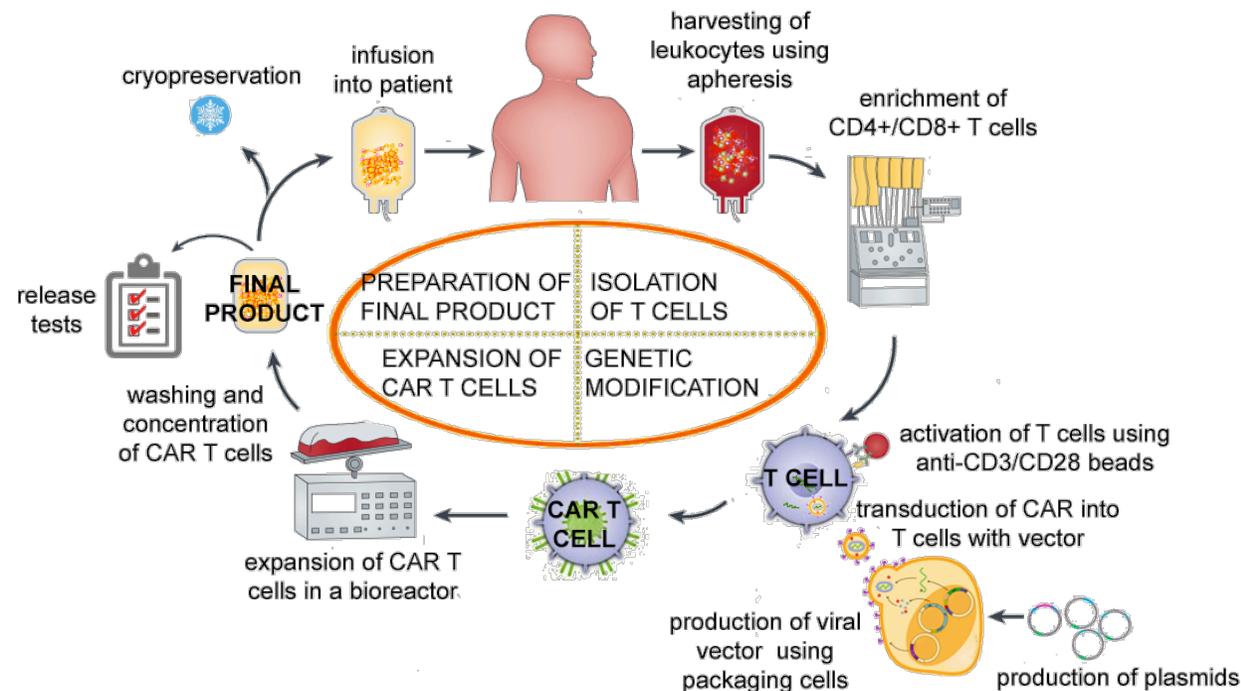
- CD19 antigen is a highly expressed marker in B-ALL
- scFv engagement redirects T-cell antigen specificity and stimulate T-cell activation
- Co-stimulation domains further enhance T-cell function, proliferation and persistence



CAR-T living cells: impressive efficacy!

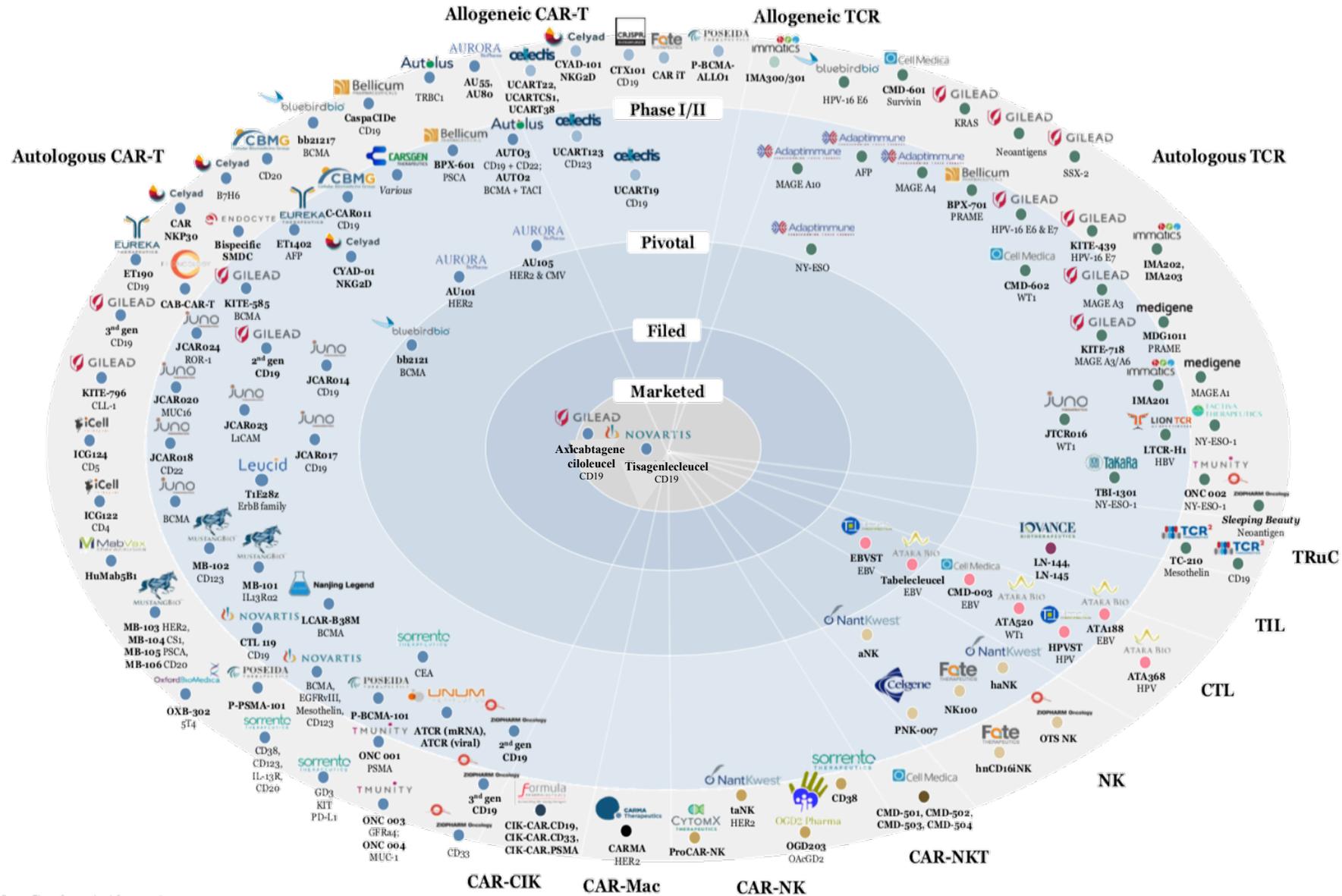
Impressive efficacy of patient-derived viral CAR-T cells in patients with very poor prognosis

- CR in 70 to 90% at 1 month and OS of 60 to 80% at 12 months in adult and pediatric patients with r/r B-ALL, whose chance of survival was 10% to 30% with conventional therapies
- CAR T cell robust *in vivo* expansion and persistence for 3 years or longer in patients



Grupp SL et al. *NEJM* (2013); 368(16):1509-1518; Maude SL et al. *NEJM* (2018); 378(5):439-448; Park JH et al. *NEJM* (2018); 378(5):449-459; Gardner R et al. *Blood* (2017); 129(25):3322-3331

Rapidly evolving CAR-T landscape



We are living the results of the “first wave” of CAR-T cell therapies.

1. Critical issues in ALL:

- Comparative data of efficacy in similar setting of pts?
- Apheresis and success of genetic manipulation;
- What’s driving CAR-T cell expansion?

2. Which patients should have access to CAR-Ts?

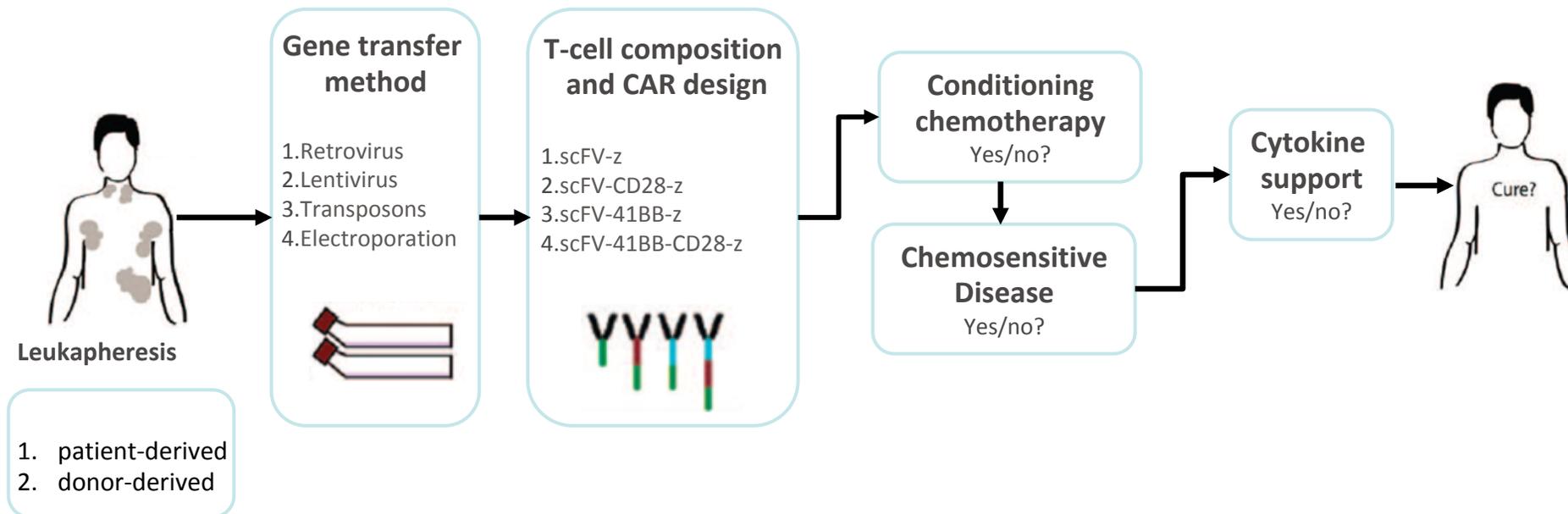
3. Future CAR-Ts with less toxicity?

