ACC MED

SEZIONE DI EMATOLOGIA

CONVEGNO

GRANDANGOLO 2020 IN EMATOLOGIA

SELEZIONE E ANALISI RAGIONATA DEI PIÙ RECENTI DATI SCIENTIFICI

XI EDIZIONE

DIGITAL EVENT, 4-5-16-17 NOVEMBRE 2020

Esperienza dell'ematologia di Monza nelle LLA

Adriana Balduzzi, Monza

Novel approach for children, adolescents and young adults with B-ALL

- most CAYA cured by modern risk-adapted chemotherapy regimens
- poor outcome of 10–15% patients suffering R/R disease
- dismal outcome of VHR B-ALL (8%) defined by persistent MRD+ EOC
- HSCT usual approach but high morbidity, low mortality, lack of efficacy if MRD+
- further intensification of cytotoxic chemotherapy regimens prevented by excess of toxicity and lack of efficacy prevent
- novel immune-based therapies under investigation
- CAR-T cells targeting CD19 demonstrated unparalleled responses in r/r ALL in multiple trials

ALL: acute lymphoblastic leukemia; BCA: B-cell aplasia; CAYA: children, adolescents, young adults; EOC: end of consolidation; EOI: end of induction; LT long-term; MRD: minimal residual disease, R/R: relapsed/refractory; VHR: very high risk

Topics - CAR-T in Pediatric ALL, Monza

Topics

- CAR-t overall
  - London: CARPALL
  - Barcelona: ARI-0001 CART-BE-01 Trial
  - Monza: FT01-CARCIK
- *Tisagenlecleucel*
  - efficacy & toxicity
    - ELIANA
    - B2001X
  - practicalities
    - enrollment
    - apheresis
    - bridge chemotherapy
    - infusion
    - CRS
    - neurotoxicity
    - flow cytometry monitoring
- Open questions & novel approaches
- Conclusions

CAR: chimeric antigen receptor, r/r: relapsed/refractory; ALL: acute lymphoblastic leukemia; CRS: cytokine release syndrome

Academic products - CAR-T in Pediatric ALL, Monza

LETTERS
https://doi.org/10.1038/s41591-019-0549-5
nature medicine

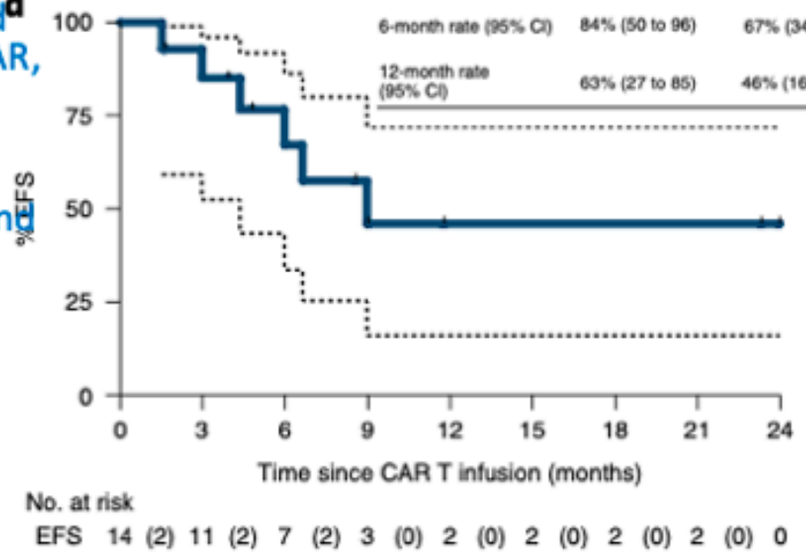
CARPALL

Enhanced CAR T cell expansion and prolonged persistence in pediatric patients with ALL treated with a low-affinity CD19 CAR

Sara Ghorashian[1], Anne Marijn Kramer[1], Shimobi Onuoha[2], Gary Wright[3], Jack Bartram[3], Rachel Richardson[1], Sarah J. Albon[1], Joan Casanovas-Company[1], Fernanda Castro[4], Bilyana Popova[4], Krystle Villanueva[4], Jenny Yeung[1], Winston Vetharoy[1], Aleks Guvenel[1], Patrycja A. Wawrzyniecka[5], Leila Mekkaoui[2], Gordon Weng-Kit Cheung[5], Danielle Pinner[3], Jan Chu[3], Giovanna Lucchini[3], Juliana Silva[3], Oana Ciocarlie[3], Arina Lazareva[3], Sarah Inglott[3], Kimberly C. Gulrukh Ahsan[6], Mathieu Ferrari[2], Somayya Manzoor[2], Kim Champion[4], Andre Lopes[4], Allan Hackshaw[4], Farzin Farzaneh[8], Robert Chiesa[3], Kancha Sujith Samarasinghe[3], Nicholas Goulden[3], Ajay Vora[3], Paul Veys[3], Rachael Martin A. Pule[5] and Persis J. Amrolia[1,3]*

lentivirally transduced[d] second-generation CAR, CD8-derived stalk/transmembrane region, 4-1BB co-stimulatory domain and a CD3ζ chain

	OS n = 14	EFS n = 14
Number of events	5 (36%)	6 (43%)
Median in months	16.5	9.97
6-month rate (95% CI)	84% (50 to 96)	67% (34 to 86)
12-month rate (95% CI)	63% (27 to 85)	46% (16 to 72)



CART-BE-01

Molecular Therapy
Available online 20 September 2020
In Press, Corrected Proof
CellPress

Original Article

CART19-BE-01: A Multicenter Trial of ARI-0001 Cell Therapy in Patients with CD19+ Relapsed/Refractory Malignancies

Valentín Ortíz-Maldonado[1], Susana Rives[2,3], Maria Castellà[1,4], Anna Alonso-Saladrigues[2], Daniel Benítez-Ribas[4,5], Miguel Caballero-Baños[5], Tycho Baumann[3,4], Joan Cid[4,6], Enric Garcia-Rey[7], Cristina Llanos[8], Montserrat Torrebadell[3,9,10], Neus Villamor[4,11,12], Eva Giné[1,4,12], Marina Díaz-Beyá[1,4], Laia Guardia[1], Mercedes Montoro[1], Albert Català[2,3], Anna Faura[2] ... Julio Delgado[1,4,12,15]

Academic CAR-T in Barcelona, Target population: ad + ped patients with relapsed/refractory CD19+ B-cell malignancies: 54 patients enrolled, 47 infused - 38 r/r ALL (27 adults, 11 children)

ARI-001: 2nd generation 41BB construct, lentiviral transduction, GMP production (ClinicMacs Prodigy)

Two lentiviral CAR19 T-cell approaches with 4–1BB- and CD3z:

- CTL019/tisagenlecleucel (scFV: FCM63)
- ARI001 cells (scFV: A3B1)

Bone Marrow Transplantation
https://doi.org/10.1038/s41409-020-01027-6

ARTICLE

Kinetics of humoral deficiency in CART19-treated children and young adults with acute lymphoblastic leukaemia

A. Deyà-Martínez[1,2] · A. Alonso-Saladrigues[3] · A. P. García[1,2] · A. Faura[3] · M. Torrebadell[3] · A. Vlagea · A. Català[3,4,5] · A. Esteve-Solé[1,2] · M. Juan[2,4,5] · S. Rives[3,4,5] · L. Alsina[1,2,5]

