CAR T cells against B cell malignancies

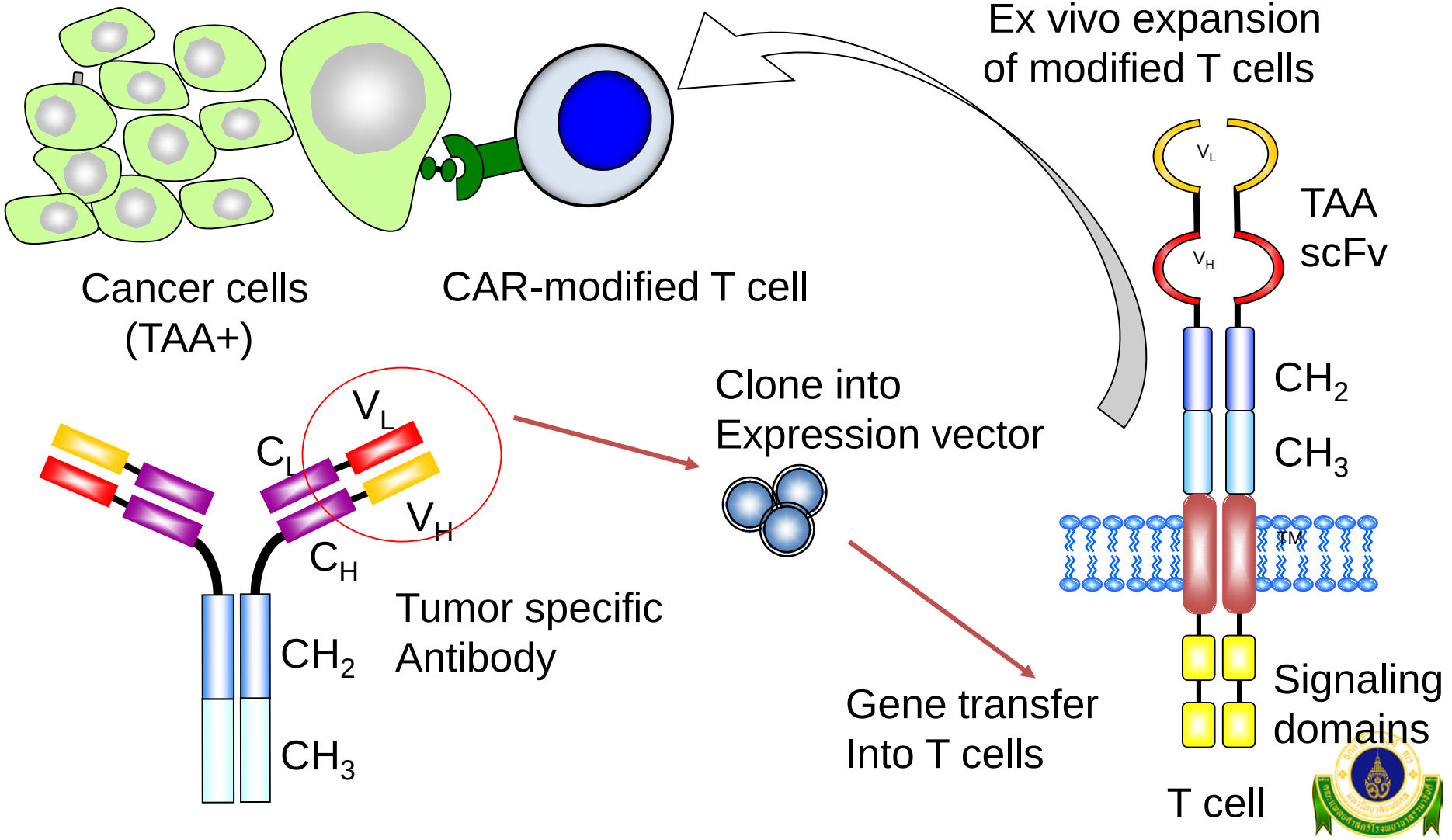
Clinical trials

Usanarat Anurathapan, MD

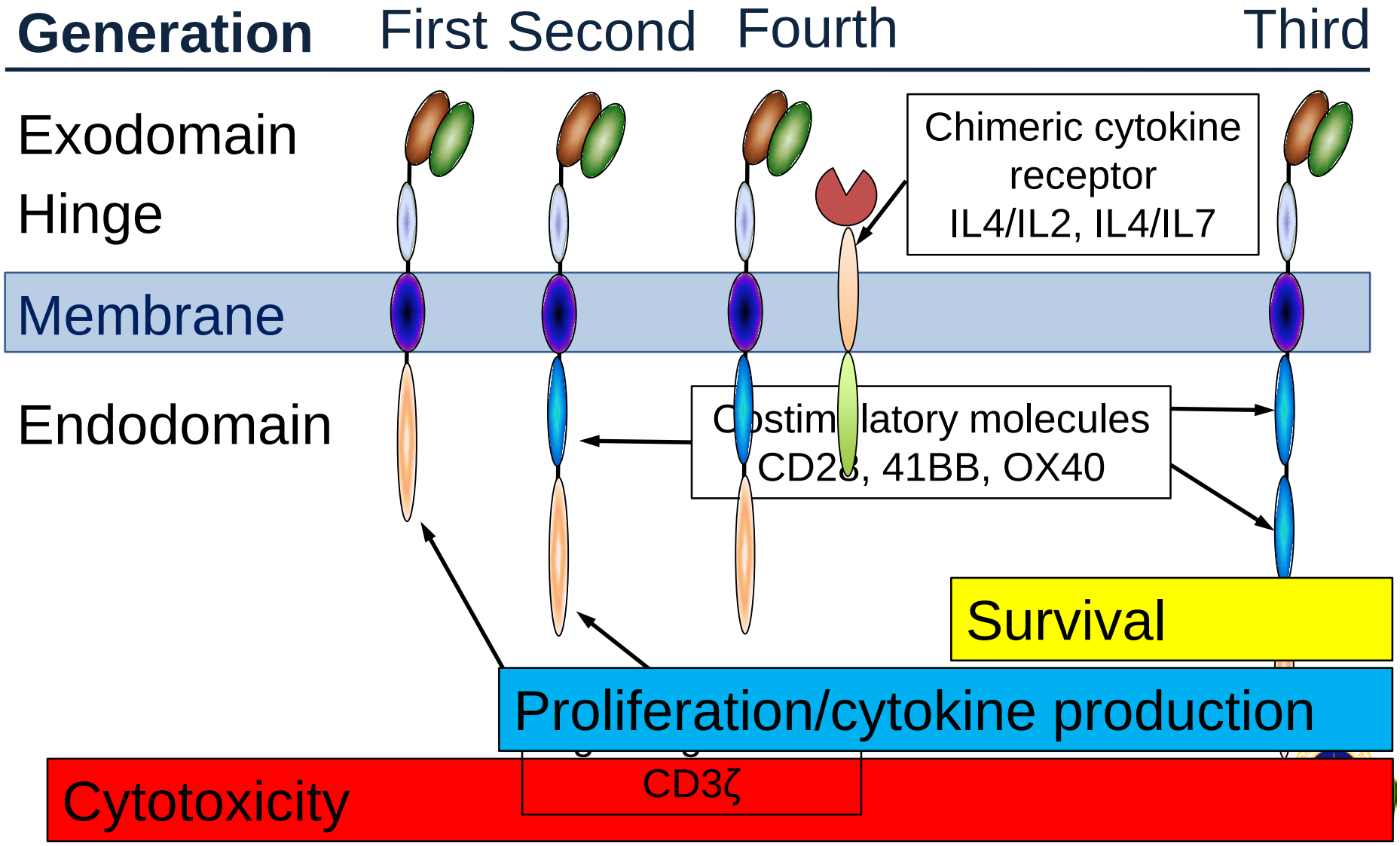
Department of Pediatrics

Faculty of Medicine Ramathibodi Hospital

Genetically modified T cells redirect the immune response against cancer cells