4. Challenges & complexity of translating success in ALL into solid tumors. What have we learned?

5. More sustainable approaches?

Several observations consistent across trials could improve the design of trials. Different variables influence patient's response:

- Lymphodepleting regime pre-CAR-T cell infusion can improve engraftment and persistence
- Subset composition of T cells (i.e. defined CD8:CD4, $T_{naive}^- T_{scm}^- T_{CM}^- T_{EM}$)
- Immunosuppressive role of the tumor microenvironment



ELIANA: pivotal phase-2 study

ELIANA is the first global, multicenter trial of CAR-T cell therapy

- Tisagenlecleucel was produced at a central manufacturing site with global distribution
- 25 sites across 11 countries in North America, Europe, and Asia-Pacific



Manufacturing sites

Pts who received a tisagenlecleucel infusion: 75 pts with at least 3 months of follow-up: OS 81%; CR 81% (45 pts) and 21% (16) CR **with incomplete hematologic recovery**

ITT analysis: 92 pts. OS 66%

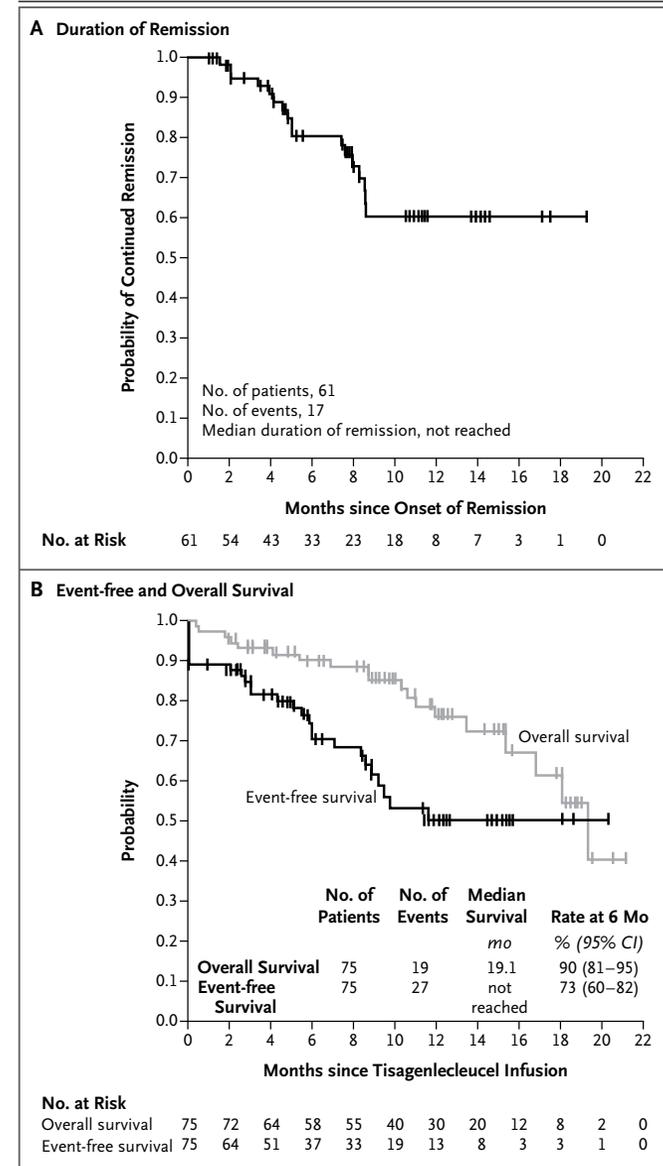
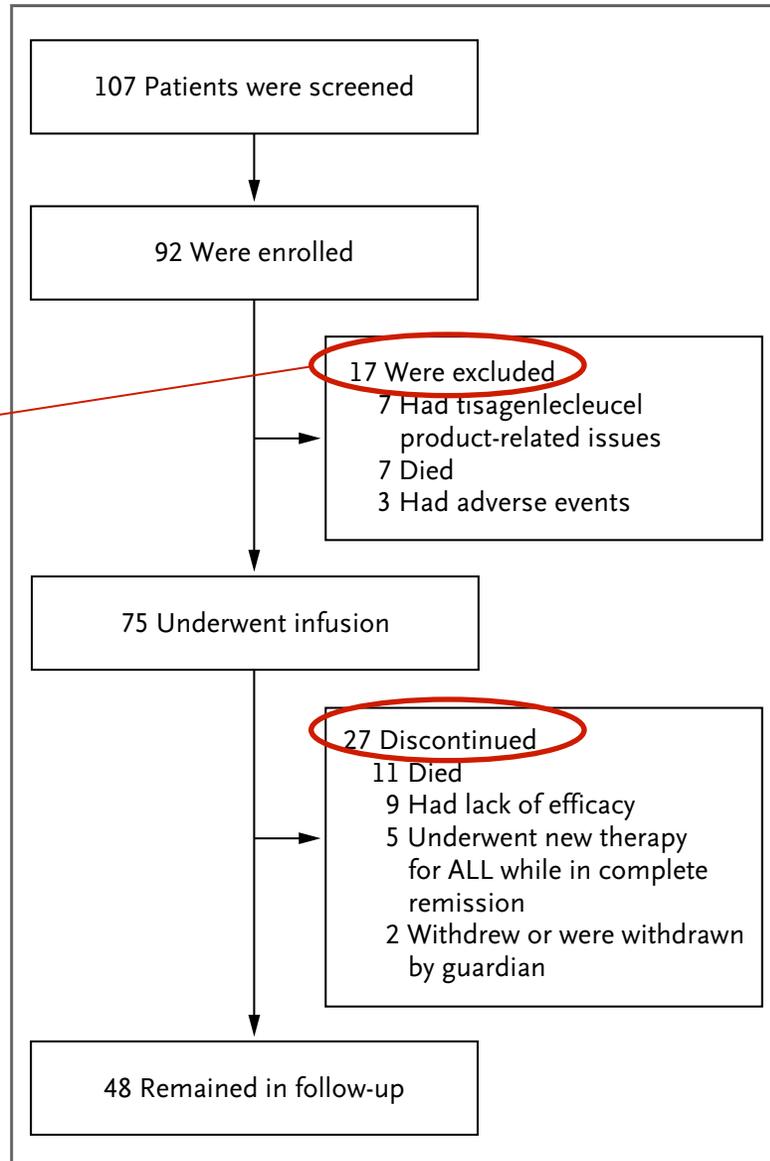
SCT: 8 pts in remission, including 2 in CR but with MRD+ and 2 with B-cell recovery within 6 months after infusion

Rate of **relapse-free survival**: 80% at 6 months and 59% at 12 months

Relapse: 1 patient had a CD19+ recurrence and 15 patients had CD19- (3 with concomitant CD19+ blasts); 6 patients had unknown CD19 status

ELIANA study: feasibility

Autologous
apheresis and
viral manipulation



Tisangelecleucel: persistence

- Persistence of tisagenlecleucel in the blood was observed for 20 months (median time 168 days)
- No relation between dose and expansion
- Patients with B-cell recovery within 6 months had earlier loss of the transgene compared with patients with sustained clinical response.

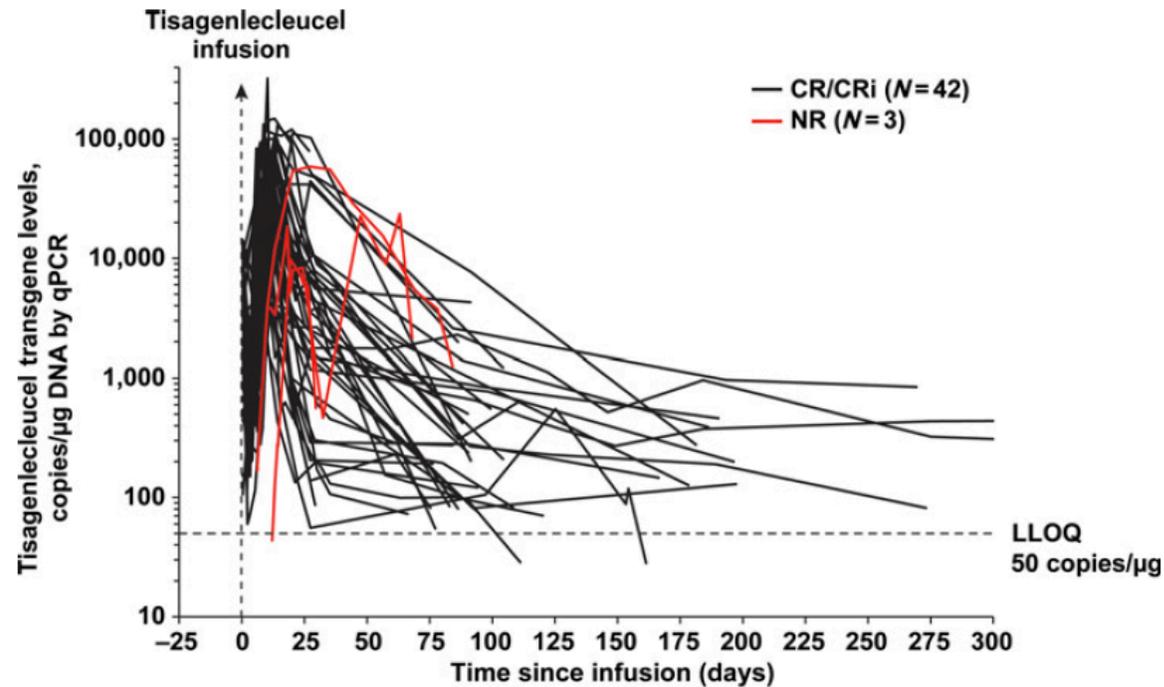


Figure 2.
Cellular kinetics of tisagenlecleucel transgene by response in the ELIANA study.

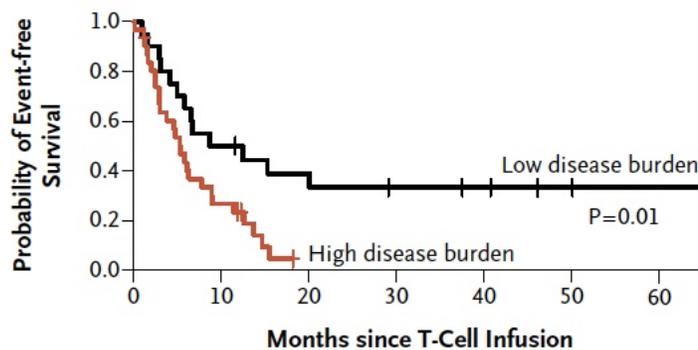
ORIGINAL ARTICLE

Long-Term Follow-up of CD19 CAR Therapy in Acute Lymphoblastic Leukemia

Jae H. Park, M.D., Isabelle Rivière, Ph.D., Mithat Gonen, Ph.D.,
Xiuyan Wang, Ph.D., Brigitte Sénéchal, Ph.D., Kevin J. Curran, M.D.,
Craig Sauter, M.D., Yongzeng Wang, Ph.D., Bianca Santomaso, M.D., Ph.D.,
Elena Mead, M.D., Mikhail Roshal, M.D., Peter Maslak, M.D.,
Marco Davila, M.D., Ph.D., Renier J. Brentjens, M.D., Ph.D.,
and Michel Sadelain, M.D., Ph.D.