Academic products - CAR-T in Pediatric ALL, Monza

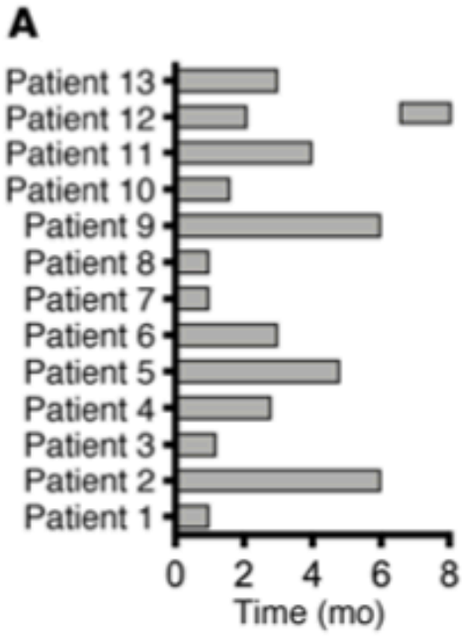
FT01-CARCIK

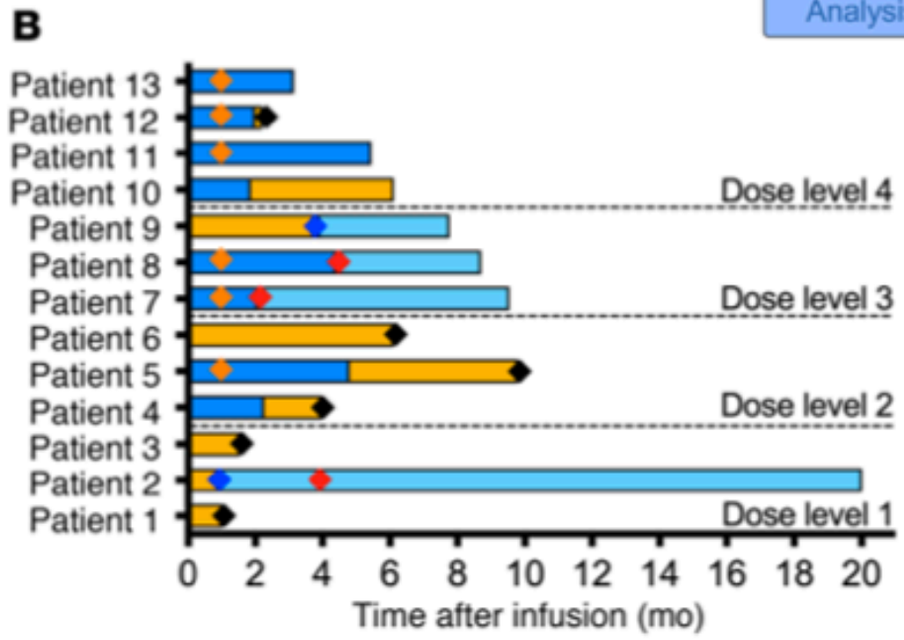
The Journal of Clinical Investigation

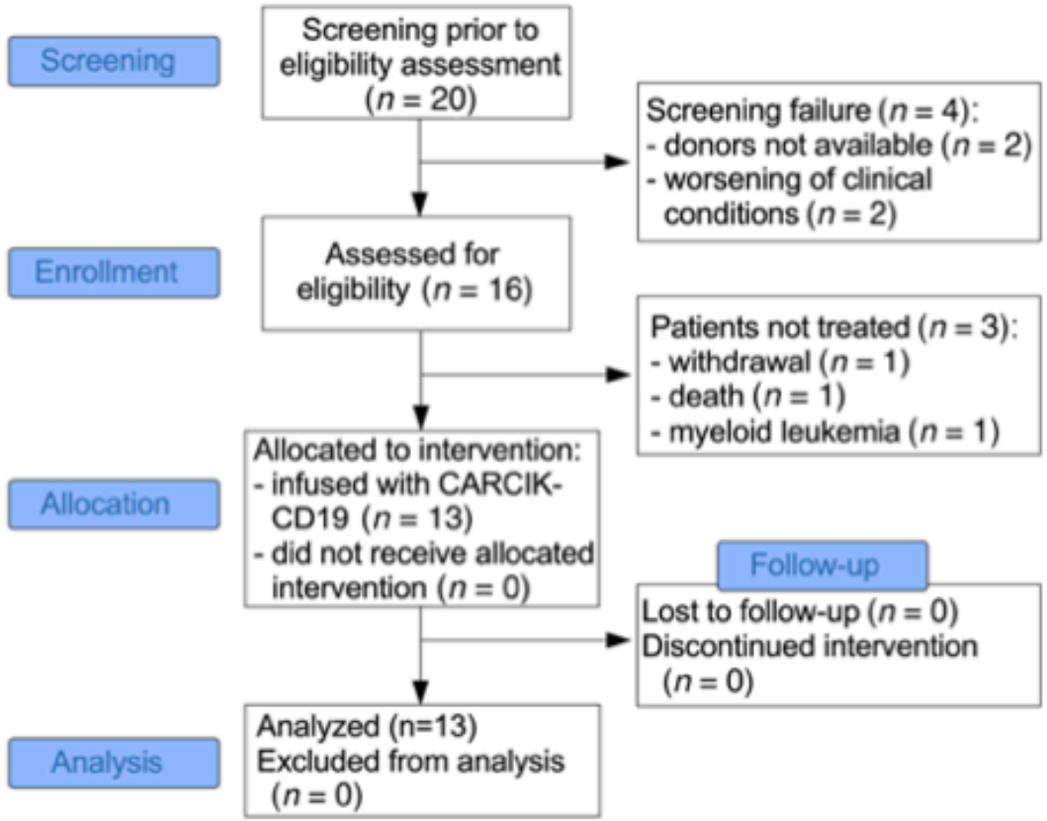
Sleeping Beauty–engineered CAR T cells achieve antileukemic activity without severe toxicities

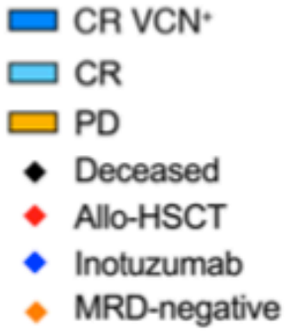
Chiara F. Magnani,[1] Giuseppe Gaipa,[1,2] Federico Lussana,[3] Daniela Belotti,[3,4] Giuseppe Gritti,[3] Sara Napolitano,[1] Giada Matera,[1,2] Benedetta Cabiati,[1,2] Chiara Buracchi,[1] Gianmaria Borleri,[3] Grazia Fazio,[1] Silvia Zaninelli,[4] Sarah Tettamanti,[1] Stefania Cesana,[1,2] Valentina Colombo,[1,2] Michele Quaroni,[1,2] Giovanni Cazzaniga,[1] Attilio Rovelli,[5] Ettore Biagi,[1,5] Stefania Galimberti,[7] Andrea Calabria,[8] Fabrizio Benedicenti,[8] Eugenio Montini,[8] Silvia Ferrari,[3] Martino Introna,[3,6] Adriana Balduzzi,[4,5] Maria Grazia Valsecchi,[7] Giuseppe Dastoli,[1] Alessandro Rambaldi,[3,9] and Andrea Biondi[1,2,5]

Donor-derived T-cell source
differentiated into cytokine-induced killer (CIK) cells
Non-viral Sleeping Beauty (SB) transposon
CAR-T construct with CD28 and OX40 costimulatory domains









