The journey of ATMPs from early development to access in EU: challenges and opportunities

Researchers perspective

Franco Locatelli, MD, PhD

Università Cattolica del Sacro Cuore – Roma

Dipartimento di Oncoematologia Pediatrica

IRCCS, Ospedale Pediatrico Bambino Gesù, Roma

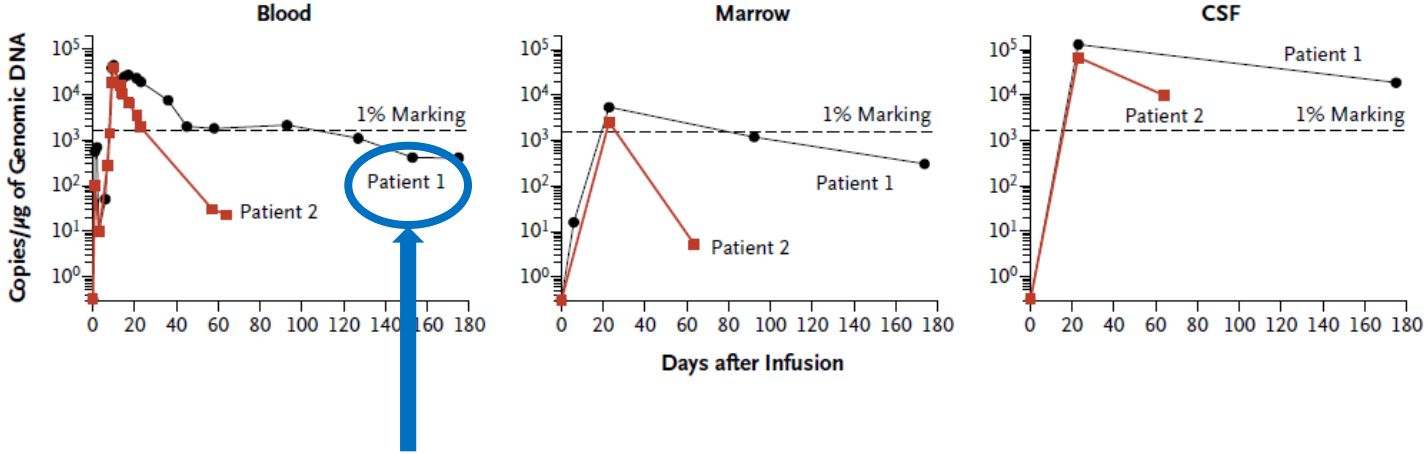
franco.locatelli@opbg.net

Disclosures of Franco Locatelli

Name of Company	Research Support	Employee	Consultant	Stockholder	Speaker's Bureau	Advisory Board	Other
Miltenyi					X		
Amgen					X	X	
Novartis					X	X	
BMS					X		
GILEAD					X		
Sanofi						X	
SOBI					X		
Vertex						X	

Emily, pt1, 12 year-leukemia-free in April 2024

B CTL019 Cells in Peripheral Blood, Bone Marrow, and CSF



Grupp S et al, N Engl J Med 2013

https://emilywhiteheadfoundation.org

EMA-approved CAR T-cell products

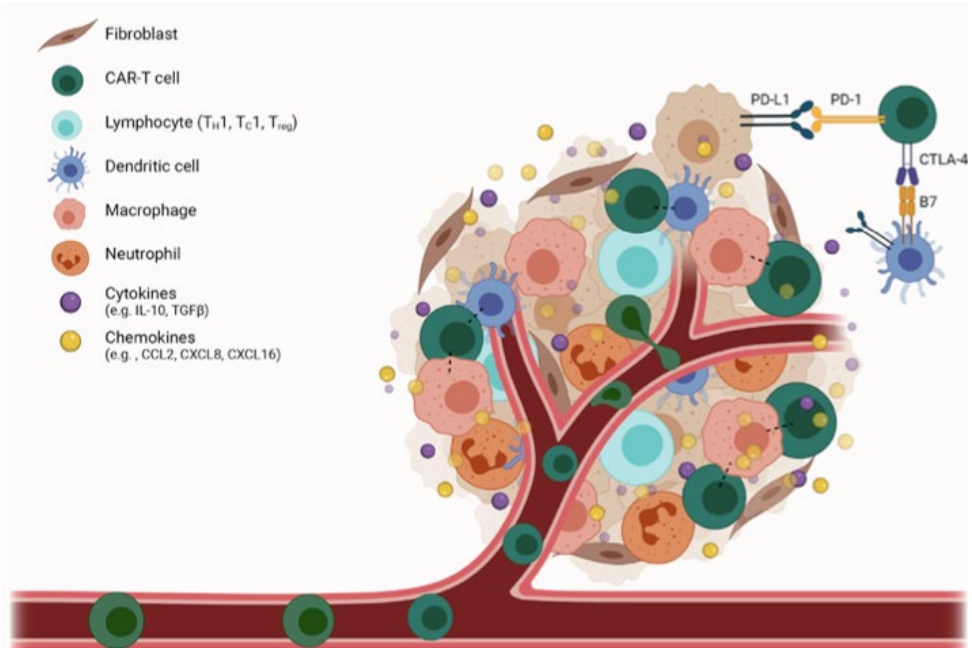
CAR T-cell product	Indication	Age	Pivotal trial	Autologous	Manufacturing
Kymriah (tisagenlecleucel) (CD19)	R/R BCP-ALL	≤25y	Single arm	✓	Centralised
	R/R LBCL	≥18y	Single arm	✓	Centralised
	R/R FL	18y	Single arm	✓	Centralised
Yescarta (axicabtagene ciloleucel) (CD19)	Refractory LBCL/FL	≥18y	Single arm	✓	Centralised
	R/R PMCL	≥18y	Single arm	✓	Centralised
Tecartus (brexucabtageneciloleuce) (CD19)	R/R mantle cell lymphoma	≥18y	Single arm	✓	Centralised
	R/R B-ALL	≥26y	Single arm	✓	Centralised
Breyanzi (lisocabtagene Maraleucel) (CD19)	R/R large B cell lymphoma	≥18y	Single arm	✓	Centralised
	R/R CLL/SLL	≥18y	Single arm	✓	Centralised
Abecma (idecabtagene) (BCMA)	R/R multiple myeloma	All	Single arm	✓	Centralised

Solid tumor: key challenges for CAR T-cell

Antigen dilemma
- Proportion of positive cells
- Level of expression
- Expression on normal counterpart

Immunosuppressive cells
- CAFs
- Tregs
- MDSCs

T-cell trafficking and infiltration
- Physical barriers
- Aberrant vasculature
- Chemokine receptors