➤ **Patients with low disease burden benefited the most from CAR-T therapy**

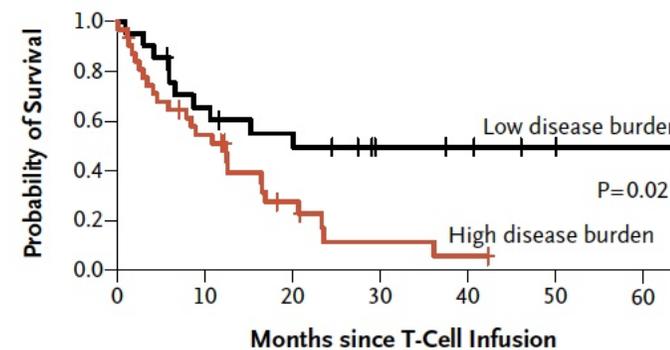
A Event-free Survival, According to Disease Burden



No. at Risk

Low burden	20	10	7	5	4	2	1
High burden	31	8	0	0	0	0	0

B Overall Survival, According to Disease Burden



No. at Risk

Low burden	21	13	10	5	4	2	1
High burden	32	16	6	2	1	0	0

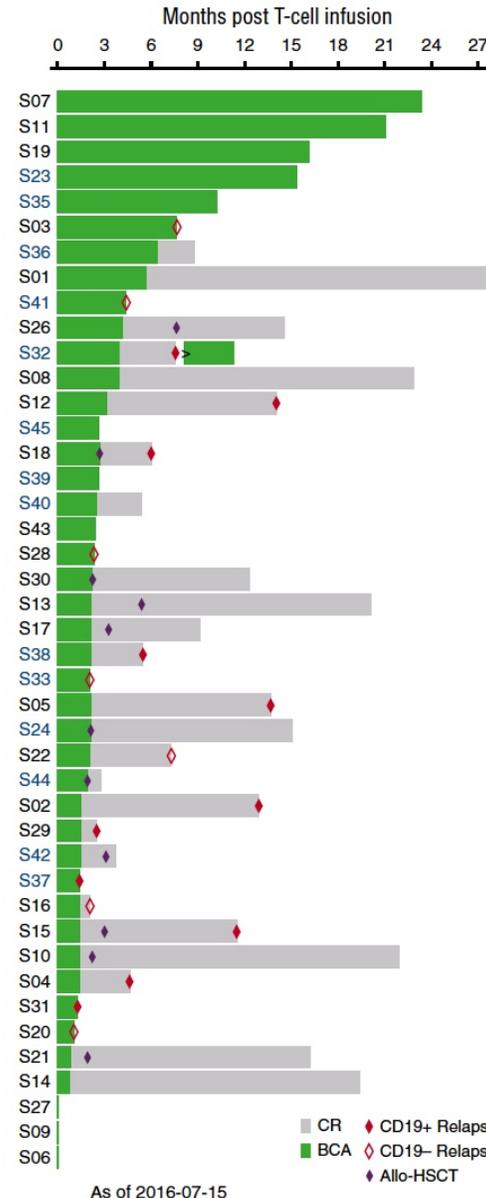
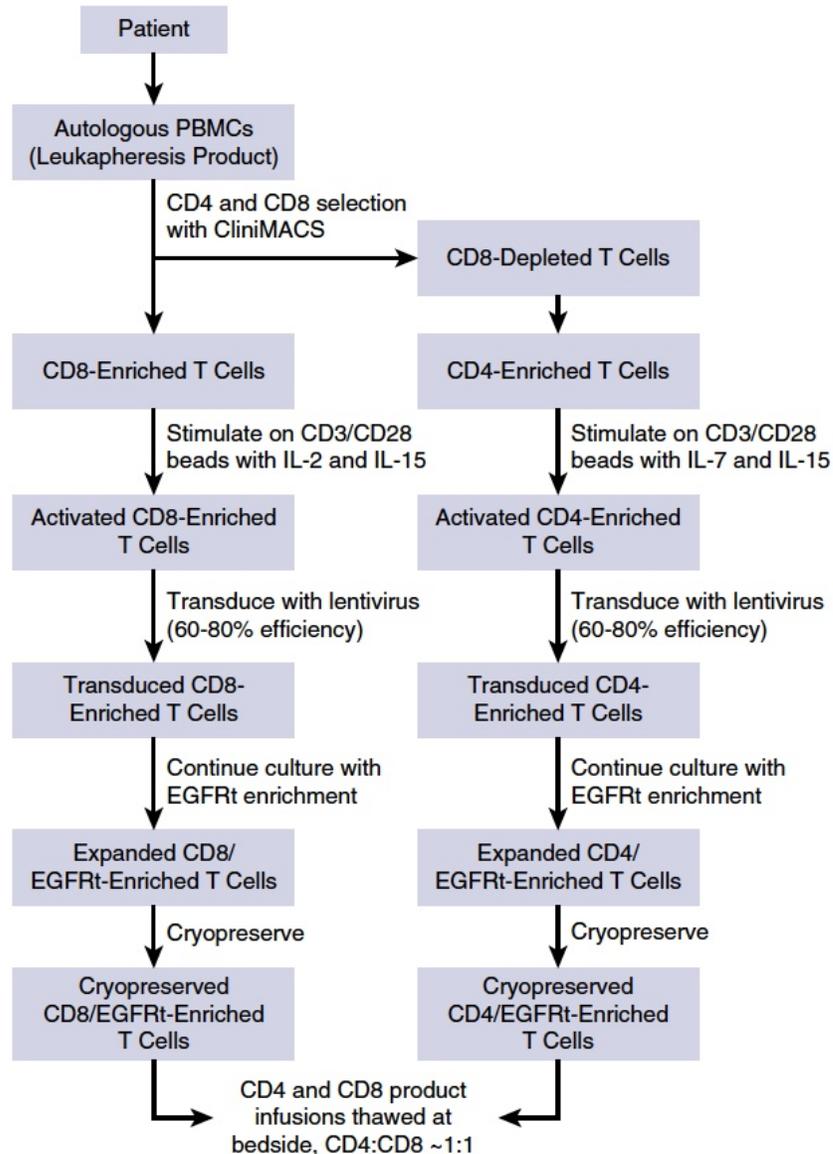
Figure 4. Event-free Survival and Overall Survival, According to Pretreatment Disease Burden.

Patients with a low disease burden (<5% bone marrow blasts) at the time of T-cell infusion had significantly longer event-free survival (Panel A) and overall survival (Panel B) than did those with a high disease burden ($\geq 5\%$ bone marrow blasts or extramedullary disease). The median event-free survival among patients with a low disease burden was 10.6 months (95% CI, 5.9 to not reached), as compared with 5.3 months (95% CI, 3.0 to 9.0) among patients with a high disease burden ($P=0.01$). The median overall survival among patients with a low disease burden was 20.1 months (95% CI, 8.7 to not reached), as compared with 12.4 months (95% CI, 5.9 to 20.7) among those with a high disease burden ($P=0.02$).

All pts except 2 showed a T-cell persistence of 10-40 days with a median persistence of 14 days. CAR-T cells were not detected beyond 68 days. They did not find correlation between persistence of CAR-T cells and LT survival.



Defined CD4⁺:CD8⁺ composition in JCAR017 contributes to remission rate



Seattle Children's Hospital:

- **93% CR** (40/43 patients)
 - 100% MRD-negative (40/40)
- 1-year OS: 69.5%
 - 10/43 severe CRS, 21/43 severe neurotoxicity

➤ **Persistence of CAR-T cells, antigen escape, and complexity of manufacturing are key aspects for further optimization**



What's driving CAR-T cell expansion?

- Robust CAR-T cell expansion was strongly associated with the probability of achieving MRD-negative CR in adult B-ALL

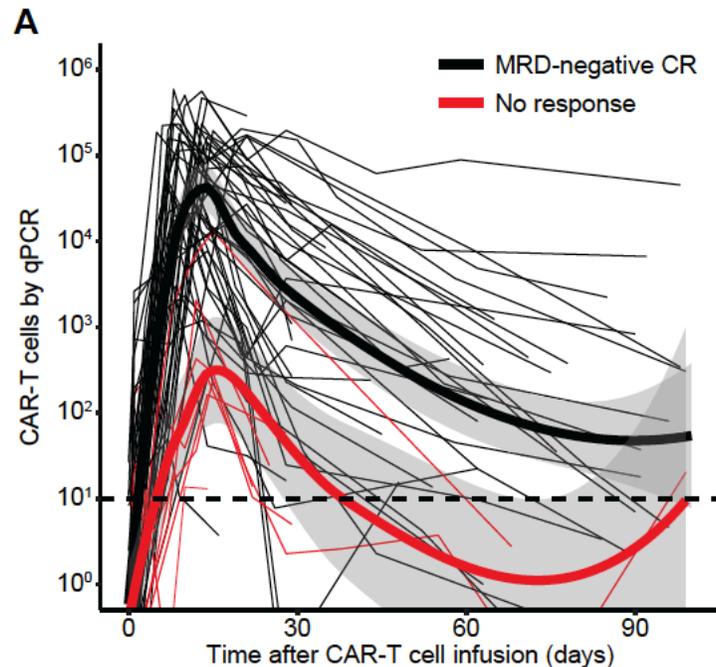


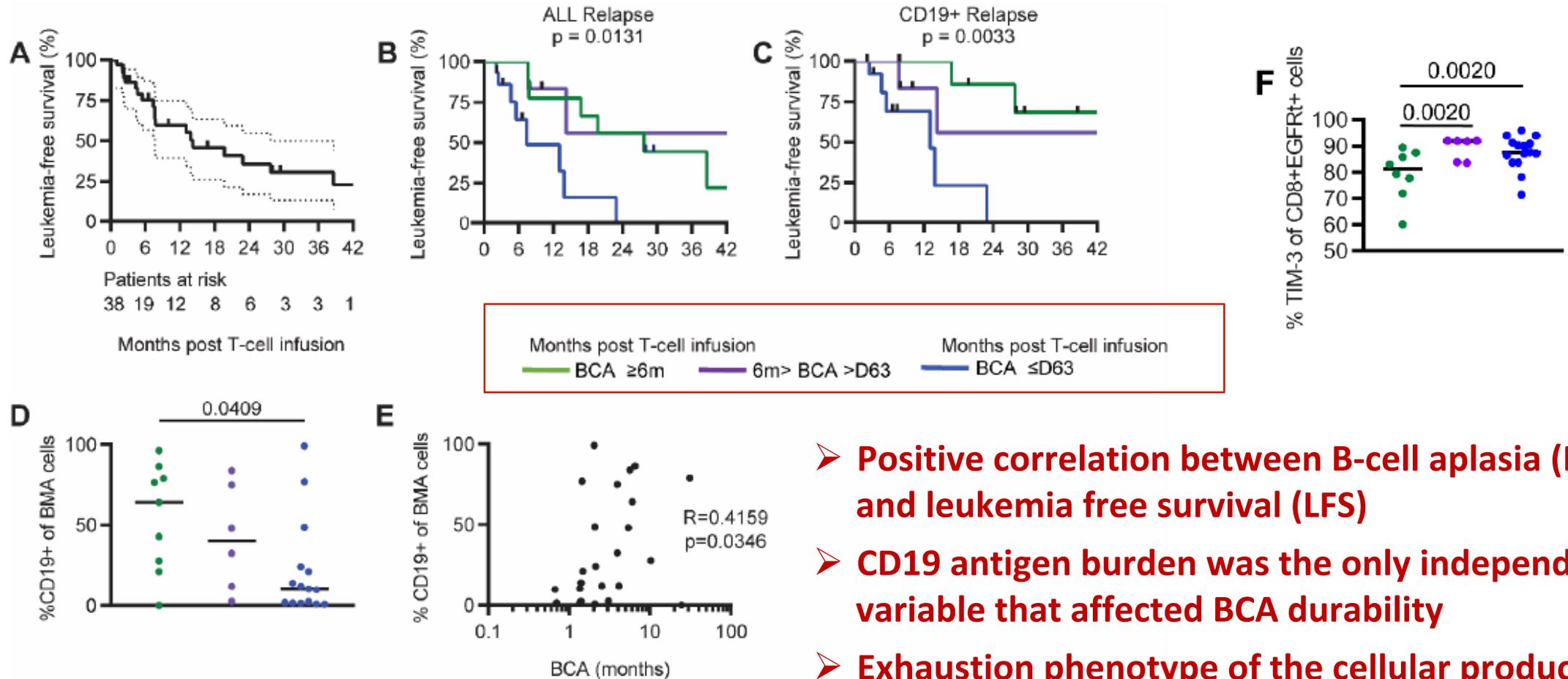
Table 2: Univariate and multivariable analyses for factors impacting event-free survival in patients who achieved MRD-negative CR.