Evolution of CAR

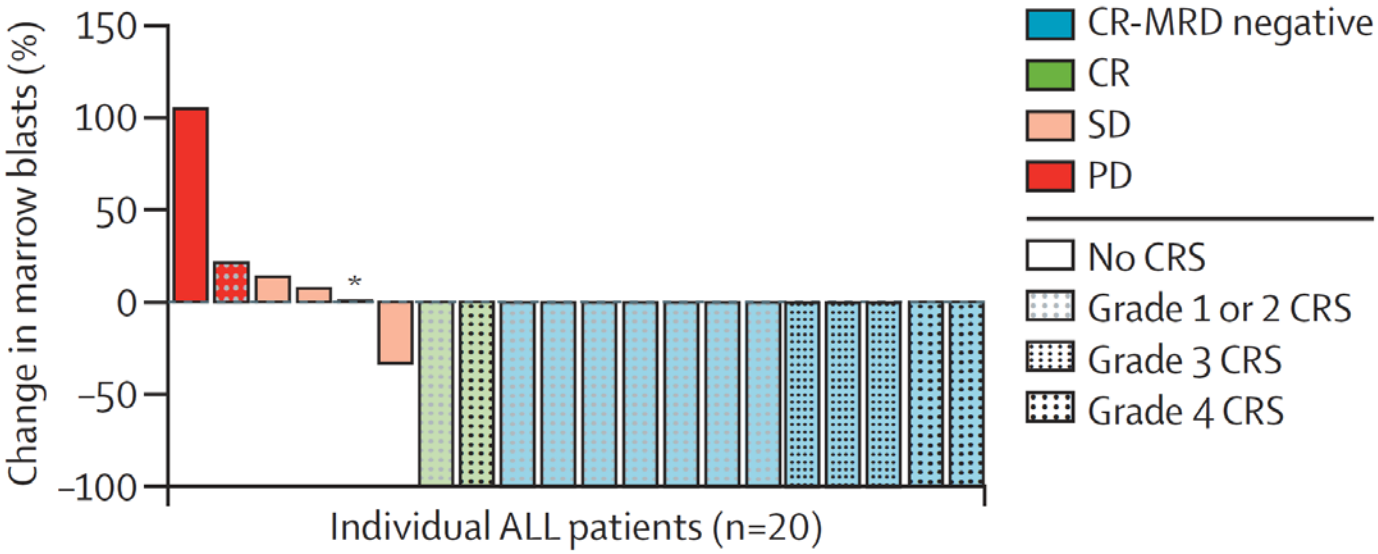


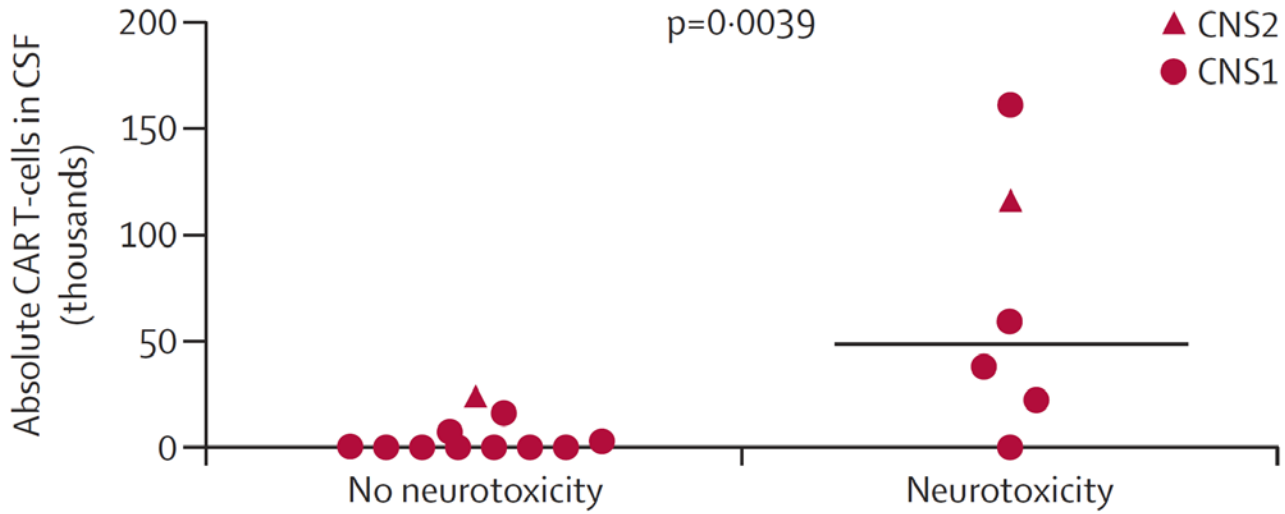
21 children & young adults in phase I trial

- All were relapsed or refractory CD19+ ALL or NHL; 8 pts undergone allo-HCT; 67% male; age 3-27y/o
- Median Leukemia burden = 26% marrow blast
- 19 pts received auto anti-CD19-CD28-CD3zeta CAR T cells (2 insufficient cells)
  - 3 + 3 design: 1 -> 3 x10^6 of CAR-T cells/kg
  - Median CAR transduction efficiency = 66%
- Prime with FLU/CY

Lee DW, et al. Lancet. 2015;385:517.