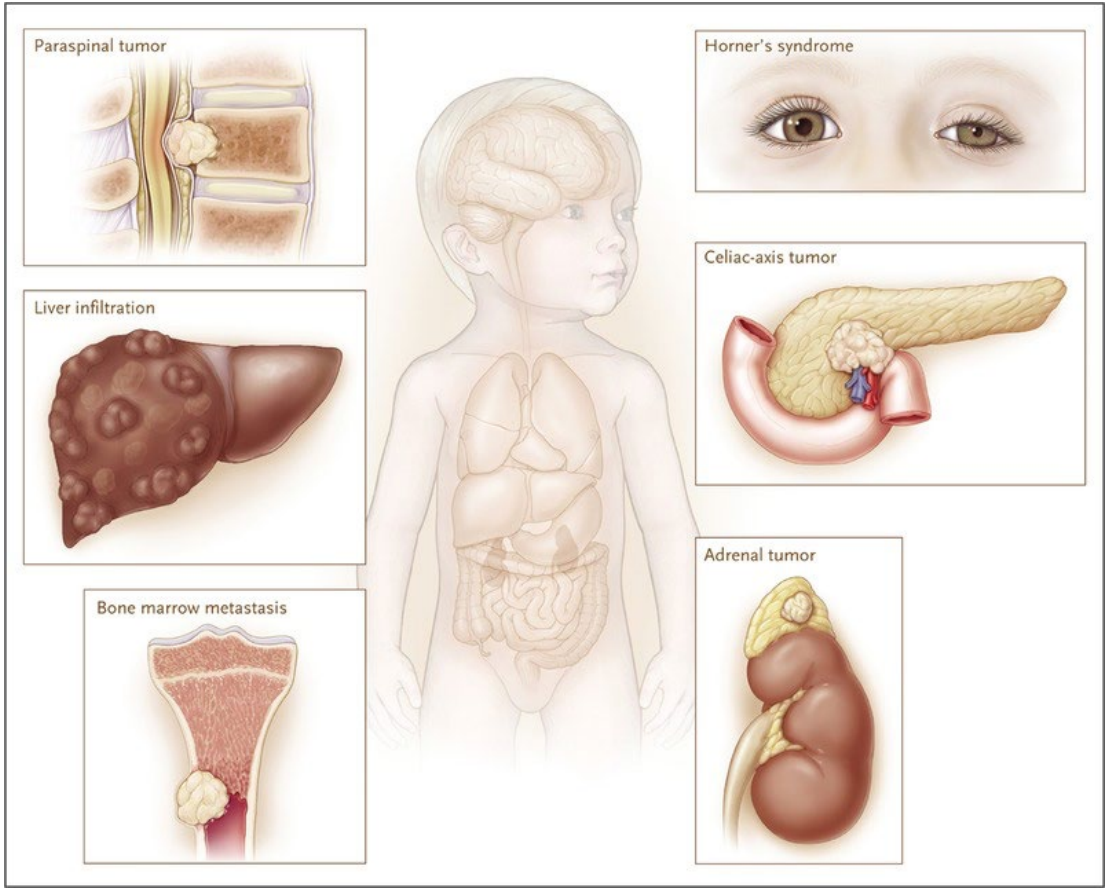


T-cell dysfunction and exhaustion
- Upregulation of inhibitory receptors

Immunosuppressive TME – soluble factors
- Reactive oxygen species
- High levels of adenosine
- Hypoxic environment
- Acid environment
- Elevated extracellular potassium

Foeng J et al, Cell Rep Med 2022

Neuroblastoma



Maris JM. Recent advances in neuroblastoma. NEJM 2010

Most common extracranial solid tumor of childhood

9,7 x 10^6 new diagnosis/year (about 110 new cases per year in Italy)

Prevalence according to age

< 1 year: 72,8%;
1-4 years: 21,7%;
5-9 years: 3,4%
>9 years: 1,5%

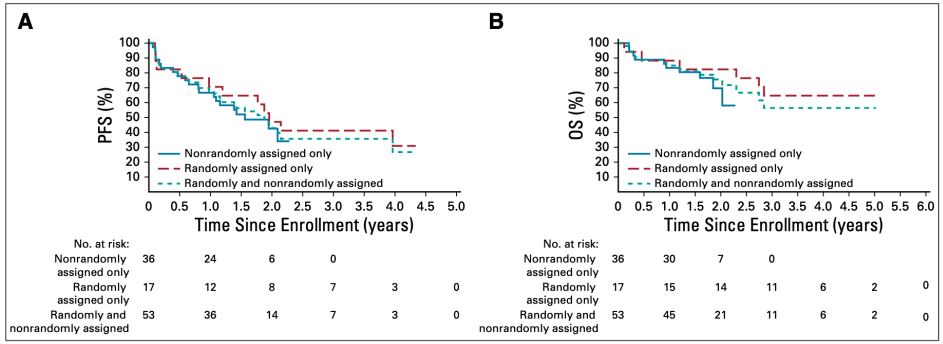
50% metastatic (bone, bone marrow, lymph nodes e liver) at diagnosis

Chances of cure with the current approaches:
- **Patients in first line = approximately 50%**
- **Patients with relapsed/refractory disease 10-15%**

R/R High-Risk Neuroblastoma: outcome with chemo-immunotherapy

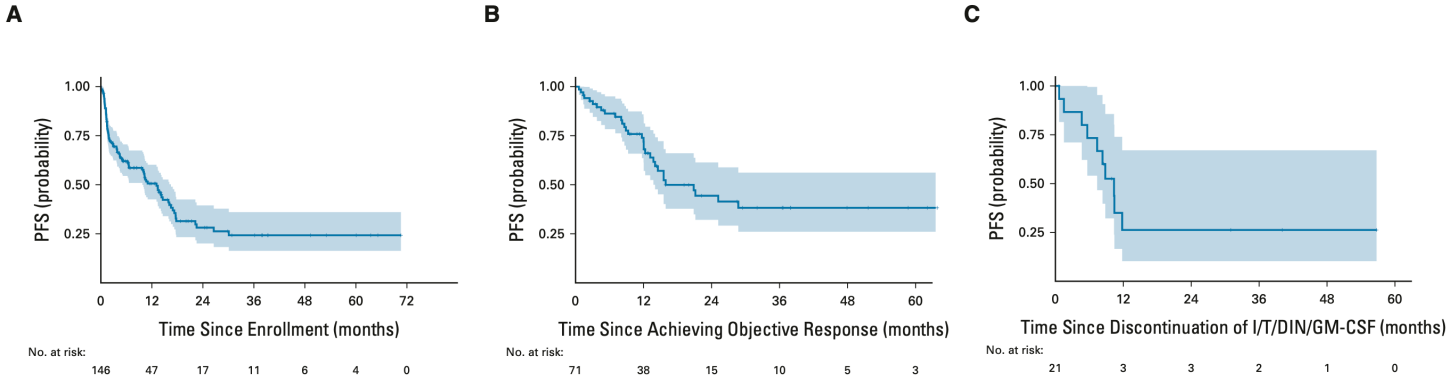
Children's Oncology Group ANBL1221 trial