Variable	Univariate HR (95% CI)	P value	Multivariable HR (95% CI)	P value
LDH (per 100 U/L, pre-lymphodepletion)	1.49 (1.22-1.80)	<.0001	1.39 (1.12-1.74)	.003
Bridging systemic therapy ^a	5.66 (2.56-12.5)	<.0001	- ^b	-
Platelet count (per 50,000/ μ L, pre-lymphodepletion)	0.57 (0.42-0.76)	.0002	0.65 (0.47-0.88)	.006
Extramedullary disease (Y)	3.57 (1.66-7.65)	.001	-	-
Fludarabine added to lymphodepletion (Y)	0.30 (0.13-0.66)	.003	0.34 (0.15-0.78)	.011
IL-6 (pg/mL, pre-lymphodepletion)	1.02 (1.01-1.03)	.005	-	-
Marrow blasts by flow cytometry (%)	1.01 (1.00-1.03)	.006	-	-
High-risk cytogenetics ^d (Y)	2.48 (1.12-5.50)	.03	-	-
Neutrophil count (1000/ μ L, pre-lymphodepletion)	0.73 (0.55-0.97)	.03	-	-
Soluble TNFRp55 (pg/mL, Day 0)	4.84 (1.07-21.8)	.04	- ^c	-
IL-2 (pg/mL, Day 0)	3.24 (1.05-10.0)	.04	-	-
IL-8 (pg/mL, pre-lymphodepletion)	1.78 (1.00-3.15)	.05	-	-
Soluble TIM-3 (ng/mL, pre-lymphodepletion)	1.05 (1.00-1.11)	.06	-	-
Dose level (2×10^5 vs 2×10^6 CAR-T cells/kg)	0.51 (0.24-1.11)	.09	-	-
No. prior regimens (n)	1.13 (0.97-1.32)	.1	-	-
Prior allogeneic hematopoietic cell transplantation (Y)	1.65 (0.79-3.44)	.2	-	-

- Pre-lymphodepletion LDH concentration and platelet count reflected an aggressive progression and increased bulk of disease requiring bridging therapy
- Allogeneic HCT after CD19 CAR-T cell therapy is associated with better EFS



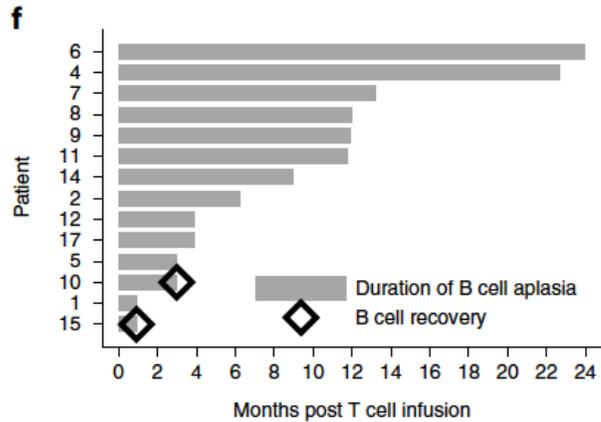
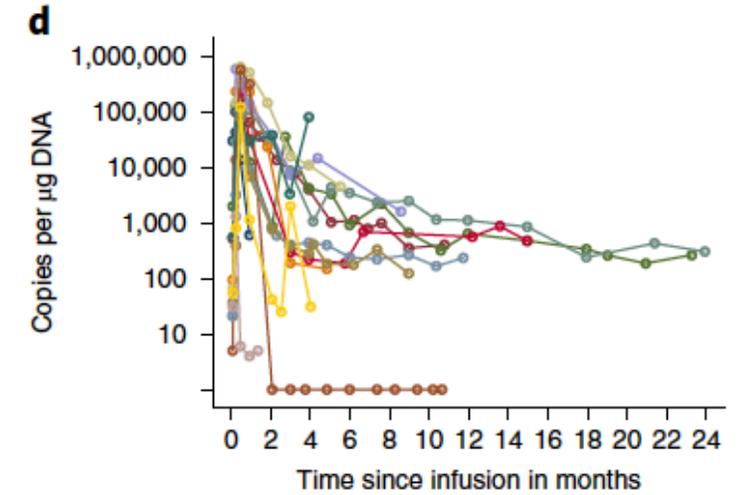
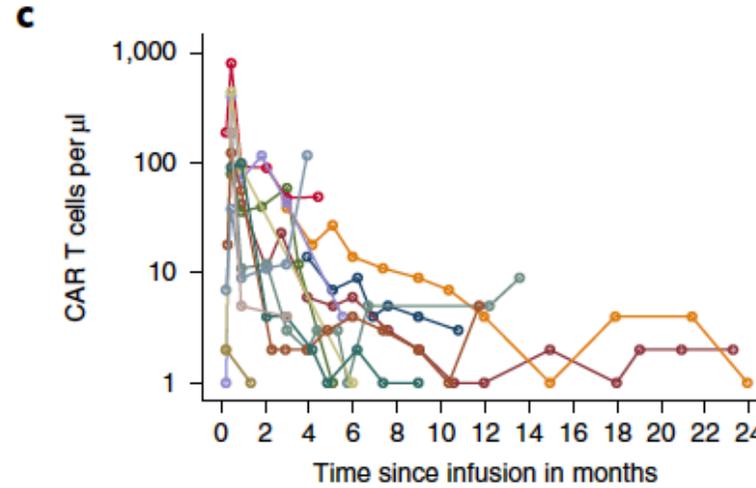
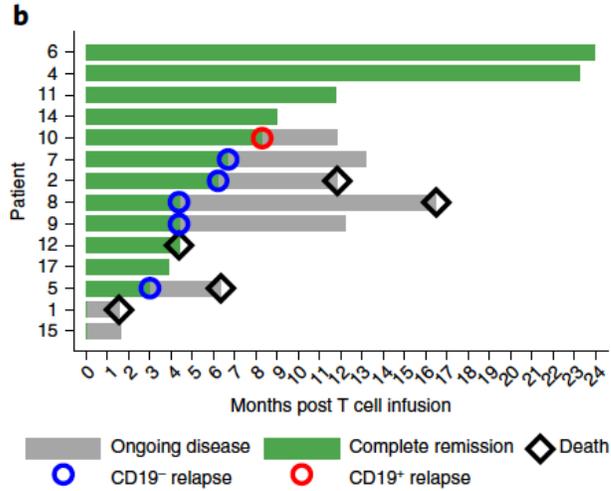
Short BCA correlates with high risk of relapse in pediatric patients



- Positive correlation between B-cell aplasia (BCA) and leukemia free survival (LFS)
- CD19 antigen burden was the only independent variable that affected BCA durability
- Exhaustion phenotype of the cellular product was associated with short BCA and reduced CAR-T cell expansion



The design of CAR construct has an impact on response and persistence



- **Low-affinity CAT CD19 CAR and PGK promoter which drives better CAR expression**
- **Excellent expansion was achieved despite the majority of the patients having lower tumor burden compared to patients treated with tisagenlecleucel**
- **Higher expression of IL-7R and Bcl-2 in CAT CAR-T cells might promote homeostatic proliferation and prevent apoptosis**



Table 1 | Reported causes of death after CAR-T-cell therapies (excluding progressive disease)

Study	Malignancy	Patient age (years)	CAR-T-cell product* (designation or name)	CAR-T-cell dose (per kg)	Day of death after CAR-T-cell infusion	Cause of death [†]
Morgan <i>et al.</i> (2010) ⁸⁸	Metastatic colon cancer	39	HER2-28-137-ζ	1 × 10 ¹⁰ total cells	5	ARDS
Brentjens <i>et al.</i> (2010) ⁹⁴	CLL	69	CD19-28-ζ (19-28z)	1.2–3.0 × 10 ⁷	2	CRS
Frey <i>et al.</i> (2014) ⁴⁴	B-ALL	>18	CD19-137-ζ (tisagenlecleucel, previously known as CTL019)	6.5 × 10 ⁶	5	CRS (+ Influenza B)
				6.7 × 10 ⁶	15	CRS (+ Pseudomonas sepsis, pneumonia)
				8.4 × 10 ⁶	15	CRS (+ Stenotrophomonas sepsis, pneumonia)
Kochenderfer <i>et al.</i> (2015) ¹¹	PMBCL	30	CD19-28-ζ	2.5 × 10 ⁶	16	Unknown (possibly cardiac arrhythmia)
Chong <i>et al.</i> (2016) ⁹⁵	FL	>18	CD19-137-ζ (tisagenlecleucel)	NA	NA	Encephalitis

Toxicities are associated with a potent immune response

Study	Malignancy	Patient age (years)	CAR-T-cell product* (designation or name)	CAR-T-cell dose (per kg)	Day of death after CAR-T-cell infusion	Cause of death [†]
			citoleucel; axi-cel, also known as KTE-C19)			
Locke <i>et al.</i> (2016) ⁹⁶ (ZUMA-1)	NHL	>18	CD19-28-ζ (axi-cel)	2 × 10 ⁶	NA	Cardiac arrest
Turtle <i>et al.</i> (2016) ¹⁷	B-ALL	48	CD19-137-ζ	11.6 × 10 ⁶ CD4 ⁺ + 8.4 × 10 ⁶ CD8 ⁺	3	CRS
		52	CD19-137-ζ	1 × 10 ⁶ CD4 ⁺ + 1 × 10 ⁶ CD8 ⁺	122	Neurotoxicity
Turtle <i>et al.</i> (2016) ¹⁸	NHL	>18	CD19-137-ζ	10 × 10 ⁶ CD4 ⁺ + 10 × 10 ⁶ CD8 ⁺	30	CRS (+ GI bleed)
				10 × 10 ⁶ CD4 ⁺ + 10 × 10 ⁶ CD8 ⁺	13	Neurotoxicity (+ CNS bleed)
ROCKET (2017) ^{64,69}	B-ALL	NA	CD19-28-ζ (JCAR015)	NA	NA	Cerebral oedema (5 cases)
ZUMA-1 (2017) ⁷⁰	NHL	>18	CD19-28-ζ (axi-cel)	NA	NA	Cerebral oedema
Turtle <i>et al.</i> (2017) ¹⁹	CLL	62	CD19-137-ζ	1 × 10 ⁶ CD4 ⁺ + 1 × 10 ⁶ CD8 ⁺	11	Cerebral oedema

ζ, T-cell receptor CD3ζ chain; 137, CD137 (4-1BB); 28, CD28; ARDS, acute respiratory distress syndrome; B-ALL, B-cell acute lymphoblastic leukaemia; CAR, chimeric antigen receptor; CLL, chronic lymphocytic leukaemia; CNS, central nervous system; CRS, cytokine-release syndrome; DLBCL, diffuse large-B-cell lymphoma; FL, follicular lymphoma; GI, gastrointestinal; HLH, haemophagocytic lymphohistiocytosis; NA, not available; NHL, non-Hodgkin lymphoma; PMBCL, primary mediastinal B-cell lymphoma. *In the format: target antigen/co-stimulatory domain/T-cell-receptor activation domain. †Single case, unless otherwise noted.