Factors associated with CRS & neurotoxicity





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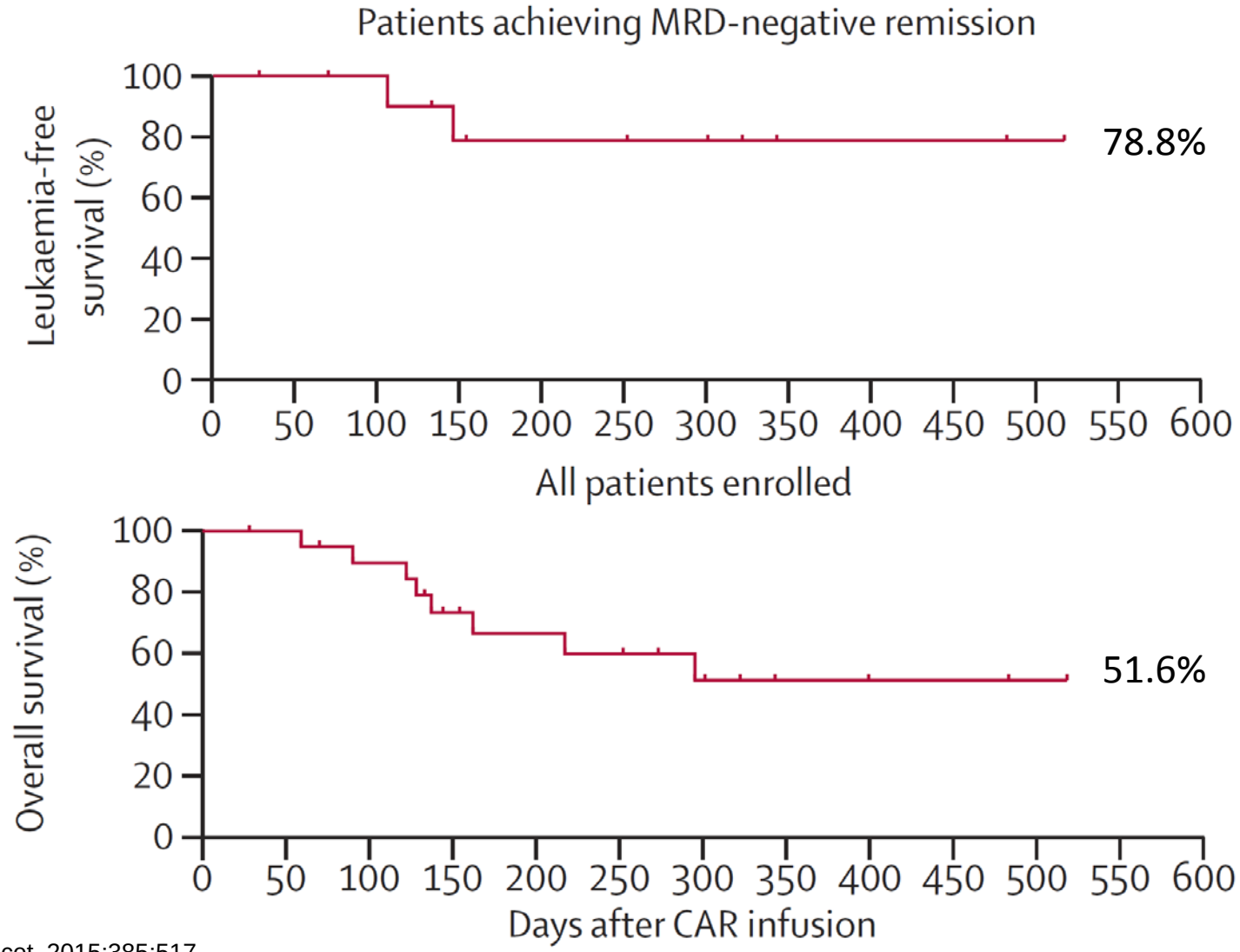
OS & LFS at median F/U 10 mo