ORR: 41.5%



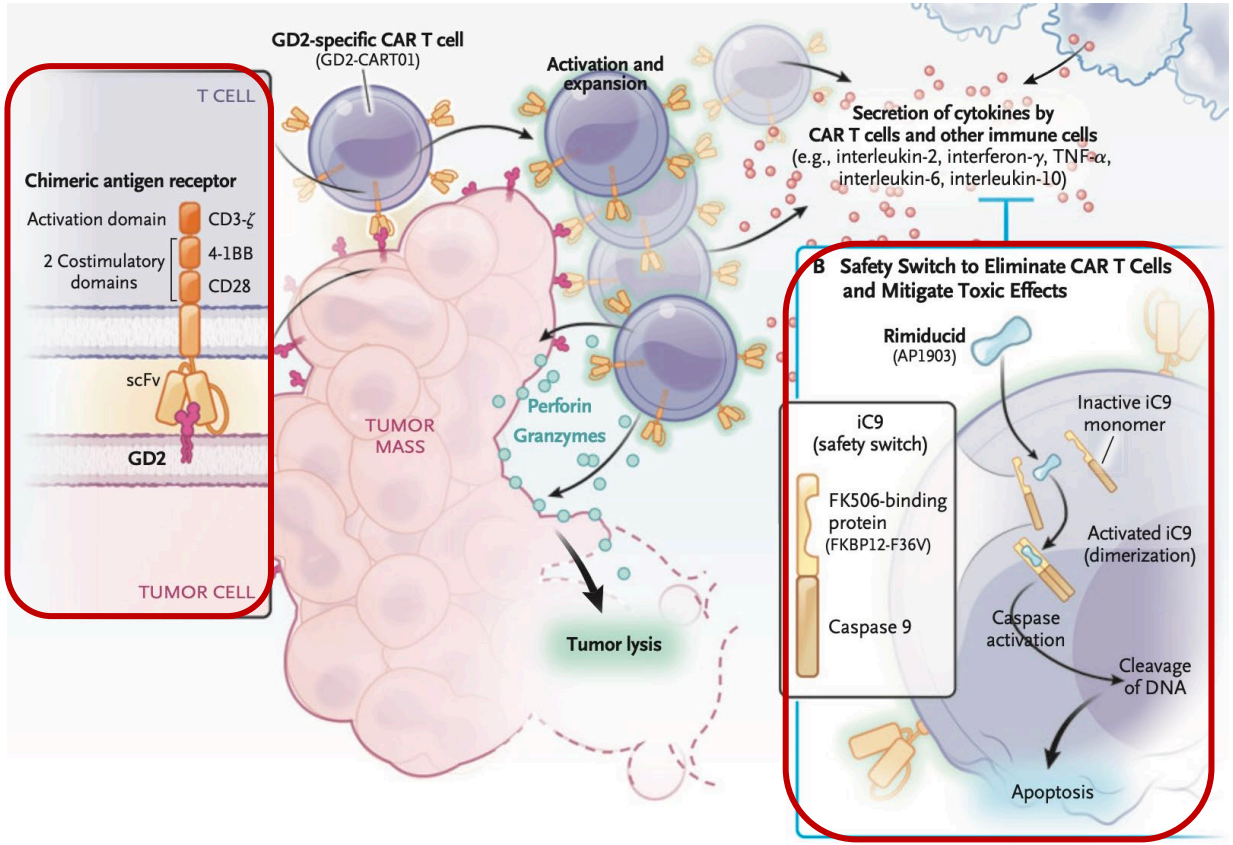
Real-world COG restrospective analysis

ORR: 49%



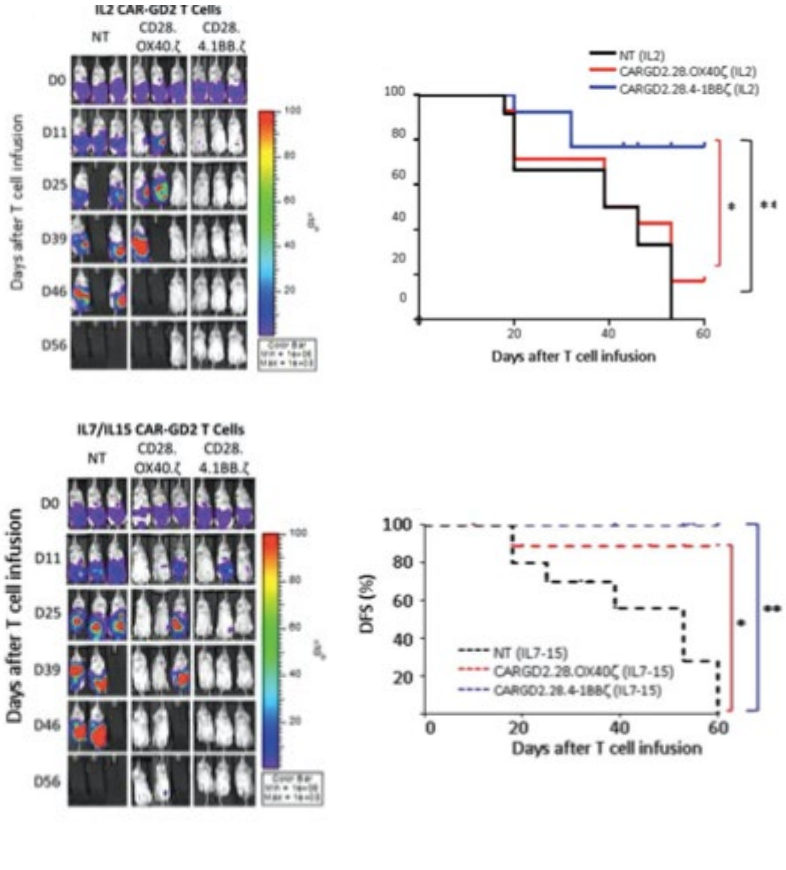
Mody R et al. JCO 2020
Lerman BJ et al. JCO 2022

GD2-CART01



Yeko OO and Longo D, NEJM 2023

Preclinical evaluation of the optimal construct and manufacturing for 3rd generation GD2-CAR