Cytokine release syndrome (CRS): Eliana study

	Patients infused (N = 79)
Patients developed CRS, n (%)	61 (77)
Time to onset, median (range), days	3.0 (1-22)
Duration of CRS, median (range), days	8.0 (1-36)
ICU admission, n (%)	38 (48)
Anticytokine therapy, n (%)	31 (39)
Tocilizumab, n (%)	31 (39)
1 dose	18 (23)
2 doses	10 (13)
3 doses	3 (4)
Corticosteroids, n (%)	16 (20)
Hypotension that required intervention, n (%)	42 (53)
High-dose vasopressors, n (%)	19 (24)
Intubation, n (%)	12 (15)
Dialysis, n (%)	8 (10)

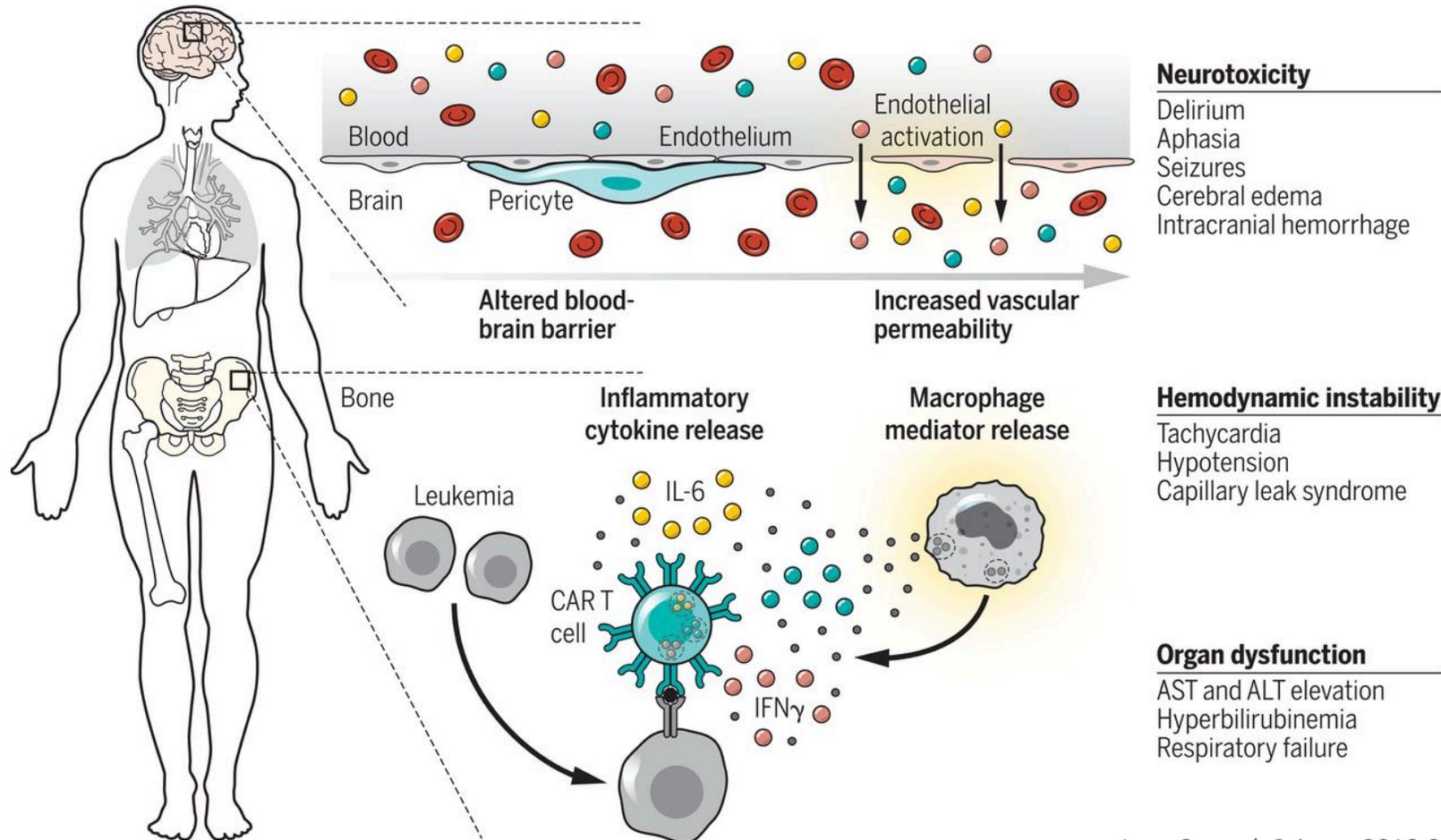
CRS was graded using the Penn scale and managed by a protocol-specific algorithm¹

ICU, intensive care unit.

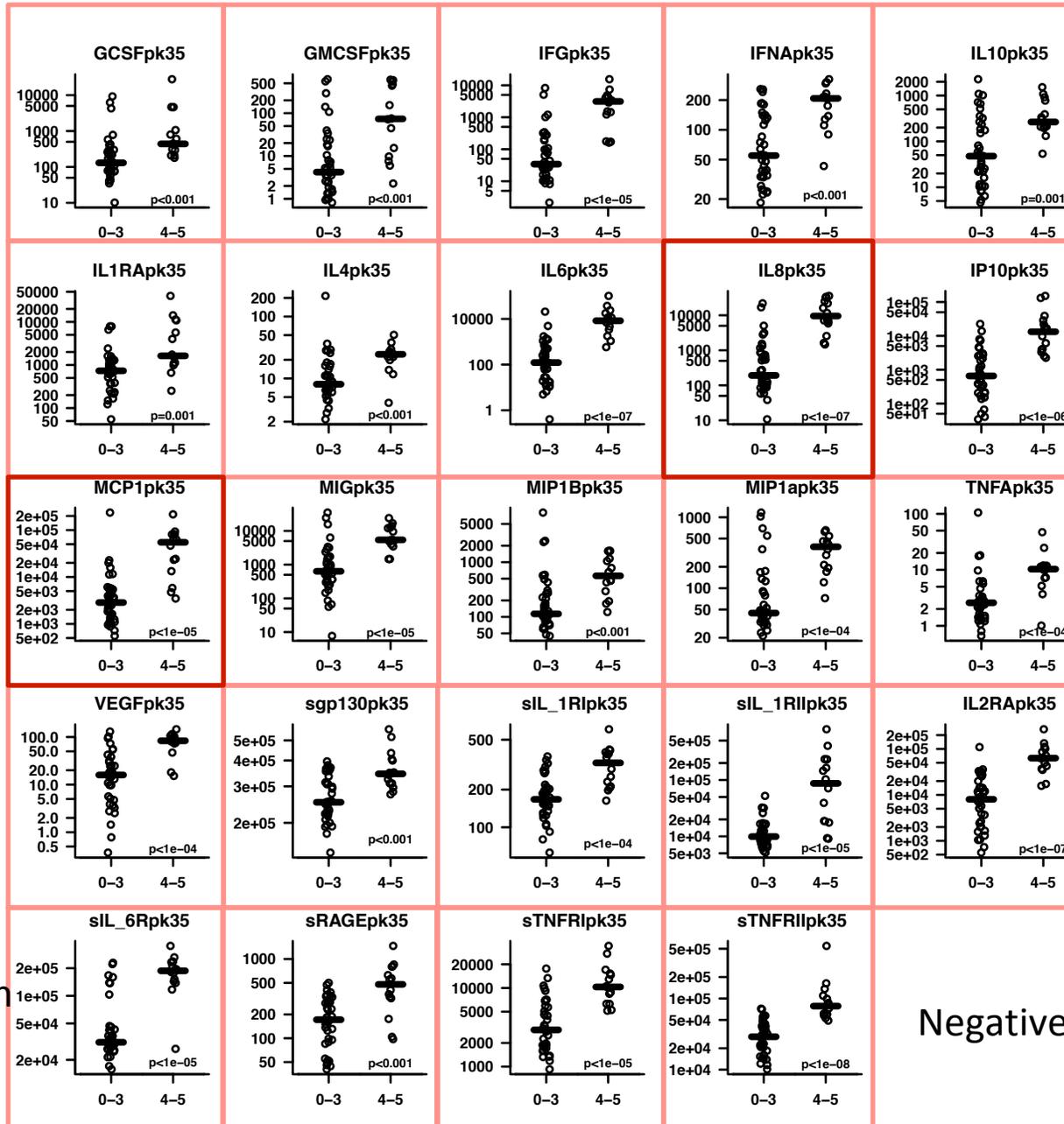
1. Porter DL, et al. *Sci Transl Med.* 2015;7(303):303ra139.

Soluble mediators in CAR-T cells account for toxicities

- Macrophages and other innate immune cells become activated and contribute to the release of soluble mediators.
- CAR-T cells are routinely observed in cerebral spinal fluid and the cytokines may increase permeability to soluble mediators.