Topics - CAR-T in Pediatric ALL, Monza

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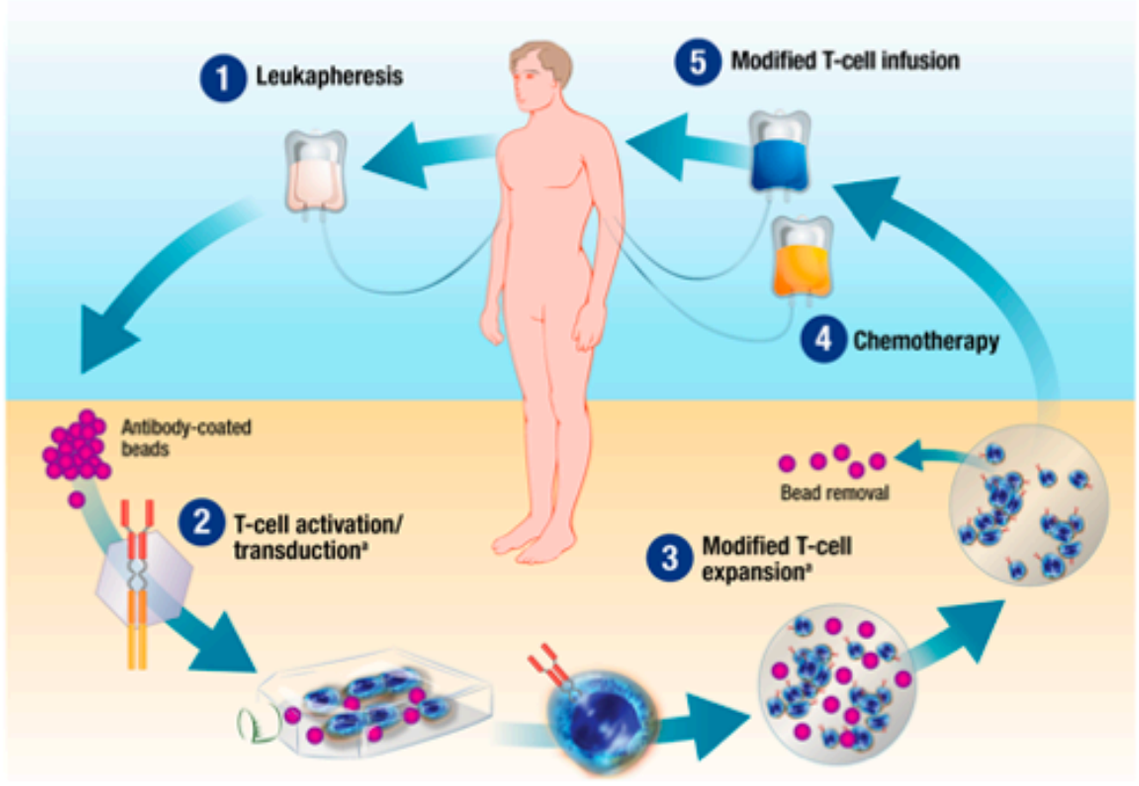
CAR: chimeric antigen receptor, r/r: relapsed/refractory; ALL: acute lymphoblastic leukemia; CRS: cytokine release syndrome

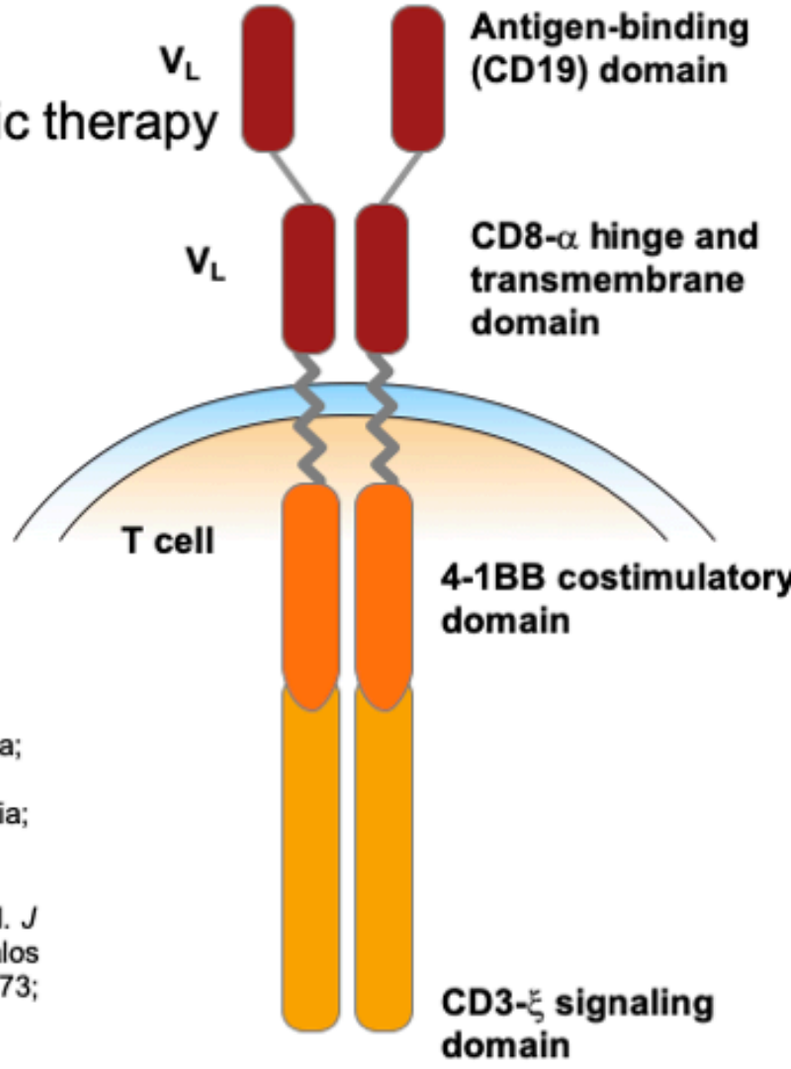
CAR T-Cell Therapy: Tisagenlecleucel (CTL019) - CAR-T in Pediatric ALL, Monza

- **First approved CAR T-cell therapy in the United States**
  - Aug 2017: for patients up to 25 years of age with r/r BCP ALL
  - May 2018: for adult patients with r/r DLBCL after ≥ 2 lines of systemic therapy
- Then approved in Europe and Canada



[a] Cellular reprogramming and ex vivo expansion are conducted at a cell processing facility.