Quintarelli C, et al. Oncoimmmunology 2018

Clinical Trial Design and treatment schema

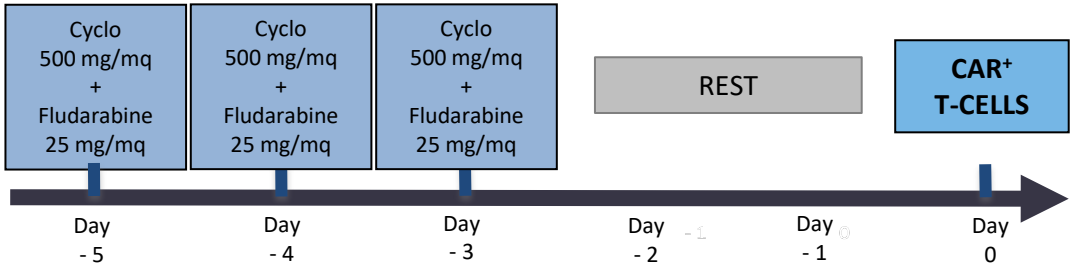
Phase I

- Five dose levels:
  - DL1: 1 x 10^6 cells/kg recipient body weight of CAR+ T cells
  - DL2: 2 x 10^6 cells/kg recipient body weight of CAR+ T cells
  - **DL3: 3 x 10^6 cells/kg recipient body weight of CAR+ T cells**
  - DL4: 6 x 10^6 cells/kg recipient body weight of CAR+ T cells
  - DL5: 10 x 10^6 cells/kg recipient body weight of CAR+ T cells

- Dose-finding 3+3 design

Phase II

- Treatment at the MTD/RD identified in phase I
- Fleming's two-stage design



Bambino Gesù
OSPEDALE PEDIATRICO

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

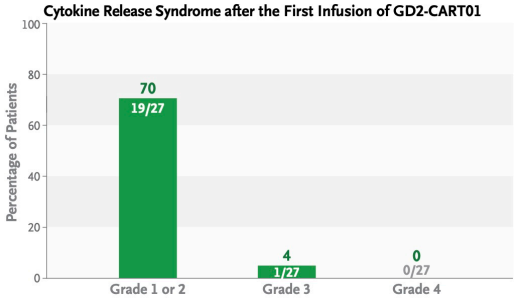
GD2-CART01 for Relapsed or Refractory High-Risk Neuroblastoma

F. Del Bufalo, B. De Angelis, I. Caruana, G. Del Baldo, M.A. De Ioris, A. Serra, A. Mastronuzzi, M.G. Cefalo, D. Pagliara, M. Amicucci, G. Li Pira, G. Leone, V. Bertaina, M. Sinibaldi, S. Di Cecca, M. Guercio, Z. Abbaszadeh, L. Iaffaldano, M. Gunetti, S. Iacovelli, R. Bugianesi, S. Macchia, M. Algeri, P. Merli, F. Galaverna, R. Abbas, M.C. Garganese, M.F. Villani, G.S. Colafati, F. Bonetti, M. Rabusin, K. Perruccio, V. Folsi, C. Quintarelli, and F. Locatelli, for the Precision Medicine Team–IRCCS Ospedale Pediatrico Bambino Gesù*

Table 1. Characteristics of the Patients at Baseline.*

Characteristic	All Patients (N=27)
Sex — no. (%)	
Male	18 (67)
Female	9 (33)
Median age (range) — yr	6.7 (2.7–18.6)
Median no. of previous treatments (range)	3 (1–6)
Disease status at enrollment — no. (%)	
Refractory	12 (44)
Relapsed	14 (52)
No evidence of disease after NB-HR01 first-line treatment†	1 (4)
Previous treatment with anti-GD2 monoclonal antibody — no. (%)	14 (52)
MYCN status — no. (%)	
Amplification	7 (26)
Gain	5 (19)
Normal	10 (37)
Unknown	5 (19)
Site of disease involvement — no. (%)	
Bone	21 (78)
Bone marrow	12 (44)
Lymph nodes	11 (41)
Abdomen	4 (15)
Paravertebral area	7 (26)
Thorax, pleura	2 (7)
Liver	1 (4)
Result of ^{123}I-labeled MIBG scan before infusion — no. (%)‡	
MIBG score ≤7	18 (67)
MIBG score >7	9 (33)

Feasibility and toxicities



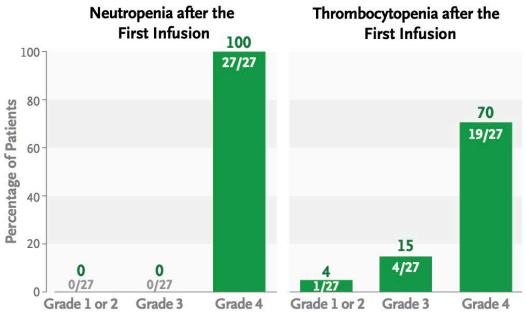


Table 2. Adverse Events in 27 Patients after the First Infusion of GD2-CART01.*

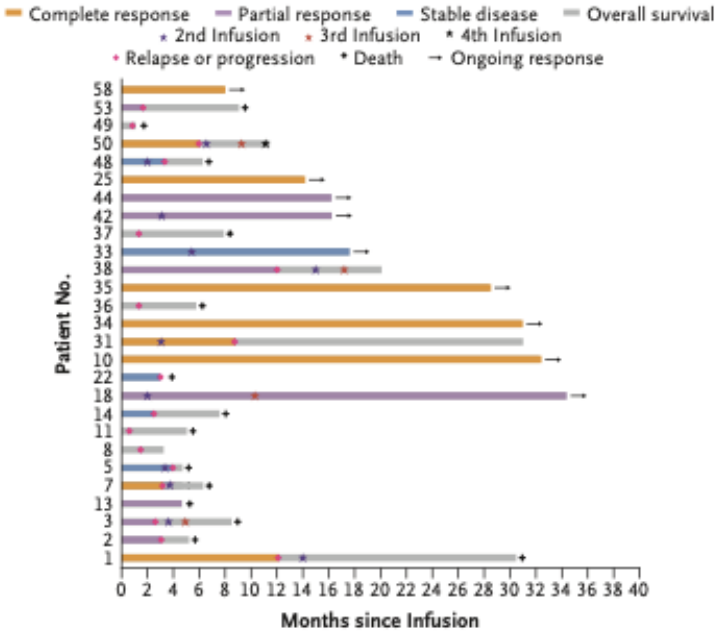
Event	Grade 1 or 2	Grade 3	Grade 4
		number of patients	
Cytokine release syndrome	19	1	0
Central neurotoxic effects	0	0	0
Peripheral neurotoxic effects or pain	6	0	0
Hematologic toxic effects			
Anemia	8	19	0
Neutropenia	0	0	27
Thrombocytopenia	1	4	19
Abnormal laboratory values			
Elevated alanine aminotransferase level, aspartate aminotransferase level, or both	13	7	0
Elevated bilirubin level	3	1	0
Electrolyte abnormalities	4	2	0
Miscellaneous			
Clostridium difficile infection	0	1	0
Rash	3	0	0
Dysuria	2	0	0
Brain hemorrhage	0	0	1

* With the exception of hematologic toxic effects, anomalies caused by the disease or by previous treatments were not included.