Biology of CRS



- IL-8 and MCP1 are associated with CRS occurrence
- CRS is also more evident with 41BB costimulation

Activated T cells

Activated macrophages

Chemotactic for macrophages

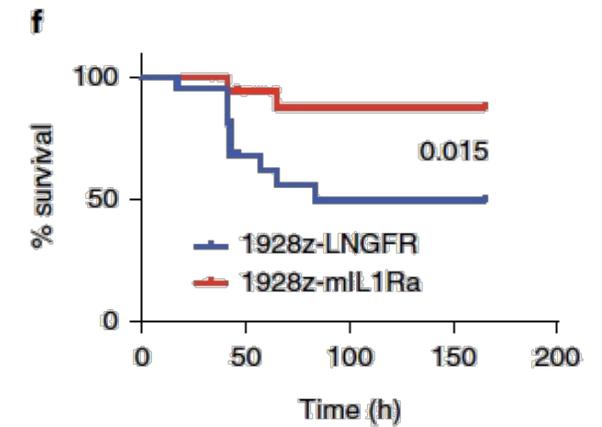
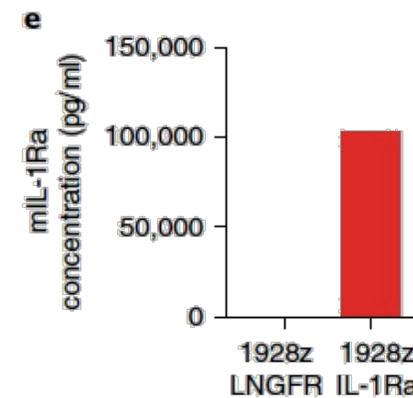
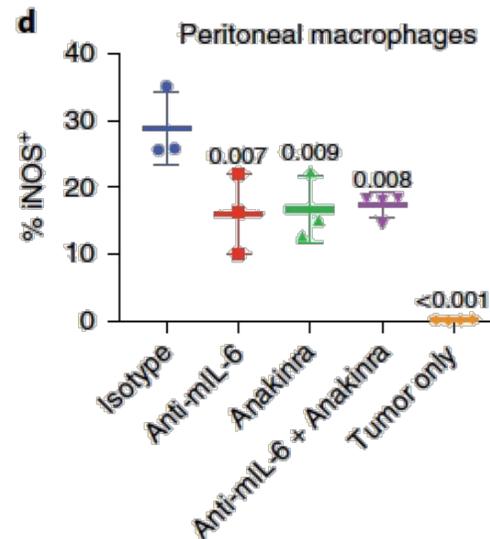
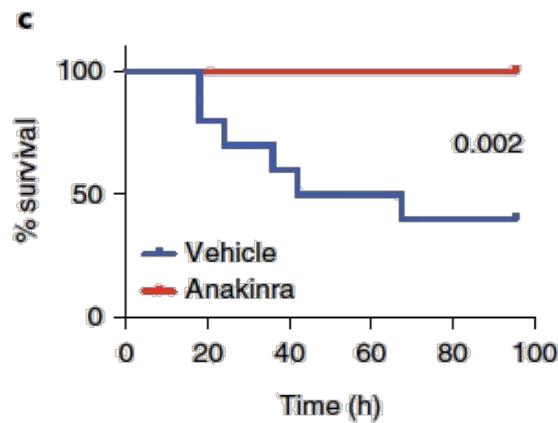
Tissue damage and inflammation

Negative regulators



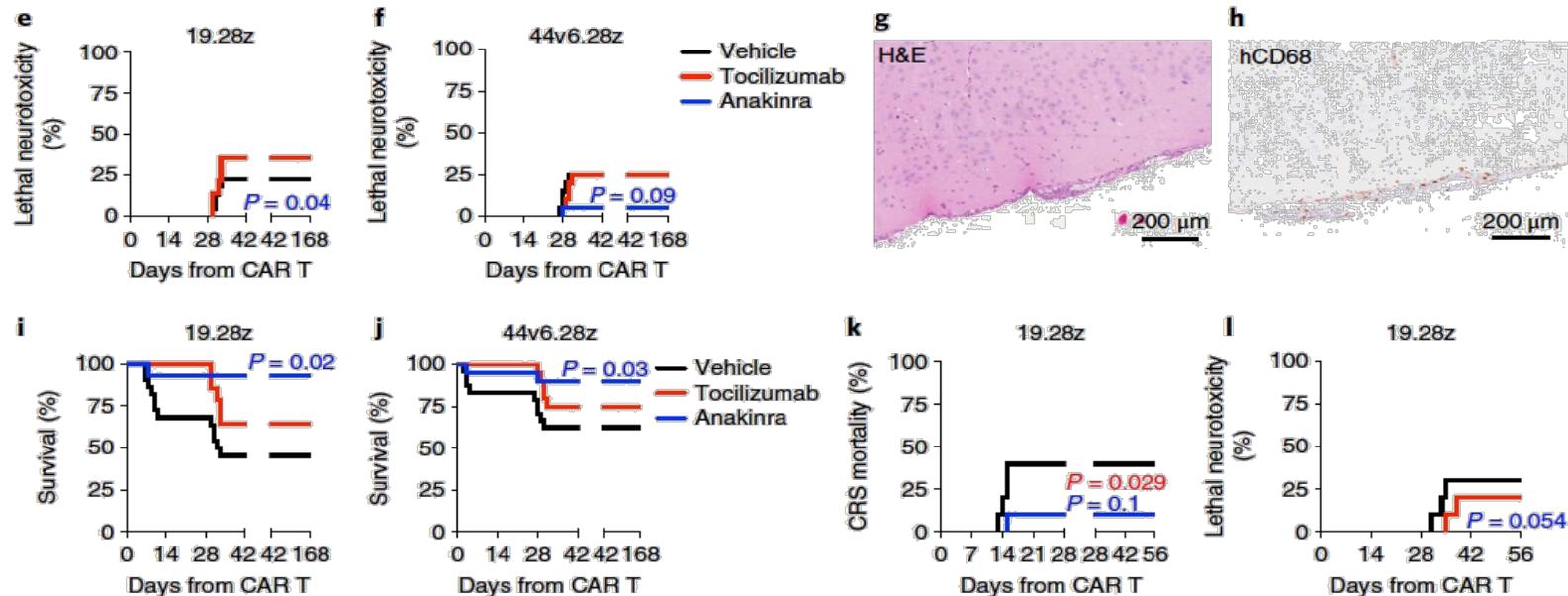
A murine model of CRS shows ablation by IL-1 blockade

- Development of a murine model of CRS that develops within 2–3 d of CAR-T cell infusion and that is potentially lethal and responsive to IL-6 receptor blockade (Raji tumor cells were intraperitoneally injected in SCID-BEIGE mice).
- Its severity is mediated not by CAR-T cell-derived cytokines, but by IL-6, IL-1 and nitric oxide (NO) produced by recipient macrophages.



Monocyte-derived IL-1 and IL-6 are differentially required for CRS and neurotoxicity

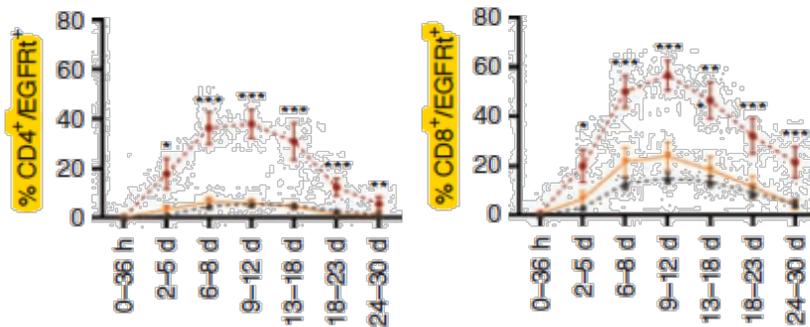
- Development of a mouse model recapitulating key features of CRS and neurotoxicity (Human HSPCs were intrahepatically injected into SGM3 mice with high leukemia burden).
- Human monocytes were the major source of IL-1 and IL-6 during CRS.
- Tocilizumab cured CRS but failed to protect mice from delayed lethal neurotoxicity.
- The IL-1 receptor antagonist anakinra abolished both CRS and neurotoxicity.



Future CAR-Ts with less toxicity?

- Higher CAR-T cell dose and use of lymphodepletion incorporating Flu were associated with development of grade ≥ 4 CRS
- Pre-existing neurological comorbidity and factors associated with higher number of CAR-T cells in vivo (Cy/Flu lymphodepletion, higher infused CAR-T cell dose and higher burden of CD19+ malignant cells in marrow) increased the risk of grade ≥ 3 neurotoxicity

Chou CK, et al. *BMT*. 2019;54:780–84.



---▲--- Grade 0 neurotoxicity
—■— Grade 1–2 neurotoxicity
---●--- Grade 3–5 neurotoxicity

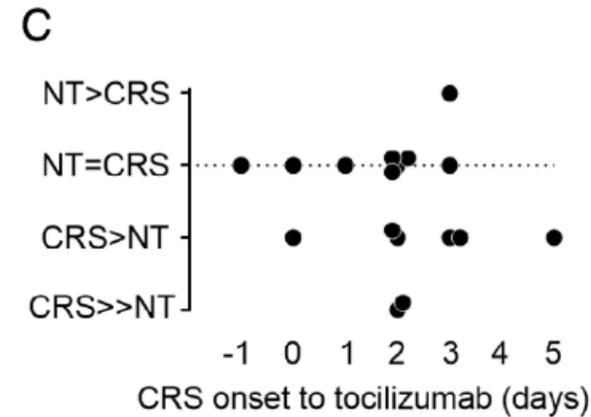
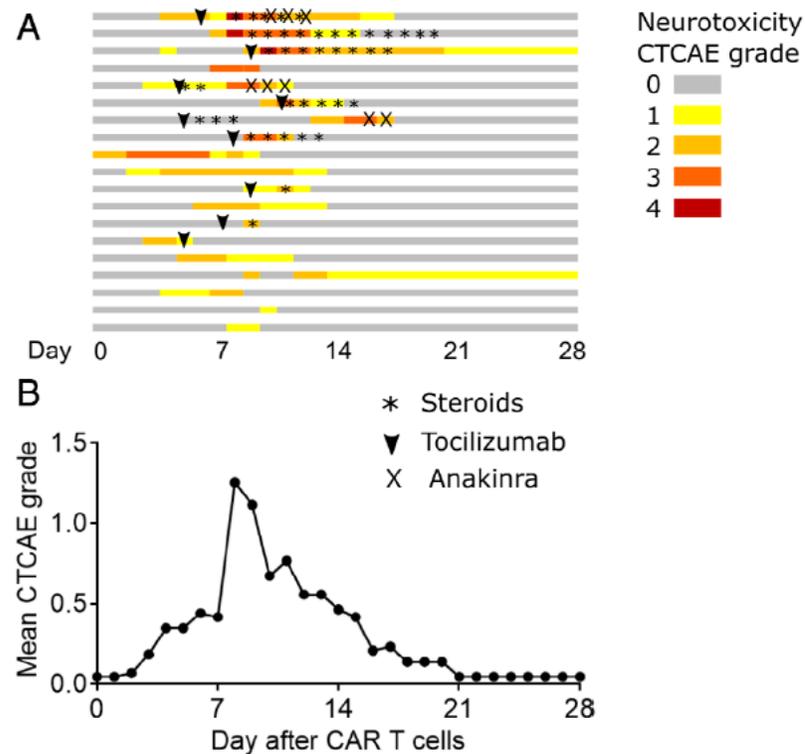
Table 1. Factors associated with neurotoxicity