tisagenlecleucel



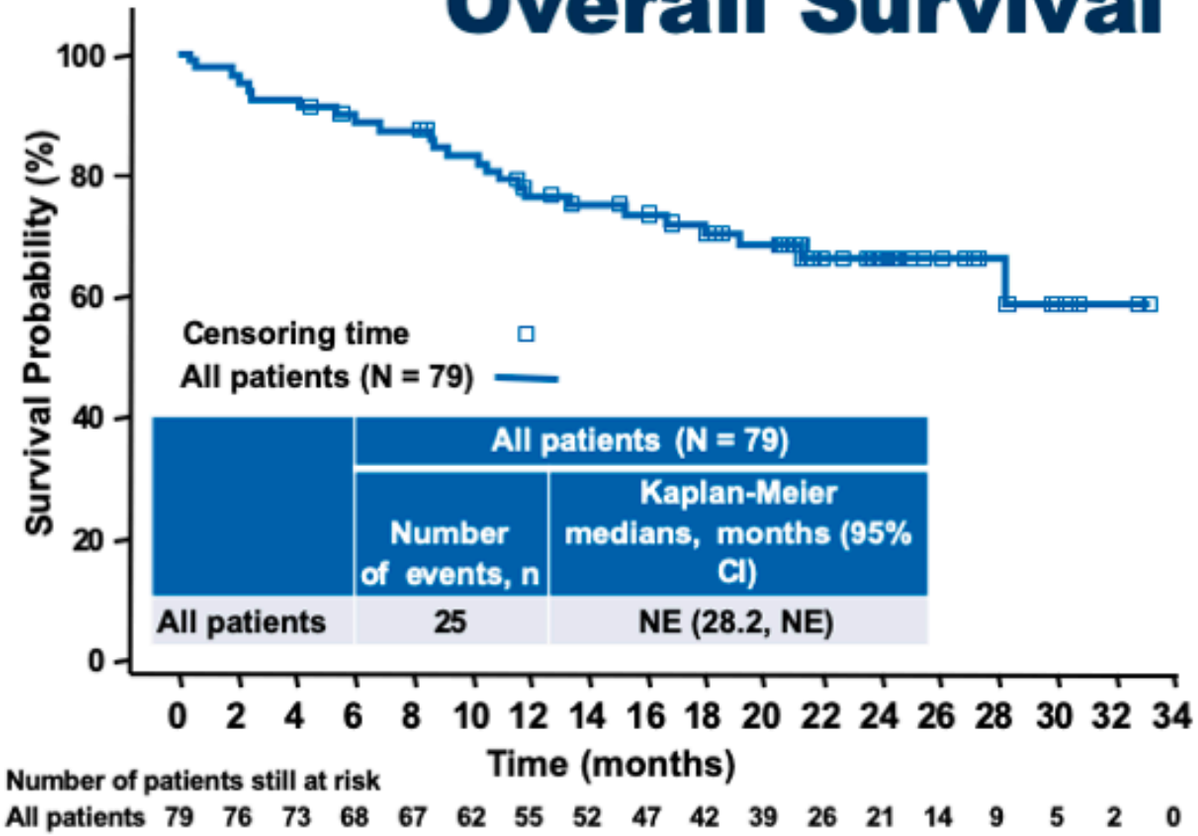
CAR, Chimeric Antigen Receptor; DLBCL, diffuse large B-cell lymphoma; EU, European Union; peds ALL, pediatric acute lymphoblastic leukemia; r/r, relapsed/refractory.

1. Milone MC, et al. *Mol Ther.* 2009;17:1453-1464; 2. Zhang H, et al. *J Immunol.* 2007;179:4910-4918; 3. Kalos M, et al. *Sci Transl Med.* 2011;3:95ra73; 4. Maude SL, et al. *N Engl J Med.* 2018;378(5):439-448.

Updated ELIANA (first global CAR T trial, 25 sites, 11 countries in N.A.+EU)

- **Inclusion:**
  - r/r B-cell ALL, aged 3-21 ys
  - BM ≥ 5% lymphoblasts
- **Exclusion:**
  - Isolated extra-med disease relapse
  - Prior CD19-directed or gene therapy

Overall Survival



	All patients (N = 79)	
	Number of events, n	Kaplan-Meier medians, months (95% CI)
All patients	25	NE (28.2, NE)

* 117 screened, 94 enrolled, 79 infused

Baseline Characteristics	N = 79*
Median age (range), years	11 (3-24)
Prior SCT, %	61
Previous lines of therapy, med (range), n	3 (1-8)
BM blasts, median (range), %	74 (5-99)

Eliana

- RFS rate among responders (N=65)
  - 12-month: 66% (95% CI, 52-77)
  - 24-month: 62% (95% CI, 47-75)
- OS among all infused patients
  - 12-month: 76% (95% CI, 65-85)
  - 24-month: 66% (95% CI, 54-76)

Note: All patients infused with tisagenlecleucel were included. Time is relative to infusion.
CR, complete remission; CRi, complete remission with incomplete blood count recovery; NE, not estimable.

ASH 2018

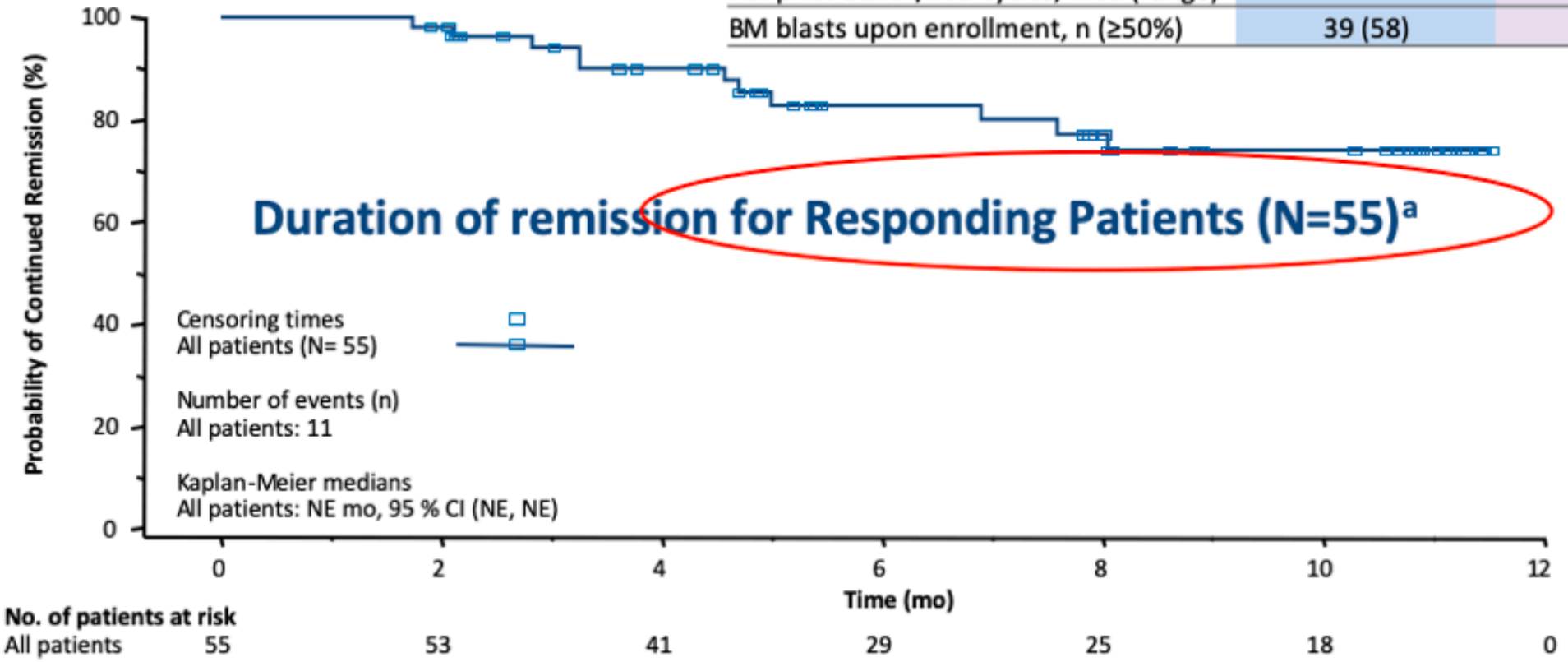
B2001X: vs Eliana: includes BLINA prior to apheresis a/o Ino as bridging

- **Inclusion:**
  - r/r B-cell ALL, aged 3 at screening, 21 ys at the time of initial diagnosis —> *emended to <26 ys at screening*
  - BM ≥ 5% lymphoblasts
  - previous blinatumomab w CD19+ expression + absence of CD19- blasts
- **Exclusion:**
  - Isolated extra-med disease relapse
  - Prior gene therapy

➡ 80 screened —> 73 enrolled —> 67 infused

Characteristics	All Patients, (N=67)	Prior BLINA, (n=15)	INO as BT, (n=9)
Median age (range), y	10 (2-33)	8 (2-24)	18 (5-24)
Prior alloSCT, n (%)	41 (61)	13 (87)	3 (33)
Prior lines of therapy, med (range), n	2 (1-8)	3 (2-8)	2 (1-4)
No. prior BLINA/INO cycles, med (range)		1 (1 to 7)	3 (1 to 6)
BM blasts upon enrollment, n (≥50%)	39 (58)	9 (60)	7 (78)