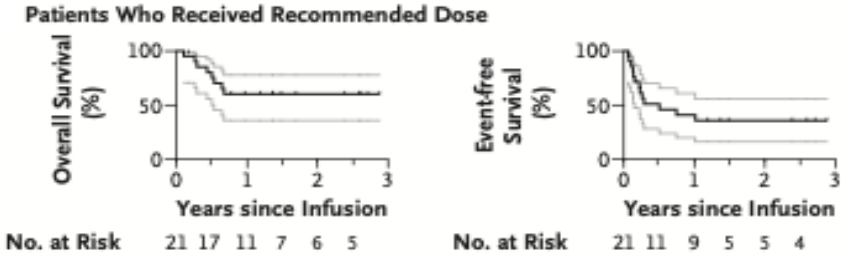
- **No DLTs were recorded in the phase I**
- **→ MTD/RD: 10,0 x 10^6 cells/kg of CAR+ T cells**

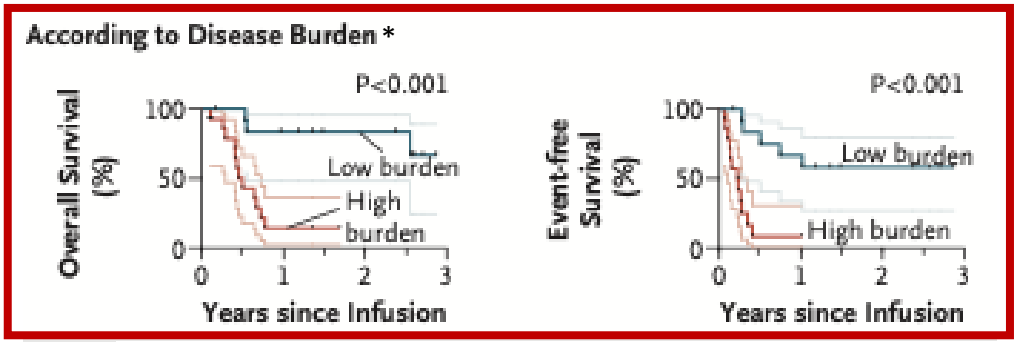
Del Bufalo F, […] Locatelli F. NEJM 2023

Clinical response and outcome



ORR: 63%





*Low disease burden:
- SIOPEN skeletal score ≤7
- Tumor mass with longest diameter <5cm
- BM infiltration ≤50%

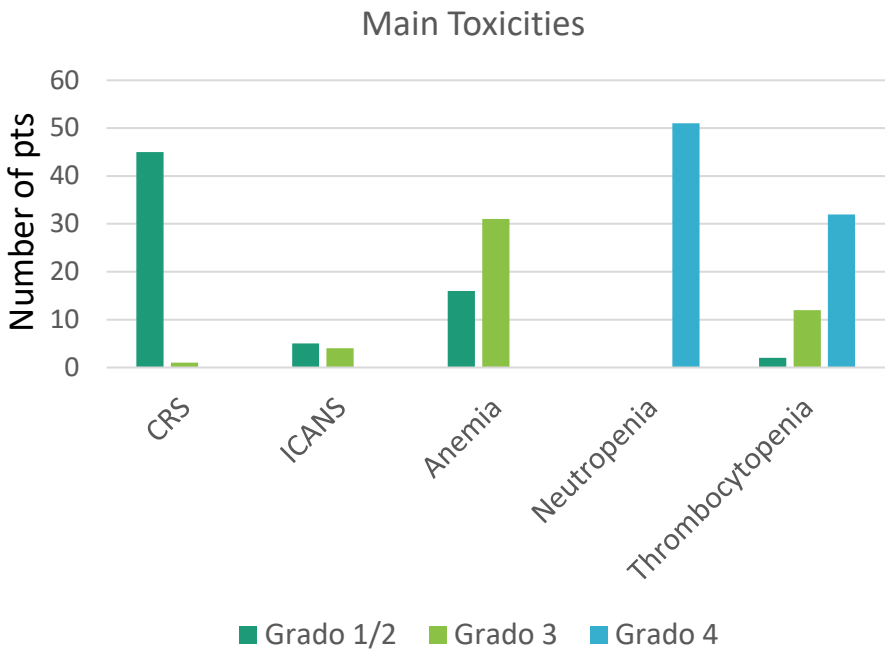
Del Bufalo F, […] Locatelli F. NEJM 2023

Updated cohort – patients characteristics

Sex	
M	35/51 (69%)
F	16/51 (31%)
Median age at infusion (range)	5,86 years (1,5 – 18,6)
Median number of previous treatments (range)	3 (1-6)
Disease status at enrollment	
Refractory	18/51 (35%)
Relapsed	26/51 (51%)
NED after NB-HR01	7/51 (14%)
Previous treatment with anti-GD2 monoclonal antibody	30/51 (59%)
N-MYC status	
Amplification	16/51 (31%)
Gain	5/51 (10%)
Non amplified	24/51 (37%)
Unknown	6/51 (12%)
Disease site involvement	
Bone	27/51 (53%)
Bone marrow	14/51 (27%)
Lymph-nodes	13/51 (25%)
Abdomen	10/51 (20%)
Paravertebral	8/51 (16%)
Thorax (pleura)	2/51 (4%)
Liver	1/51 (2%)
^{123}I-MIBG pre-infusion	
MIBG score >7	9/51 (18%)
MIBG score ≤7	42/51 (82%)

Data cut-off: 15.10.24

Updated cohort –toxicity



Adverse event	Grade 1 or 2	Grade 3	Grade 4
	Number of patients		
CRS	45	1	0
ICANS	5	4	0
Peripheral neurotoxicity/pain	12	0	0
Hematologic toxicity			
Anemia	16	31	0
Neutropenia	0	0	51
Thrombocytopenia	2	12	30
Laboratory values			
Elevated transaminases	18	11	0
Elevated bilirubin	6	2	0
Electrolyte abnormalities	9	5	0
Miscellaneous			
Acute kidney injury	0	1	0
Cl. difficilis infection	0	1	0
Skin rash	5	0	0
Dysuria	3	0	0
Brain hemorrhage	0	0	1