Neurotoxicity CTCAE grade		Grade 0 ^a	Grade 1–2 ^a	Grade 3–5 ^a	Total	Univariate ^b	Multivariable ^c
comorbidities, n (%)	PN ⁱ	14 (47)	7 (23)	9 (30)	30	0.2	
	CNS involvement	6 (43)	5 (36)	3 (21)	14	0.2	
	Headache disorder	6 (43)	5 (36)	3 (21)	14	0.2	
	Other	5 (50)	2 (20)	3 (30)	10	0.7	
	ICH ^h	4 (67)	1 (17)	1 (17)	6	1	
	Seizures	2 (33)	2 (33)	2 (33)	6	0.3	
	Cog impairment ⁱ	1 (25)	2 (50)	1 (25)	4	0.1	
	MTX CNS toxicity ^j	1 (50)	1 (50)	0	2	0.4	
	Marrow disease, %	Median (range)	0.6 (0–97)	0.4 (0–93)	25.8 (0–97)	1.3 (0–97)	0.072
Total CD19 ⁺ cells in marrow, %	Median (range)	5.3 (0–99)	12.4 (0–93)	29.1 (0–97)	8.8 (0–99)	0.062	
CD8 ⁺ central memory enriched CAR-T cells ^k , n (%)	Selected	48 (67)	11 (15)	13 (18)	72 (54)	0.242	
Lymphodepletion regimen ^l , n (%)	Cy/Flu	58 (56)	23 (22)	23 (22)	104	0.11	0.0259
	Non-Cy/Flu	22 (76)	2 (7)	5 (17)	29		
CAR-T cell dose, n (%)	2 × 10 ⁵ cells/kg	20 (57)	10 (29)	5 (14)	35	<0.0001	0.0009
	2 × 10 ⁶ cells/kg	55 (64)	15 (17)	16 (19)	86		
	2 × 10 ⁷ cells/kg	5 (42)	0	7 (58)	12		
Cytokine release syndrome, n (%)	None (G 0)	35 (88)	5 (13)	0	40	<0.0001	n/a
	Mild (G 1–2)	44 (57)	19 (25)	14 (18)	77		
	Severe (G 3–5)	1 (6)	1 (6)	14 (88)	16		



Time of tocilizumab administration

- Early tocilizumab (pre-emptive/+2 days post infusion) is not recommended: might increase CNS IL-6 levels through binding of the IL-6 receptor in serum, potentially aggravating neurotoxicity
- Administration of tocilizumab and steroids in mild/early stage CRS was not associated with a change in neurotoxicity grade and did not impact on CAR-T cell engraftment and persistence

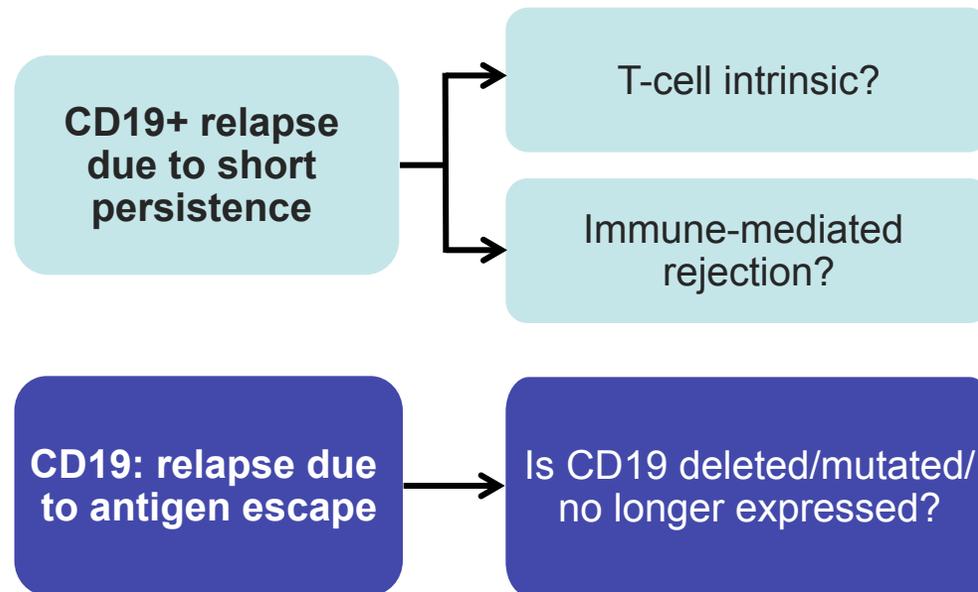


Early tocilizumab treatment of CRS is not associated with a change in neurotoxicity risk relative to CRS severity



Relapse after CAR-T in ALL

Eliana study (75 patients) : 1 patient had a CD19+ recurrence and 15 patients had CD19- (3 with concomitant CD19+ blasts); 6 patients had unknown CD19 status.



- Removal of CAR-recognized epitope as a result of alternative exon splicing forms of the CD19 gene where exon 2 was spliced out (*Sotillo et al. Cancer Discov. 2015*);
- Myeloid switch and loss of B lymphoid antigens in patients with Mixed-phenotype leukemia and MLL rearrangement (*Gardner et al. Blood, 2016*);
- Trafficking alteration of CD19 protein to the cell membrane of blast cells (*Braig et al. Blood, 2016*)
- Induction of resistance to chimeric antigen receptor T-cell therapy by transduction of a single leukemic B cell (*Ruella M et al. Nature Med, 2018*)

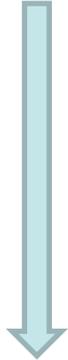


What do we want now?

- To know who needs **or not** HSCT
- Longer term results
- More effective CARs
 - Controlling the clonal escape (in fact already present and maybe more if pre-exposure to blinatumomab) : bispecific CARs
 - CARs for T-cell ALL but avoiding fratricide fight and not persisting too long
- Reduce toxicity of CARs:
 - Preemptive treatment with tocilizumab?
 - Other treatment to prevent and/or treat CRS & neurotoxicity:
 - Anti R-IL1 : anakinra (*Nat Medicine* 2018 x 2)
 - Anti-GMCSF : lenzilumab (Stern RM et al, *Blood* 2019)
 - Reduced affinity for target: **(CARPALL, NCT02443831)** : *Nature Medicine* 2019
 - Better design : Ying et al, *Nature Medicine* 2019
 - Inducible or suicidable CARs?

Possible concepts in a near future for B-ALLs

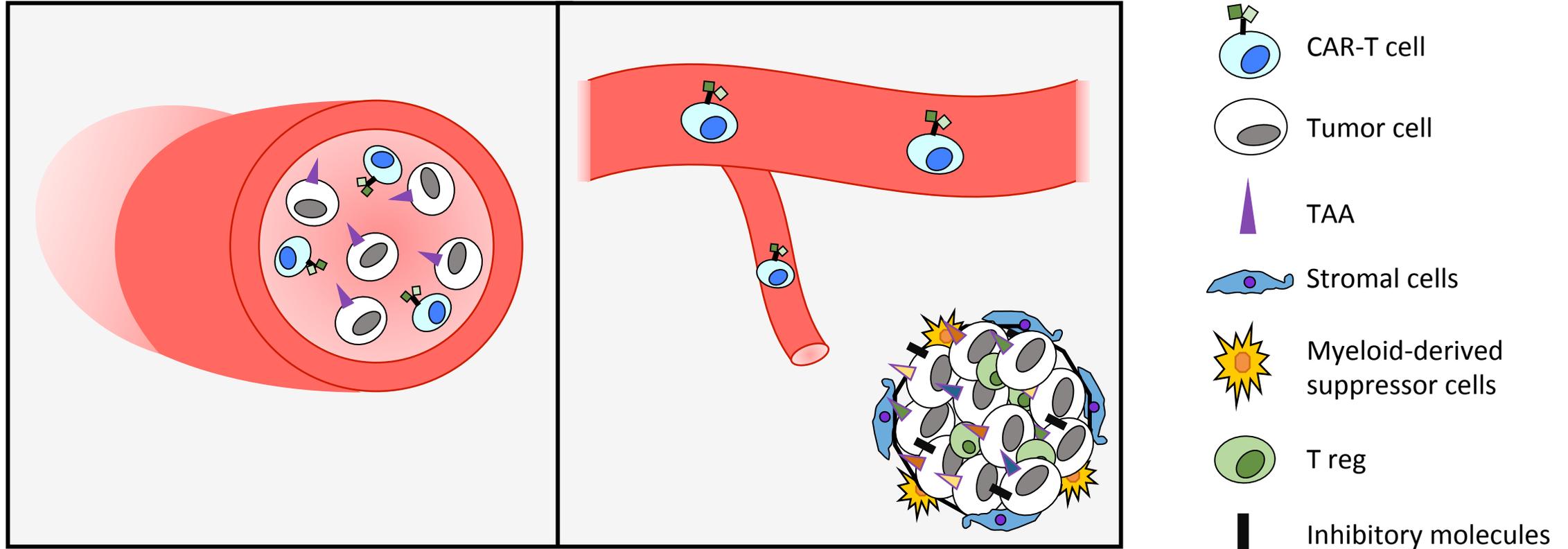
- Replace allo HSCT by persisting CARs
 - Advanced disease (relapse ≥ 2 , relapse post HSCT)
 - 1st high-risk relapse
 - 1st line very-high risk ALL (non responding to chemo)
 - 1st line HR ALL (replacement of prolonged intensive chemo?)



time

- Allogenic CARs: more a bridge to transplant except if repeated infusions are possible after milder LD chemotherapies

From hematological malignancies to solid tumors



Hurdles to overcome for targeting solid tumors

- Trafficking to the tumor site
- Tumor heterogeneity
- Unfavorable and immunosuppressive microenvironment

CAR-T cell therapy in neuro-oncology

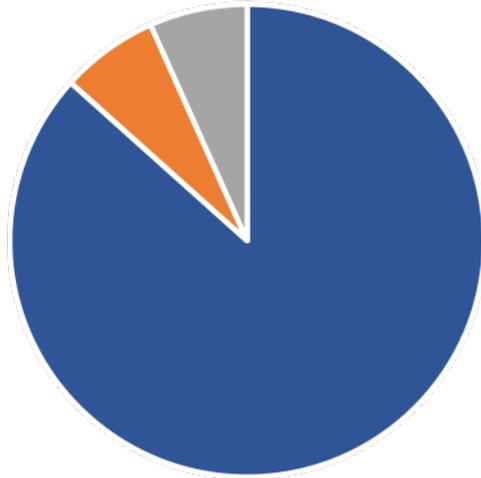
Disease	Target antigen	Type of CAR	Clinical trial	Phase	Location
Neuroblastoma	GD2	3rd generation	NCT02919046	I-II	Nanjing Children's Hospital (China)
		3rd generation + iC9	NCT03373097	I-II	Ospedale Pediatrico Bambino Gesù, Roma, Italy
		3rd generation + IL15 + iC9	NCT03721068	I	Lineberger Comprehensive Cancer Center - Chapel Hill, North Carolina
		2nd generation in invariant NKT cells	NCT03294954	I	Texas Children's Hospital Houston, Texas
		3rd generation + iC9 in VZV-specific T cells	NCT01953900	I	Texas Children's Hospital Houston, Texas
		4th generation	NCT02765243	II	Zhujiang Hospital of Southern Medical University, China
		2nd generation	NCT02761915	I	Great Ormond Street Hospital for Children NHS Foundation Trust United Kingdom
		2nd generation + C7R	NCT03635632	I	Texas Children's Hospital Houston, Texas
	L1CAM (CD171)	2nd generation + EGFRt in 1:1 CD4:CD8 T cell ratio	NCT02311621	I	Seattle Children's Hospital, Washington
EGFR	2nd generation + EGFRt	NCT03618381	I	Seattle Children's Hospital, Washington	
Glioblastoma	EGFRvIII	2nd generation + Pembrolizumab N/A	NCT03726515 NCT03283631	I I	University of Pennsylvania Duke University Medical Center, North Carolina
	IL13R α 2	2nd generation + CD19t in Tn/mem	NCT02208362	I	City of Hope Medical Center Duarte, California
	Her2	2nd generation + CD19t in Tn/mem	NCT03389230	I	City of Hope Medical Center Duarte, California