Duration of remission for Responding Patients (N=55)[a]



B2001X

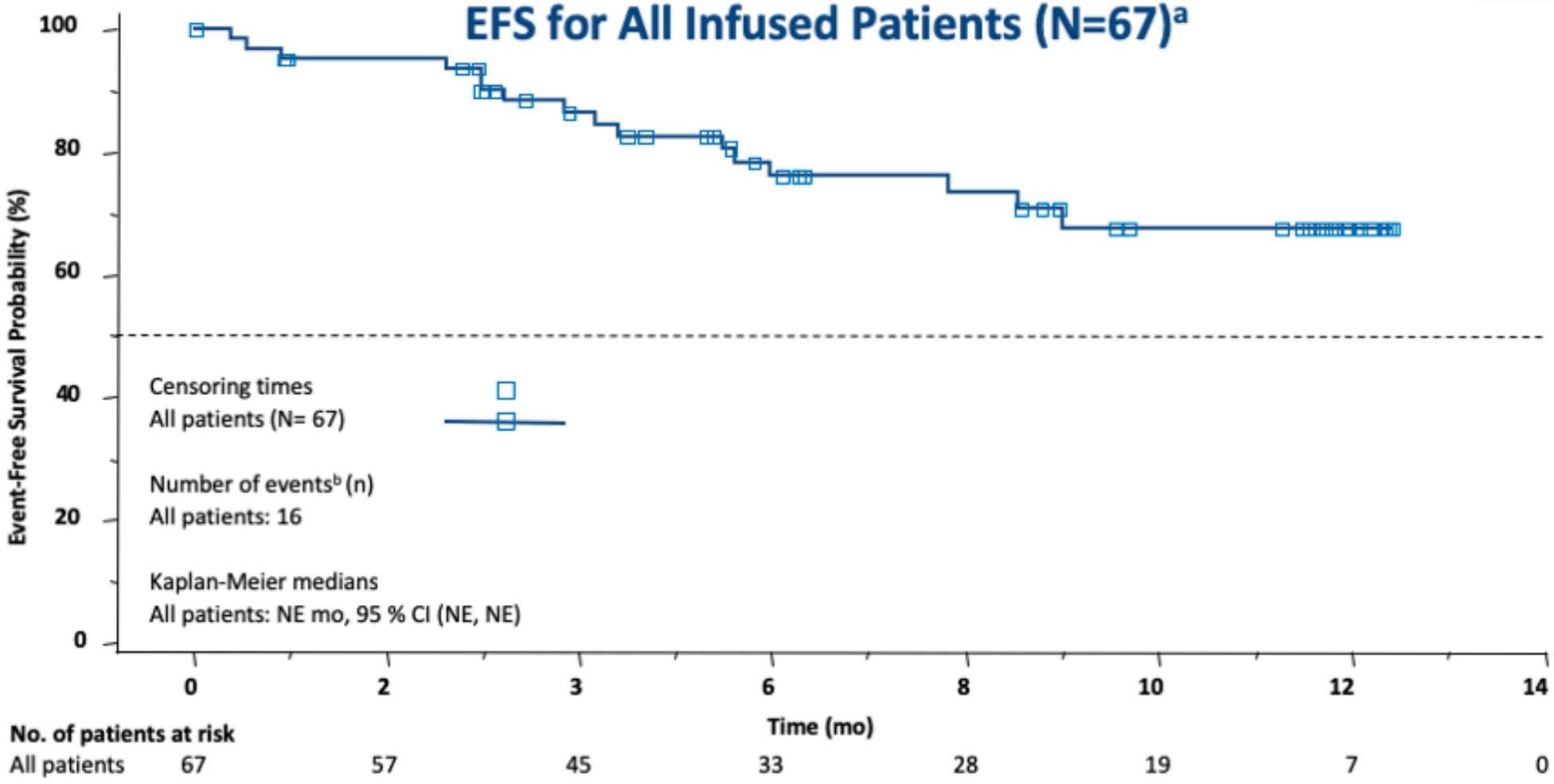
EHA - ASCO 2020

[a]With censoring for alloSCT. [b]An event is defined as the time from infusion to the earliest death from any cause, relapse, or treatment failure. alloSCT, allogenic stem cell transplant; CI, confidence interval; EFS, event-free survival; NE, not estimable.

B2001X: EFS - CAR-T in Pediatric ALL, Monza

B2001X

EFS for All Infused Patients (N=67)[a]



No. of patients at risk

All patients	67	57	45	33	28	19	7	0

[a]With censoring for alloSCT. [b]An event is defined as the time from infusion to the earliest death from any cause, relapse, or treatment failure.
alloSCT, allogenic stem cell transplant; CI, confidence interval; EFS, event-free survival; NE, not estimable.

EHA - ASCO 2020

B2001X: CRS & Adverse events - CAR-T in Pediatric ALL, Monza

B2001X

Selected AEs ≤8 ws, n (%)	All Grades	Grade 3/4
CRS[a]	43 (64)	19 (28)
Infections	26 (39)	11 (16)
Cytopenias >28 ds	32 (48)	26 (39)
Neurological events[b]	16 (24)	7 (10)

- Majority of adverse events occurred ≤8 weeks after tisagenlecleucel infusion with no cerebral edema reported
- 4 deaths reported within 30 days post infusion: ALL progression (n=2), CRS with progressive refractory ALL (n=1), and infection with multiorgan failure (n=1)

Cytokine Release Syndrome	All Infused Patients (N=67)
Time to onset, median (range), d	5 (1-13)
Duration of CRS, median (range), d	7 (1-27)
Time to grade 3/4 CRS, median (range), d	6 (1-11)
High-dose vasopressors, n (%)	15 (22)
Tocilizumab, n (%)	18 (27)
Corticosteroids, n (%)	4 (6)
ICU admission, n (%)	19 (28)
Intubation, n (%)	5 (7)
Dialysis, n (%)	2 (3)

[a]CRS was graded using the Penn scale (Porter DL, et al. *Sci Transl Med*. 2015;7[303]:303ra139). [b]Neurological events were graded by CTCAE v4.03.
AE, adverse event; ALL, acute lymphoblastic leukemia; CRS, cytokine release syndrome; CTCAE, Common Terminology Criteria for Adverse events.

EHA - ASCO 2020

B2001X: Efficacy - by subset - CAR-T in Pediatric ALL, Monza

B2001X	All Patients N=65 (OS, N=67)	Prior BLINA n=15	No Prior BLINA n=50	INO as BT n=9	No INO as BT n=56
ORR (CR+CRi),[a] n (%) (95% CI)	55[b] (85) (74-92)	10 (67) (38-88)	45 (90) (78-97)	6 (67) (30-93)	B2001X (76-95)
MRD (–) in CR/CRi,[c] %	96	100	95	100	95
DOR, relapse-free probability, % (95% CI)					
Month 6	83 (69-91)	88 (39-98)	82 (66-91)	67 (20-90)	86 (70-93)
Month 9	74 (57-85)	70 (23-92)	75(57-86)	67 (20-90)	76(58-87)
Relapse in pts with CR/CRi at any time, n	14[d]	2	12	4	10
CD19 status at relapse, n (–)/n (+)	9/5	2/0	7/5	1/3	8/2
12-month OS, % (95% CI)	83 (69-92)	53 (19-78)	91 (74-97)	71 (23-92)	85 (69-93)

- Efficacy and safety outcomes of tisagenlecleucel in B2001X consistent with ELIANA
- Trend toward a lower rate of severe CRS in B2001X vs ELIANA
- Trend toward suboptimal outcomes in patients with prior BLINA or INO as bridging therapy (caveat: cohort size, short follow-up, and potential confounding factors, eg, chemo-refractory disease requiring prior MoAbs)

[a]Patients who had ≥3 months of follow-up or discontinued earlier. [b]The remaining 10 patients (10/65): 3 patients with no CR/CRi, 5 early progression, 2 deaths precluding disease evaluation. [c]MRD measured ≤3 months after infusion. [d]13/14 relapses were medullary (isolated or combined with extramedullary) and 1 was extramedullary.