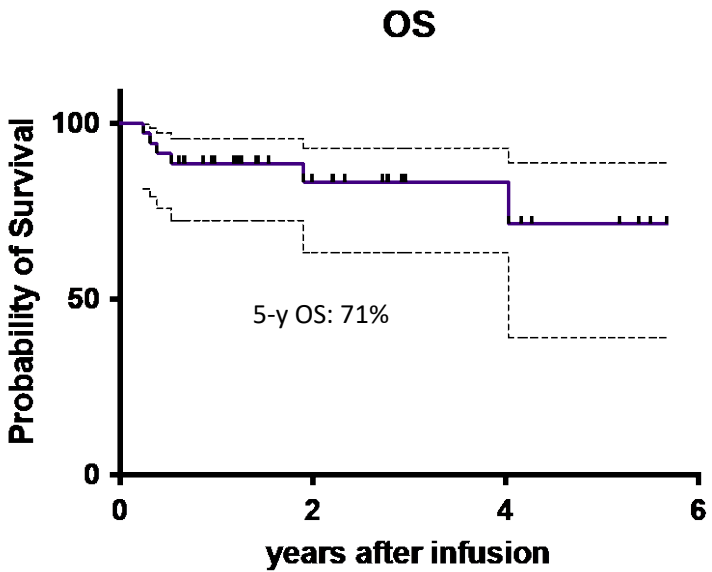
Data cut-off: 15.10.24

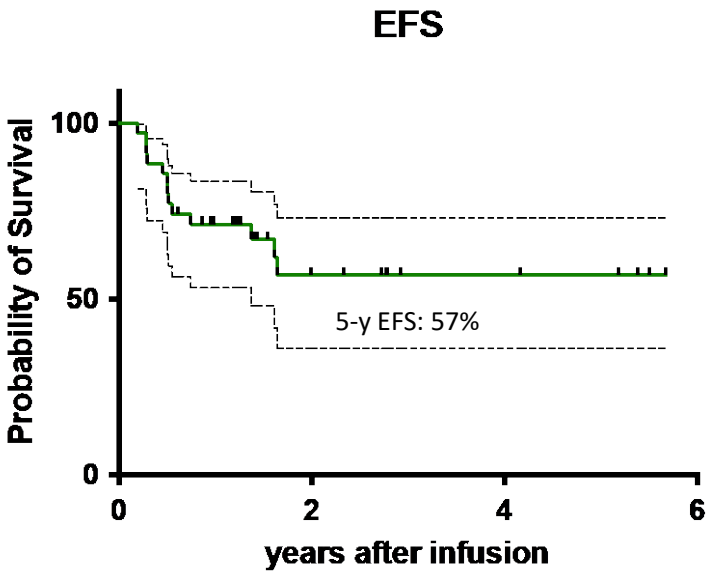
Unpublished – please do not post

Outcome of patients with low disease burden/NED, treated with the recommended dose (n=35)

CR: 20/35 (7 are the patients treated in NED)
PR: 7/35 (3 with negative PET and long-term duration of response)

ORR: 27/35 (77%)