Summary of CAR-T cell active and recruiting clinical trials for brain tumors; source: Clinicaltrials.gov

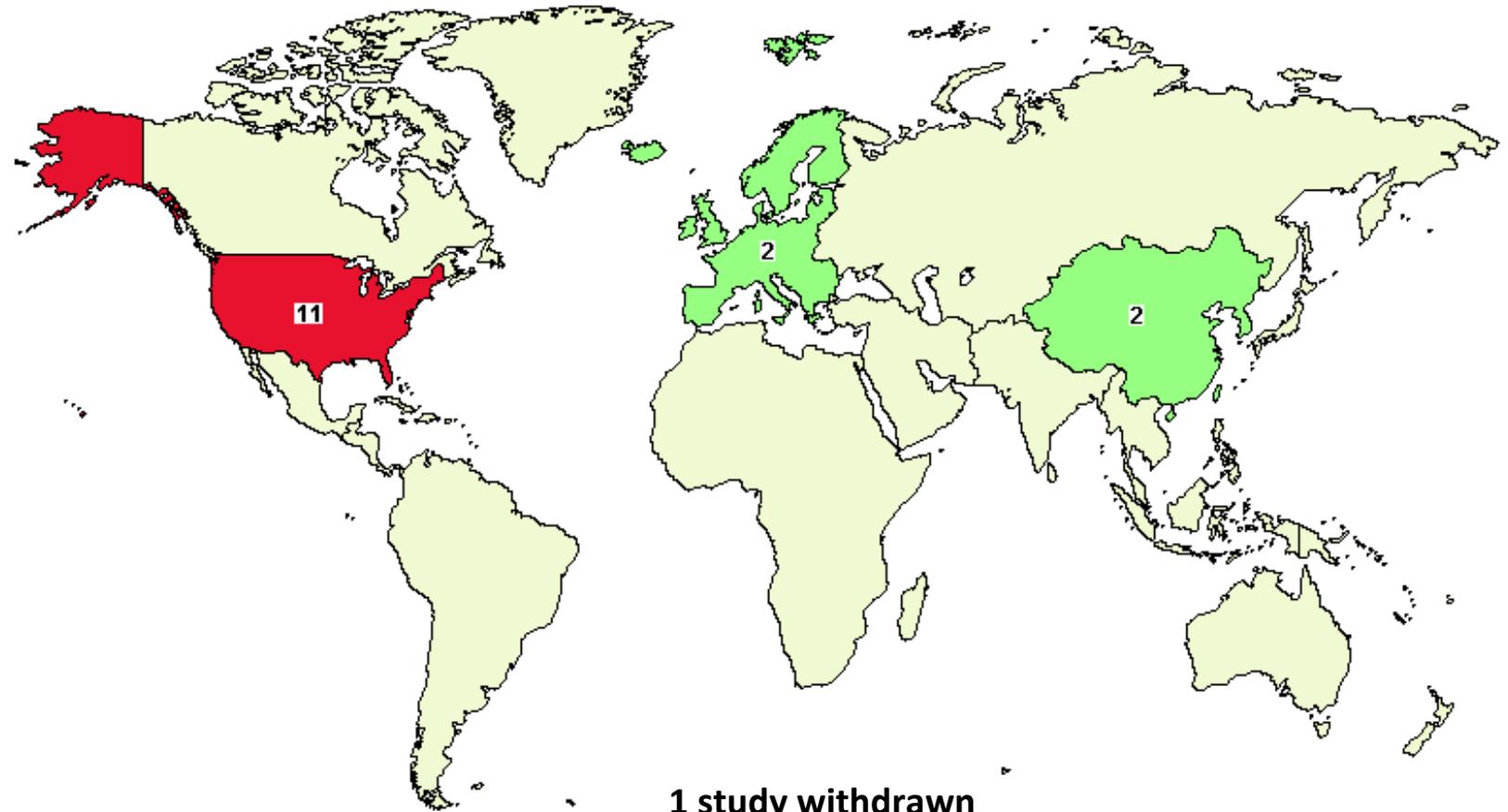


Targeting neuroblastoma

Target antigens



■ GD2 ■ L1CAM (CD171) ■ EGFR



1 study withdrawn
2 studies completed
2 studies active, not recruiting

Targeting neuroblastoma: *clinical evidence*

- **Product: GD2.z in EBV CTLs (NCT00085930)** (Baylor College of Medicine) → 11 patients treated, CR 27% (3/11) (2 sustained). Low T-cell persistence associated with longer survival. Transient pain (≤ grade 3) only at site of disease in 3/19 patients (*Louis et al. 2011; Pule et al. 2008*).
- **Product: iC9-GD2.28.Ox40.z (NCT01822652)** (Baylor College of Medicine) → 11 patients treated. No objective responses. T-cell expansion after lymphodepletion, but transient persistence; no benefit to addition of anti-PD-1 therapy (*Heczey et al. 2017*).
- **Product: L1CAM.z** (Seattle Children's Hospital, City of Hope) → 6 patients treated. No objective responses. No lymphodepletion; minimal T cell persistence. (*Park et al. 2007*).

Improving CAR-T cell efficacy in solid tumors: next generation CARs

Chemokine receptor engineering of CAR-T:

CCR4.CD30.CAR against HL

(Di Stasi A et al, Blood 2009, NCT03602157)

Matrix degrading enzyme engineering of CAR-T:

GD2.heparanase.CAR

(Caruana I et al, Nat Med 2015)

CAR-T + Immune Checkpoints:

- PD-1, CTLA4 in addition to CARs

(Li S Clin Cancer Res 2017)

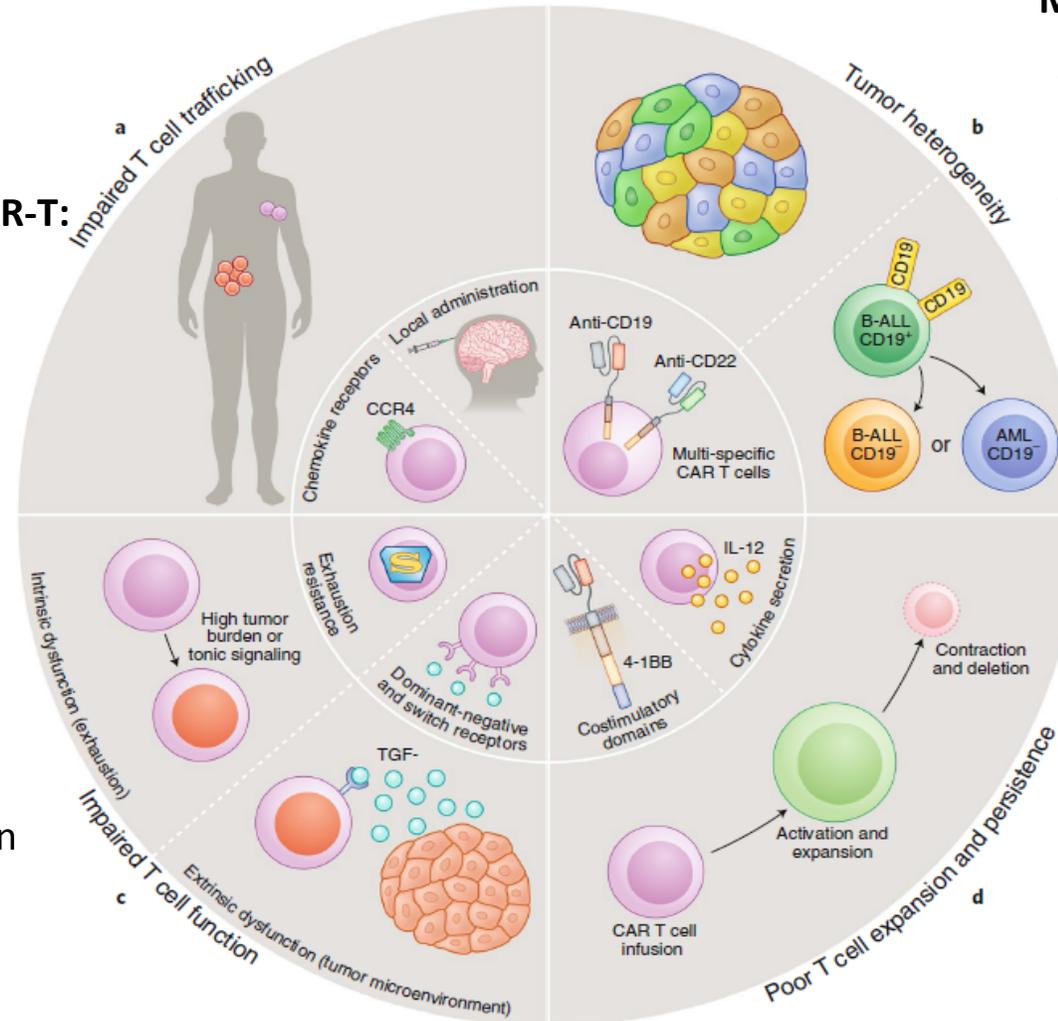
- UNIVERSAL CAR T resistant to PD-1 inhibition

(by CRISPR/Cas9 editing)

(Ren J, Clin Cancer Res 2017)

Some of these strategies are already exploited in clinical trials

(i.e. for Glioblastoma EGFRVII CAR + PEMBROLIZUMAB NCT03726515)



Multispecific CARs:

- Tandem CAR Her2/IL13Rα2

(Hedge M et al., J Clin Invest 2016)

- Trivalent CAR Her2/IL13Rα2 /EphA2

(Bielamowicz K Neuro Oncol. 2018)

Armored CAR-T:

- CEA.CAR+ IL12, CD30.CAR + IL12

Chmielewski M, Cancer Res 2011

- CD19.CAR+ IL18, MESO.CAR + IL18

Hu B Cell Rep 2017

- GD2.CAR+ IL15

Chen Y Clin Cancer Res. 2019

NCT03721068

Conclusions

1. Adoptive transfer of CAR modified T cells is a **realistic** therapeutic opportunity, although at very high cost!
2. Major breakthrough but still many questions!
3. Efficacy vs. **toxicity balance**.
4. Composition of the different cell products (different **gene delivery methods**.... NON-VIRAL: trasposons, mRNA; **T-cell subsets**...).
5. Need for “**suicide**” gene control?
6. Bridge to SCT or substitute for its need?
7. How and when CAR can be considered in the ALL strategy?
8. Engagement of large pharmaceutical companies ... still place for academic involvement?