BLINA, blinatumomab; BT, bridging therapy; CD, cluster of differentiation; CI, confidence interval; CR, complete remission; CRi, complete remission with incomplete blood count recovery; DOR, duration of remission; INO, inotuzumab; MRD, minimal residual disease; ORR, overall remission rate; OS, overall survival.

EHA - ASCO 2020

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Enrollment issue - CAR-T in Pediatric ALL, Monza

CAR-T Monza

 # 30 BCP ALL patients referred to Monza for CAR-t CTL019 in the first 4 ys

the vast majority could have been referred earlier!

 # 26 screening

 # 3: not evaluated (cross-border)

#1 extramedullary only

#2 CD19 negative ALL

#2 medullary blasts <5%

#4 protocol temporarily closed 20 (IR 12-25)

 # 2 over-age for ped Dept.

1 lost to f-up

 # 17: eligibility criteria matched

enrolled in CARCIK

1 exitus before apheresis

1 exitus before infusion

1 waiting CD3 for apheresis

14 infused tisagenlecleucel

1 moved to US

Apheresis/Bridging chemotherapy related issues - CAR-T in Pediatric ALL, Monza

CAR-T Monza

Apheresis

- chemotherapy washout: 1 week post-ITT, VCR, 4 weeks post PEG-ASP, etc
  - timing balance: optimal CD3 count, clinical condition, ideally lowest disease burden
  - close collaboration between local referring physicians/apheresis team/anesthesiologist
- venous access: balance between CBC (CD3 & lymph), patient features (weigh, compliance, CVC)
  - in our experience: all but 2 patient needed an additional femoral access, 8/14 patients needed sedation to perform apheresis (4-hr sedation: challenging, > low-weight)
  - good in vitro expansion in 13/14 pts overall despite high disease burden

Bridging chemotherapy

- 3-5 weeks time required after apheresis, depending on manufacturing slot availability
- no need to achieve/maintain CR, but disease control required (issues with local physicians, families)
  - ideally lowest disease burden upon infusion to optimize outcome and decrease risk of CRS: leukemia level definition of a critical threshold?
- in our experience:
  - low-dose chemotherapy (St. Jude rotational maintenance: VCR+ster, VP+CY, ARA-C+VP, 6MP+MTX; no anthracyclines, some ASP), + ITT; no MoAbs: no Ino (no Blina!)

ALL: acute lymphoblastic leukemia; ARA_C: cytosine arabinoside; CBC: cell blood counts; CR: complete remission; ITT: intra-tecal therapy; MTX: methotrexate; 6MP: 6-mercapto-purine; VCR: vincristine; VP: etoposide;

Infusion planning/Lymphodepletion chemotherapy- CAR-T in Pediatric ALL, Monza

CAR-T Monza

- Pharmacy for lymphodepletion and CRS drugs
- Cell Therapy Lab for thawing
- ICU (alerted for infusion & CRS)
- Nurse staff (plan daily activities, avoid simultaneous graft infusion)
- Clinical trial unit for sample shipping / Courier for Kymriah PK samples
- Patient reassessment

- patient re-assessement: disease, CNS, infection, toxicity —> *delay if major toxicity or infection*
- prior to infusion (start at -9 ds) (6-14 ds prior to infusion) —> *recommended, regardless of CBC*
  - Fludarabine 30 mg/m2 IV daily for 4 doses
  - Cyclophosphamide 500 mg/m2 IV daily for 2 doses

- dose range tisagenlecleucel (single infusion)
  - 0.2 to 5.0 × 10^6 cells/kg for patients ≤ 50 kg
  - 0.1 to 2.5 × 10^8 cells for patients > 50 kg
- premedication: anti-histaminic, paracetamol,

CBC: cell blood count; CNS: central nervous system

Nota bibliografica

Synopsis of CRS grading according to the U-Penn, Lee and ASTCT classifications

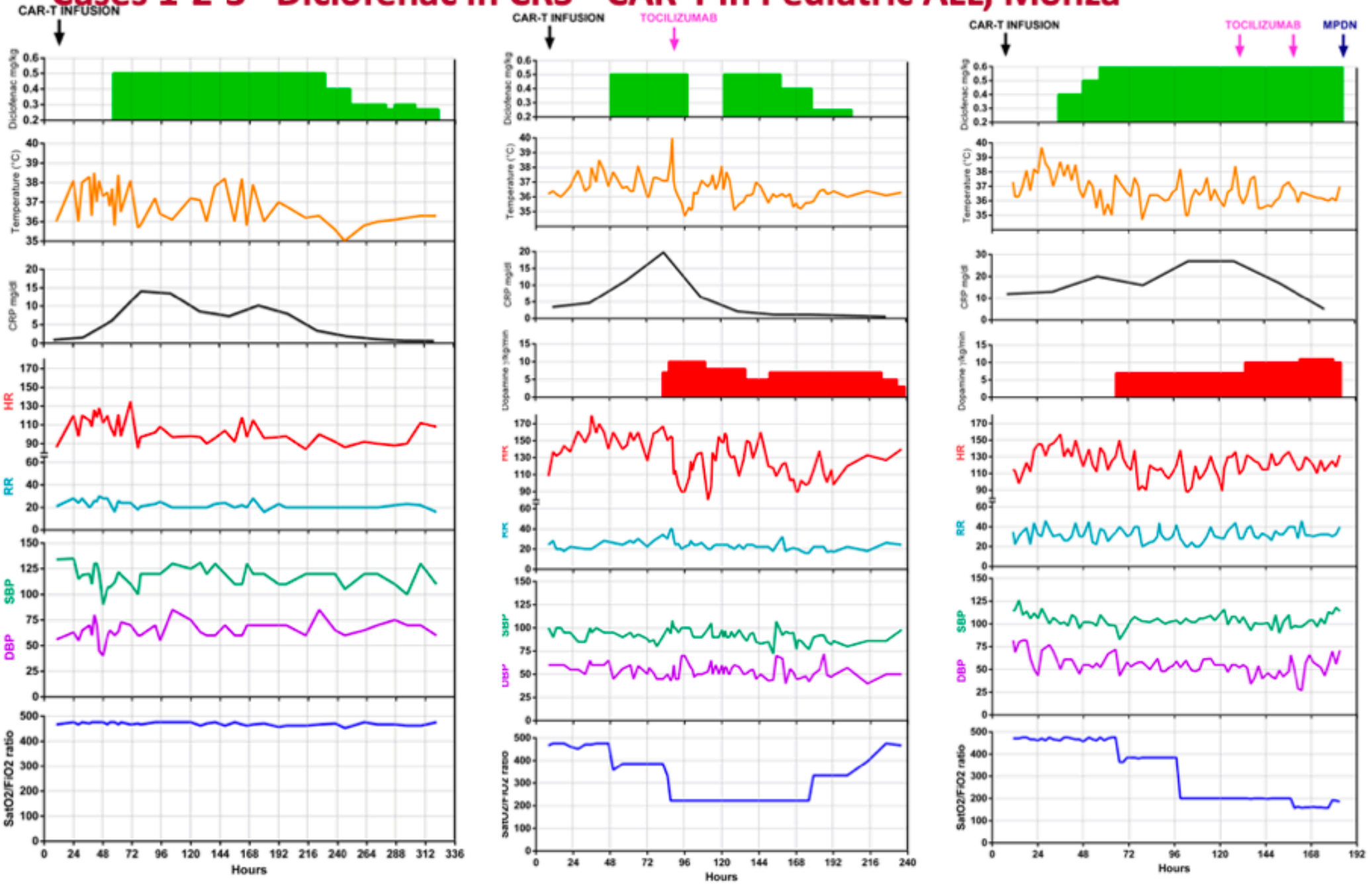
	Hypertension			Hypoxia			Organ Toxicity		
Grade	Penn[1]	Lee[2]	ASTCT[3]	Penn[1]	Lee[2]	ASTCT[3]	Penn[1]	Lee[2]	ASTCT[3]
1	-	-	-	-	-	-	-	-	NA
2	-	1 LD VP	No VP	–	O2 <40%	O2≤6L	I–II	II	NA
3	1 LD VP	2 or HD VP	1 VP	any O2	O2 ≥40%	O2 >6 L	III, trans IV	III, LFTs IV	NA
4	≥2 or HD VP	≥2 or HD VP	≥2VP	intub	intub	B/C-PAP/ intub	IV	IV	NA