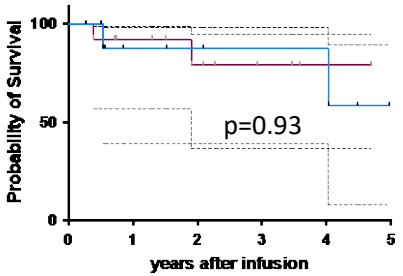


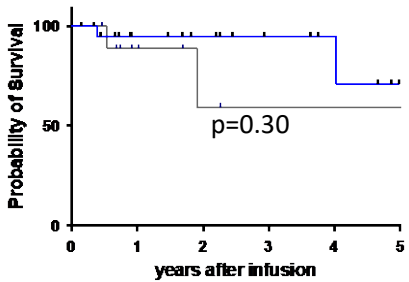
Data cut-off: 15.10.24

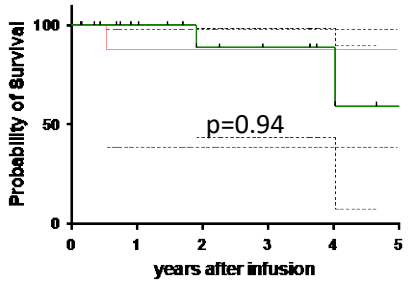
Unpublished – please do not post

Early treatment is associated with significantly improved outcomes

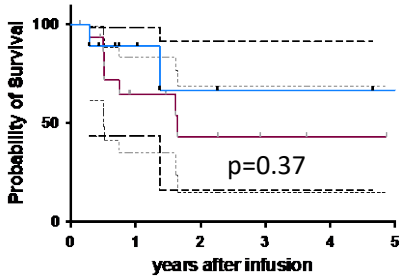
OS

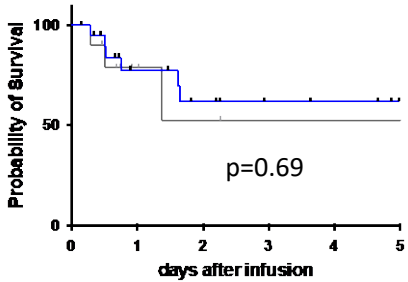


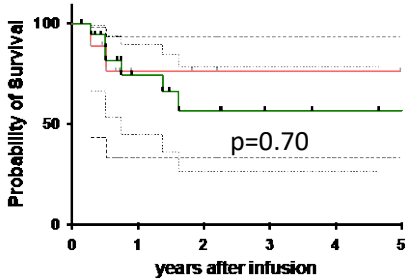




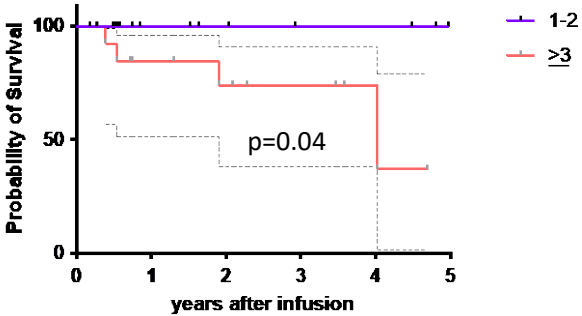
EFS



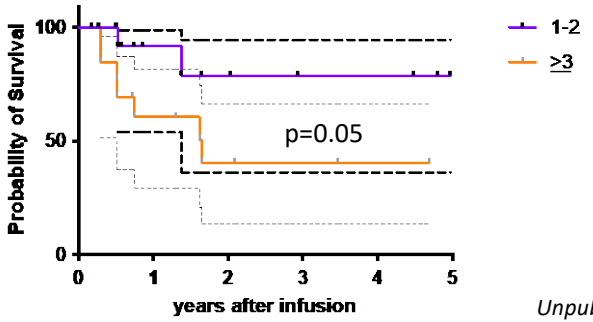




OS



EFS

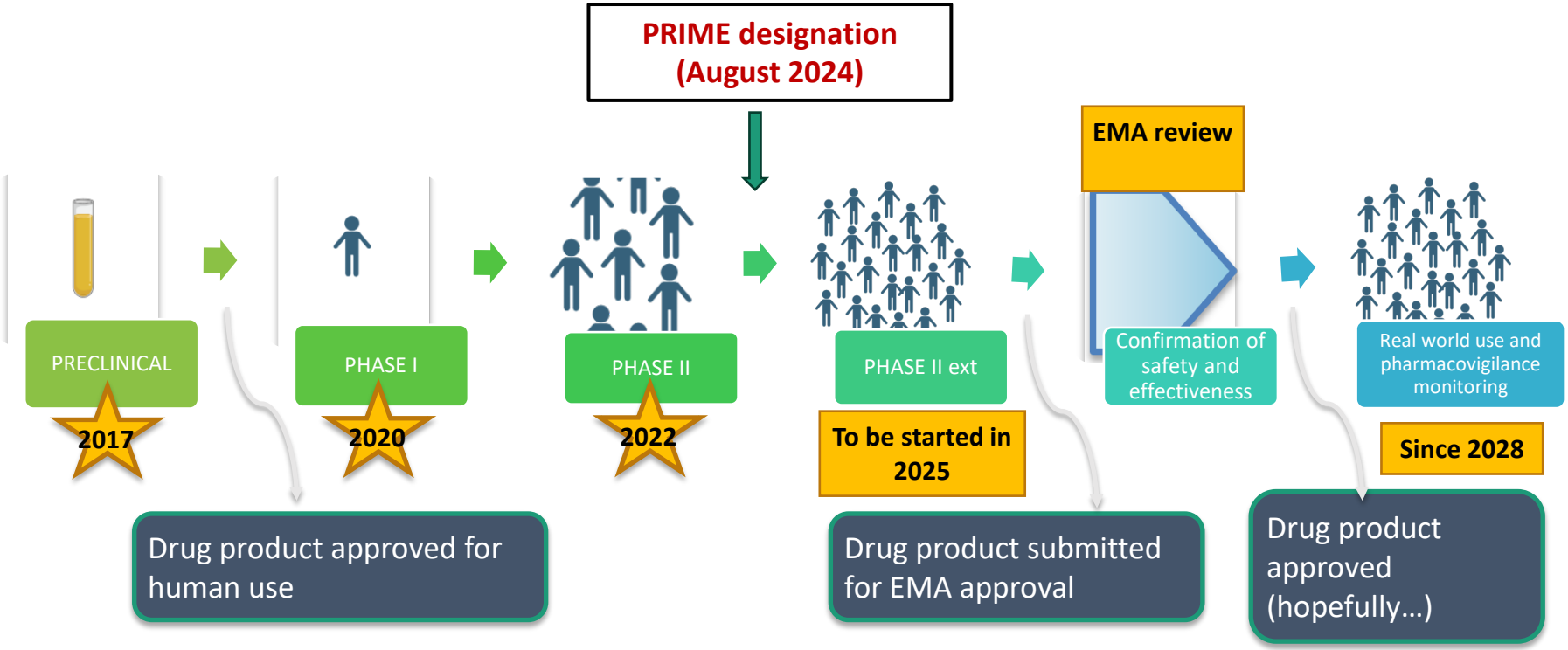


Data cut-off: 09.04.24

Unpublished – please do not post

What’s next?

GD2-CART01 for Neuroblastoma: update and next regulatory steps



EU-GD2-CAR01: multicenter, phase II trial (in collaboration with SIOPEN)

Phase II

Patients with NB relapsed/progressing during/after first line

↓

Reinduction treatment to reduce tumor burden
(choice of each treating center)

↓

Achievement of low tumor burden

↓

Enrollment in EU-GD2-CAR01

Bridging therapy ↓

GD2-CART01

Main inclusion criteria for treatment

- Diagnosis of High-Risk NBL
- Relapse/progression during/after first line treatment
- **Low disease burden (SIOPEN score ≤7; maximum diameter of soft tissue lesions ≤5±0.5 cm; BM infiltration <50%)**
- **No more than 2 previous lines of therapy**
- Disease under control (SD or MR or PR or CR) after reinduction
- Whenever technically feasible, a biopsy aimed at documenting GD2 expression is mandatory and GD2 positivity is required for inclusion
- Life expectancy > 3 months
- Age: 18 months – 25 years
- Voluntary informed consent is given
- Clinical performance status: ≥ 60% (Karnofsky or Lansky)

Clinical trial design – sample size

- Expected 3y-EFS in the experimental arm (GD2-CART01): 50%
- - 3y-EFS in the control arm (historical/AI-generated?): 30%
- - Delta of 20%
- - Alpha=5%
- - Two-sided test
- - Power: 80%
- → the number of patients required is around 50 patients.

- An interim analysis will be planned at the end of the second year
- A data safety monitoring committee will be appointed to oversee the study conduction

CAR.GD2 DP COSTS in the Academic setting

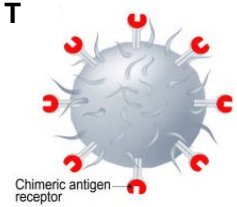
	Cost FOR SINGLE PATIENT (€)
Personell	9.000
Reagents (including virus)	55.000
Infrastructure Mantainance	1.000
Transport costs (including 1 dedicated dry shipper)	6.500
Total cost for single patient	71.500

Phase II, single arm study – budget estimate

Phase 2 Study - SINGLE ARM - Reference State Member Italy	
COST SUMMARY	**TOTAL (€)**
Overall Trial Development	80.260,00
Contracts, legal and financing	38.660,00
Regulatory submission and notifications	46.500,00
Trial Management and Trial Conduct (local project management)	54.625,00
Site Management	76.310,00
Data Management	71.440,00
Pharmacovigilance	44.070,00
Patient Treatments	1.633.699,00
Study Performance	66.798,00
Medicinal Product Supply Management	**3.271.800,00**
Monitoring and Audit	1.499.440,00
Data Provision, Statistical Analysis and Publication	16.800,00
Central Laboratories	1.758.050,00
Pharmacy	72.011,00
End of Trial	13.640,00
FINAL COST	**8.744.103,00**

GD2 as a suitable target for CAR T cells

GD2.CAR T

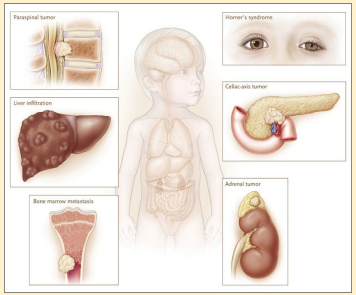


One biological drug for multiple tumors

neuroblastoma · glioma · medulloblastoma · alveolar rhabdomiosarcoma · Osteosarcoma · embrional rhabdomiosarcoma · Ewing's sarcoma · Desmoplastic small-round-cell tumor · small cell lung cancer · Non-small cell lung cancer · melanoma

High Risk metastatic neuroblastoma

Phase I/II Trial 2017



Pre-clinical results:

- Oncoimmunology. 2018 Mar 15;7(6):e1433518.
- J Immunother Cancer. 2021 Mar;9(3):e001502.
- J Hematol Oncol. 2021 Nov 12;14(1):191.

Clinical Application

Phase I/II Clinical trial NCT03373097

- N Engl J Med. 2023 Apr 6;388(14):1284-1295
- N Engl J Med. 2023 Jun 15;388(24):2303-2304

High Risk Brain tumors

Phase I Trial 2023

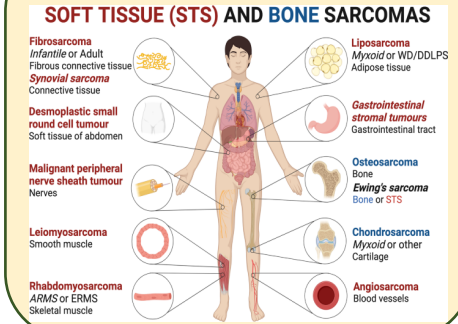
Pre-clinical results:

- Neuro Oncol. 2022 Jul 1;24(7):1150-1163.
- Clin Cancer Res. 2024 Jun 3;30(11):2545-2557

Clinical application

- Phase I/II Clinical trial **NCT05298995**.