CRS management algorithm

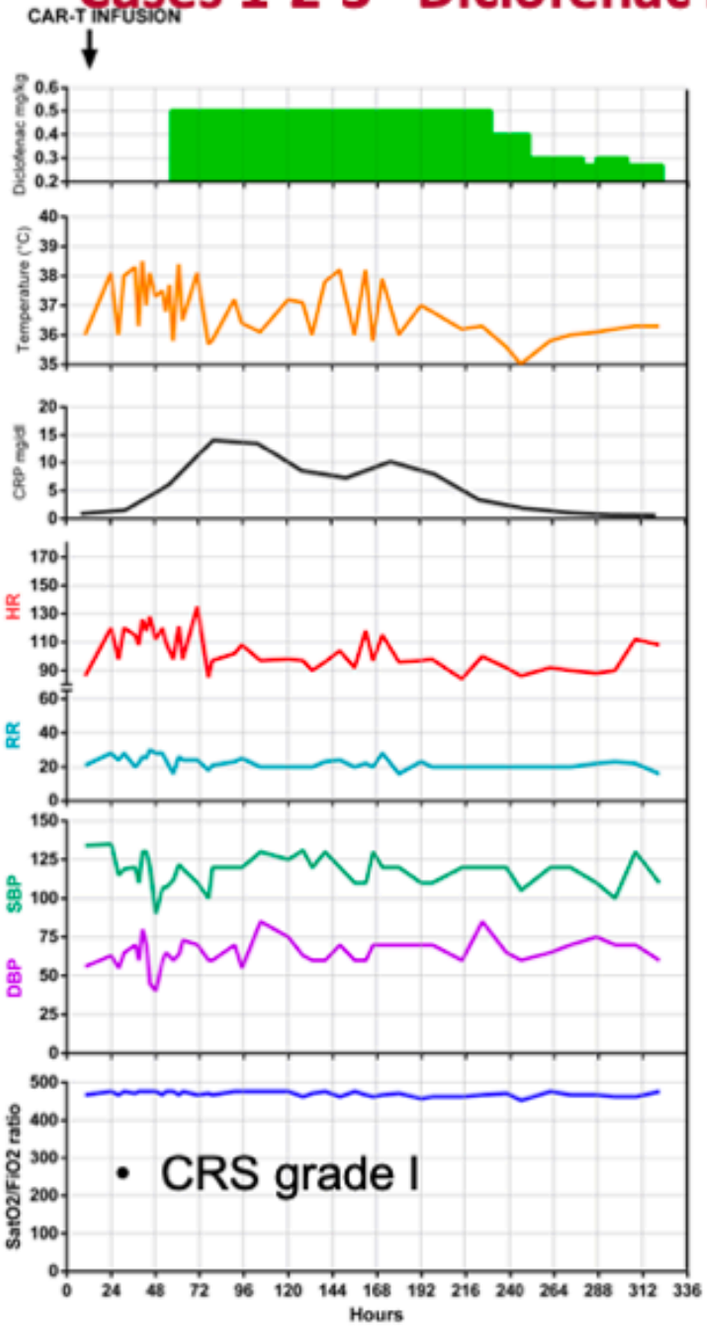
- ✦ vasopressors (single, low-dose, high-dose, double)
- ✦ tocilizumab
- ✦ steroid
- ✦ siltuximab