High Risk metastatic sarcoma



Pre-clinical results:

- Paper under revision

Clinical application

Phase II Clinical trial NCT03373097

GD2-CAR T cells in CNS Tumours

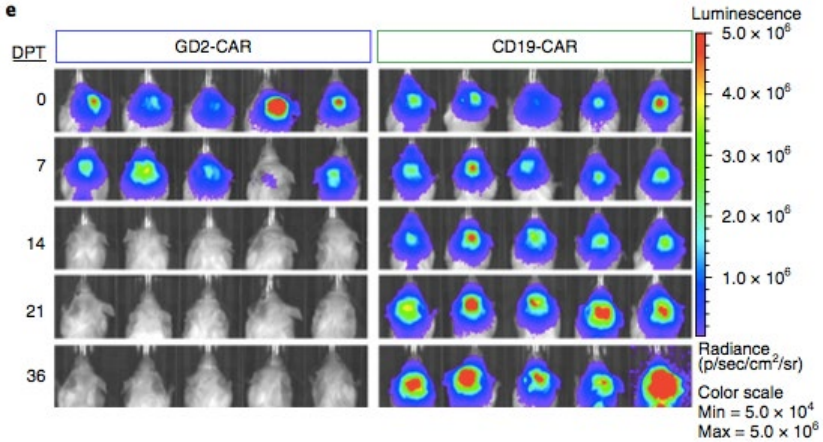
nature medicine

LETTERS

https://doi.org/10.1038/s41591-018-0006-x

Potent antitumor efficacy of anti-GD2 CAR T cells in H3-K27M$^+$ diffuse midline gliomas

Christopher W. Mount[1,2,3,12], Robbie G. Majzner[4,12], Shree Sundaresh[1], Evan P. Arnold[1], Meena Kadapakkam[4], Samuel Haile[4], Louai Labanieh[4,5], Esther Hulleman[6], Pamelyn J. Woo[1], Skyler P. Rietberg[4], Hannes Vogel[1,4,7,8], Michelle Monje[1,4,7,8,9,10]* and Crystal L. Mackall[4,9,11]*



Article

GD2-CAR T cell therapy for H3K27M-mutated diffuse midline gliomas

https://doi.org/10.1038/s41586-022-04489-4

Received: 2 August 2021

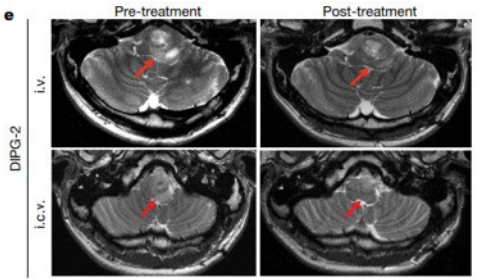
Accepted: 28 January 2022

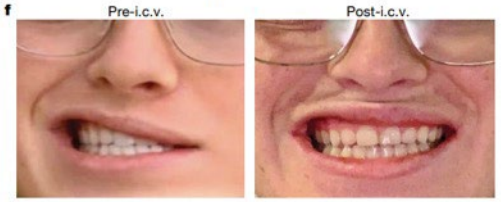
Published online: 7 February 2022

Open access

Check for updates

Robbie G. Majzner[1,2,3,13], Sneha Ramakrishna[1,2,13], Kristen W. Yeom[4], Shabnum Patel[1], Harshini Chinnasamy[1], Liora M. Schultz[1,2], Rebecca M. Richards[1,2], Li Jiang[3], Valentin Barsan[1,2], Rebecca Mancusi[5], Anna C. Geraghty[6], Zinaida Good[1,3,7], Aaron Y. Mochizuki[8], Shawn M. Gillespie[6], Angus Martin Shaw Toland[6], Jasia Mahdi[6], Agnes Reschke[1,2], Esther H. Nie[6], Isabelle J. Chau[6], Maria Caterina Rotiroti[2], Christopher W. Mount[6], Christina Baggott[1], Sharon Mavroukakis[1], Emily Egeler[1], Jennifer Moon[1], Courtney Erickson[1], Sean Green[2], Michael Kunicki[1,2], Michelle Fujimoto[1,2], Zach Ehlinger[2], Warren Reynolds[2], Sreevidya Kurra[2], Katherine E. Warren[5], Snehit Prabhu[1], Hannes Vogel[9], Lindsey Rasmussen[9], Timothy T. Cornell[9], Sonia Partap[6], Paul G. Fisher[6], Cynthia J. Campen[6], Mariella G. Filbin[5], Gerald Grant[10], Bita Sahaf[1,2], Kara L. Davis[1,2], Steven A. Feldman[1], Crystal L. Mackall[1,2,3,11,14] & Michelle Monje[1,2,6,8,10,12,14]





Third Generation CAR Targeting GD2

Platform: RETROVIRUS

Phase I study of anti-GD2 Chimeric Antigen Receptor-Expressing T cells in pediatric and young adult patients with relapsed/refractory CNS Tumors

ClinicalTrials.gov Identifier: NCT05298995

Active for recruitment

CAR T-cell pipeline in development and manufactured at OPBG GMP-facility

At the patient bedside

- **GD2-CAR T cells in neuroblastoma;**
- **GD2-CAR T cells in CNS neoplasia;**

At pre-clinical levels:

- **CD30-CAR T cells in HD/ALCL;**
- **FOLR-1 CAR T cells in infant AML, OS and other solid tumours;**
- **B7H3 in CNS and solid tumours;**
- **Allogeneic CD123 CAR NK cells in AML;**
- **X-antigen CAR T cells in AML.**

In collaboration with Pharma Industries and already at patient's bedside

- **PEBL CD7CAR T cells in T-ALL;**
- **CD19 CAR T cells in B-cell mediated AID.**

ACKNOWLEDGEMENTS

Department of Hematology/ Oncology

Clinical Unit

Gene and Cell Therapy Unit

Concetta Quintarelli, Biagio De Angelis, Francesca Del Bufalo, ***Enrico Velardi, Chiara Agrati, Mara Vinci***

Fred Hutchinson Cancer Research Center

Soheil Meshinchi

Cell Manipulation Unit

Giuseppina Li Pira

Simone Biagini

GMP Facility

Monica Gunetti

Stefano Iacovelli

Rossana Bugianesi

Stefania Macchia

All Team!

National University of Singapore

Dario Campana, Allen Yeoh Eng Jug, Elaine Coustan-Smith

Medisix

Andrew Bruce, Peiying Chuan

BCM/CAGT Houston

Malcolm Brenner