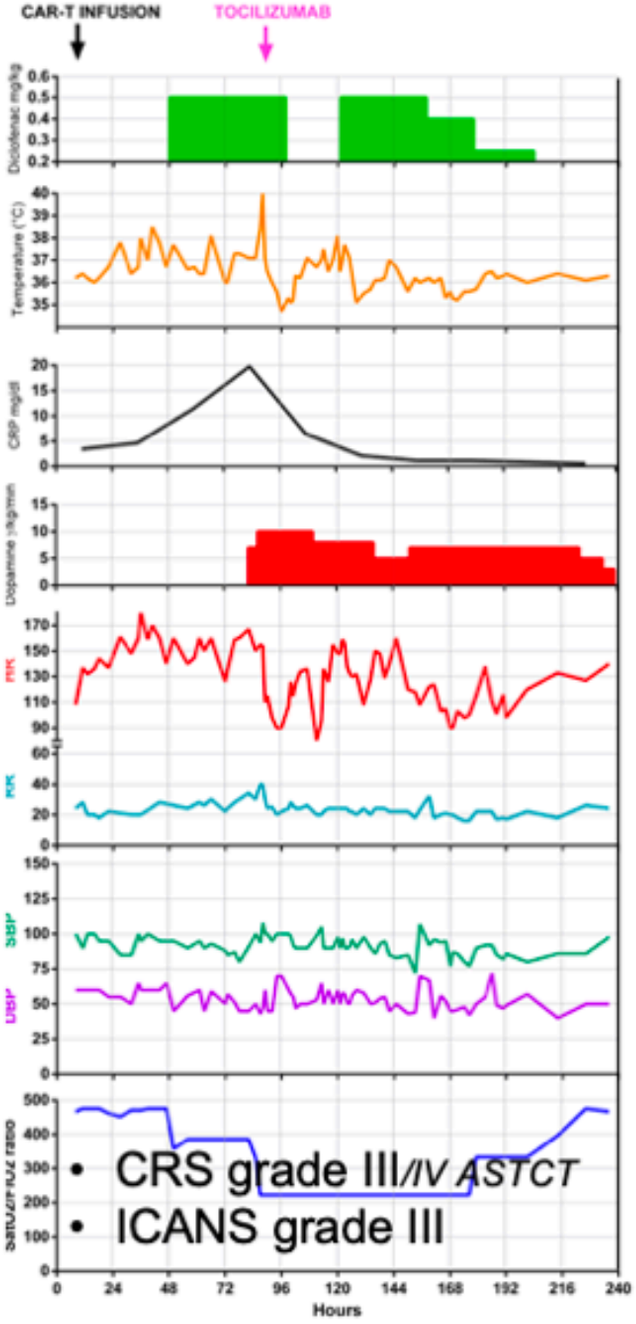
Nota bibliografica

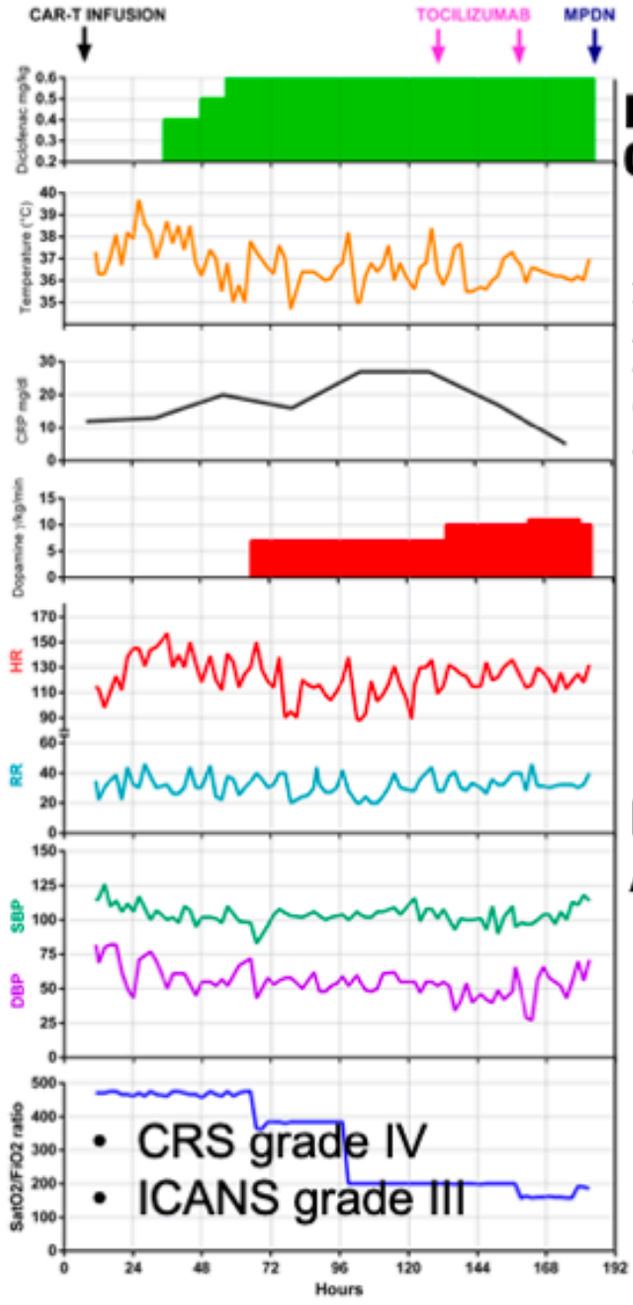
Cases 1-2-3 - Diclofenac in CRS - CAR-T in Pediatric ALL, Monza



Cases 1-2-3 - Diclofenac in CRS - CAR-T in Pediatric ALL, Monza



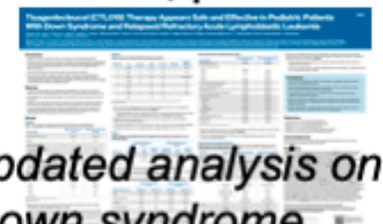




Diclofenac i.v. c.i. 0.3mg/Kg die

mean 3,07 febrile peaks over 24 hs w/o diclofenac and 0,95 w diclofenac

Experience on 7 pts ASH 2017, poster

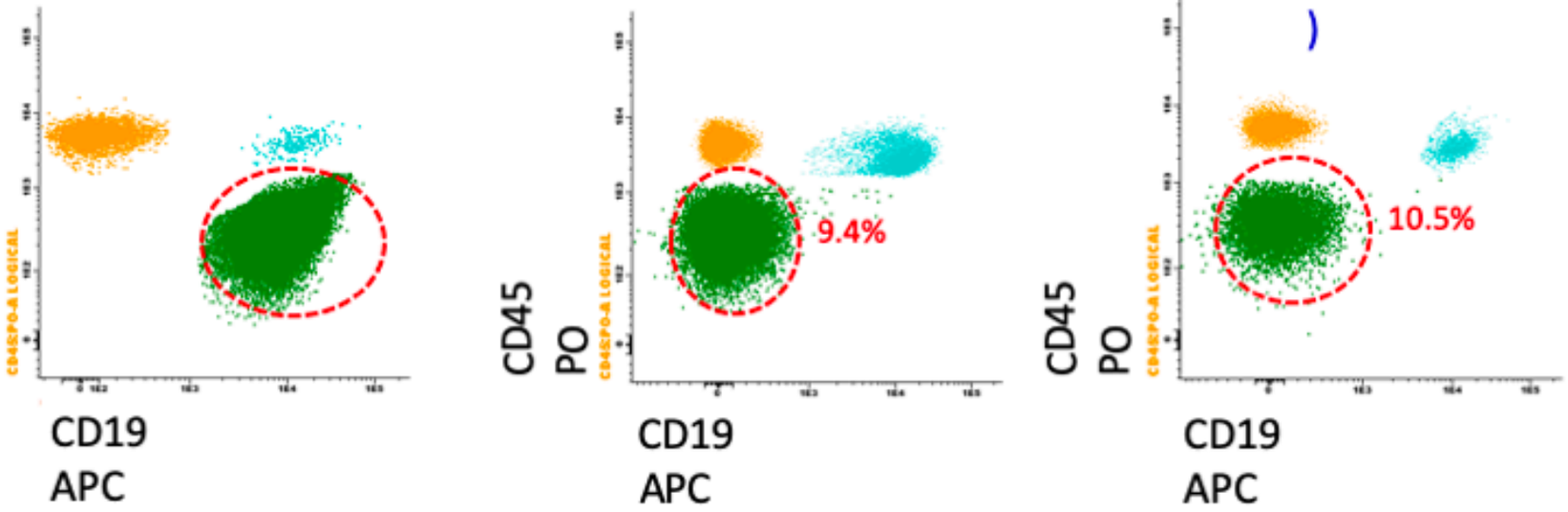


updated analysis on Down syndrome patients ongoing more fragile, similar efficacy

B-cell aplasia and Disease monitoring - CAR-T in Pediatric ALL, Monza

MRD with CD19 down-modulation

Addition of anti-CD22 is crucial



B-Lymphos T-Lymphos Blasts

Topics - CAR-T in Pediatric ALL, Monza

Topics

- CAR-t overall
  - London: CARPALL
  - Barcelona: ARI-0001 CART-BE-01 Trial
  - Monza: FT01-CARCIK
- *Tisagenlecleucel*
  - efficacy & toxicity
    - ELIANA
    - B2001X
  - practicalities
    - enrollment
    - apheresis
    - bridge chemotherapy
    - infusion
    - CRS
    - neurotoxicity
    - flow cytometry monitoring
- Open questions & novel approaches
- Conclusions

CAR: chimeric antigen receptor, r/r: relapsed/refractory; ALL: acute lymphoblastic leukemia; CRS: cytokine release syndrome

Open issues - CAR-T in Pediatric ALL, Monza

- eligibility:
  - isolated extramedullary leukemia
  - MRD+ post-HSCT
- efficacy:
  - MLL rearranged patients: increased risk of CD19- relapse
  - tissue trafficking
  - CNS leukemia
- timing?
  - patients eligible for HSCT
  - HR relapses?
  - frontline? - Cassiopeia
    - Could CAR-t substitute HSCT?
    - Could HSCT be placed as a rescue whenever CAR-t had failed?
- safety:
  - toxicity:
    - CRS management
    - neurotoxicity
- monitoring:
  - disease monitoring
    - CD19- relapse
  - BCA monitoring: impact of loss by timing
- what if failure?
  - second infusion
    - association with anti-PD1/PDL1
    - bridge to SCT vs standing alone

Could immunotherapy spare toxicity and maintain/increase efficacy?
What makes the difference... quality of life in the show and long term...