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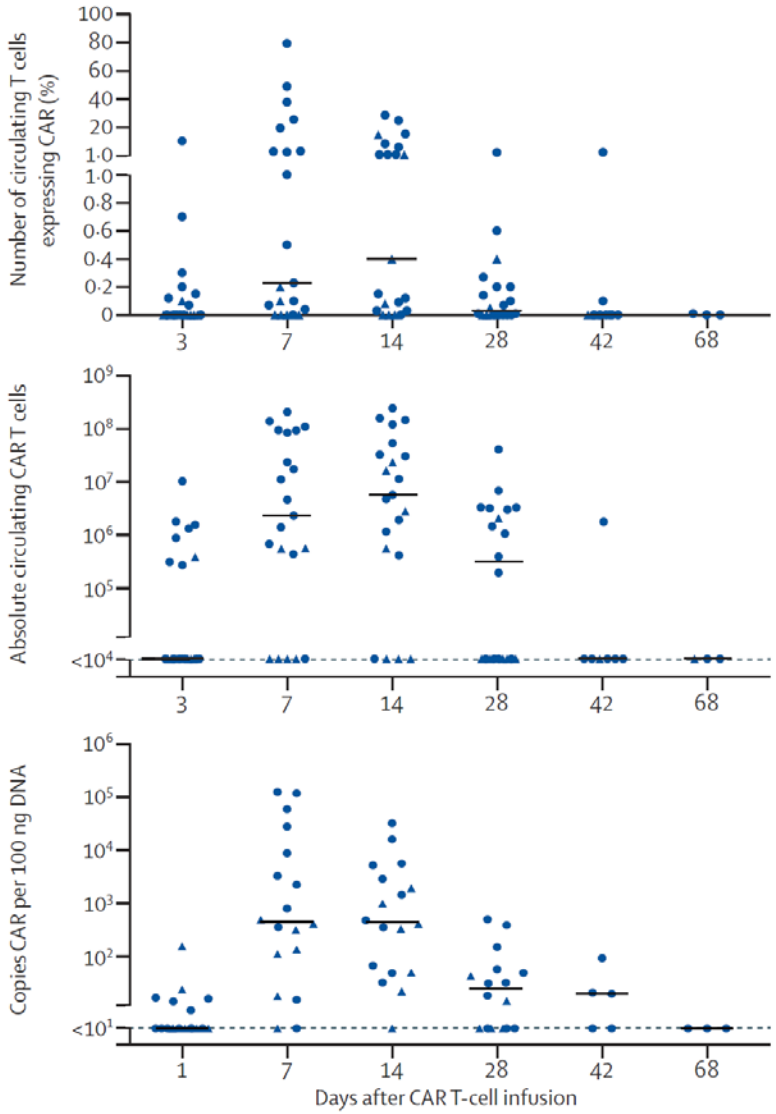
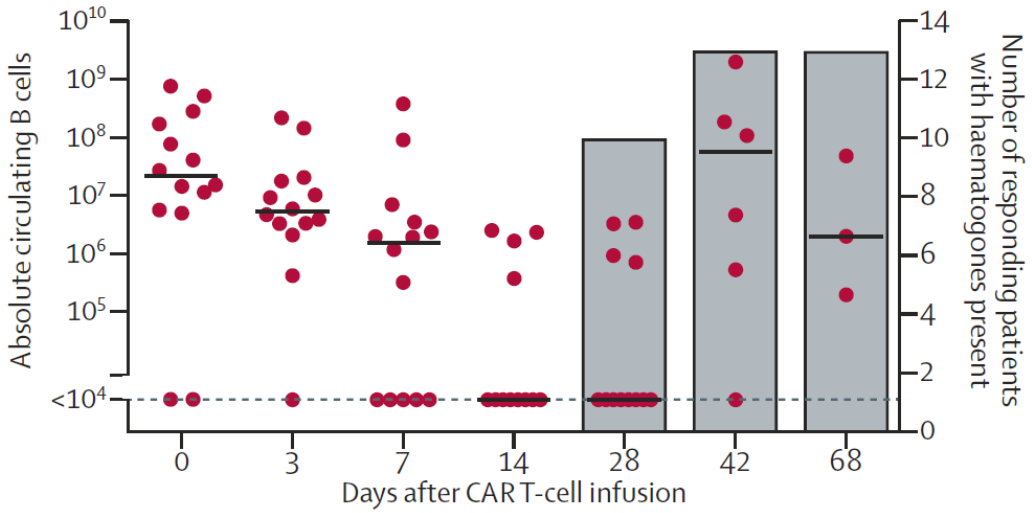
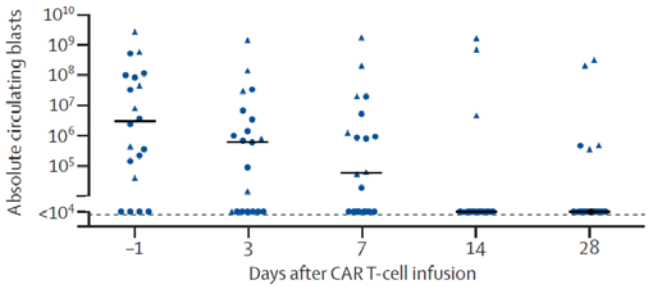
CAR T Persistence & B cell aplasia

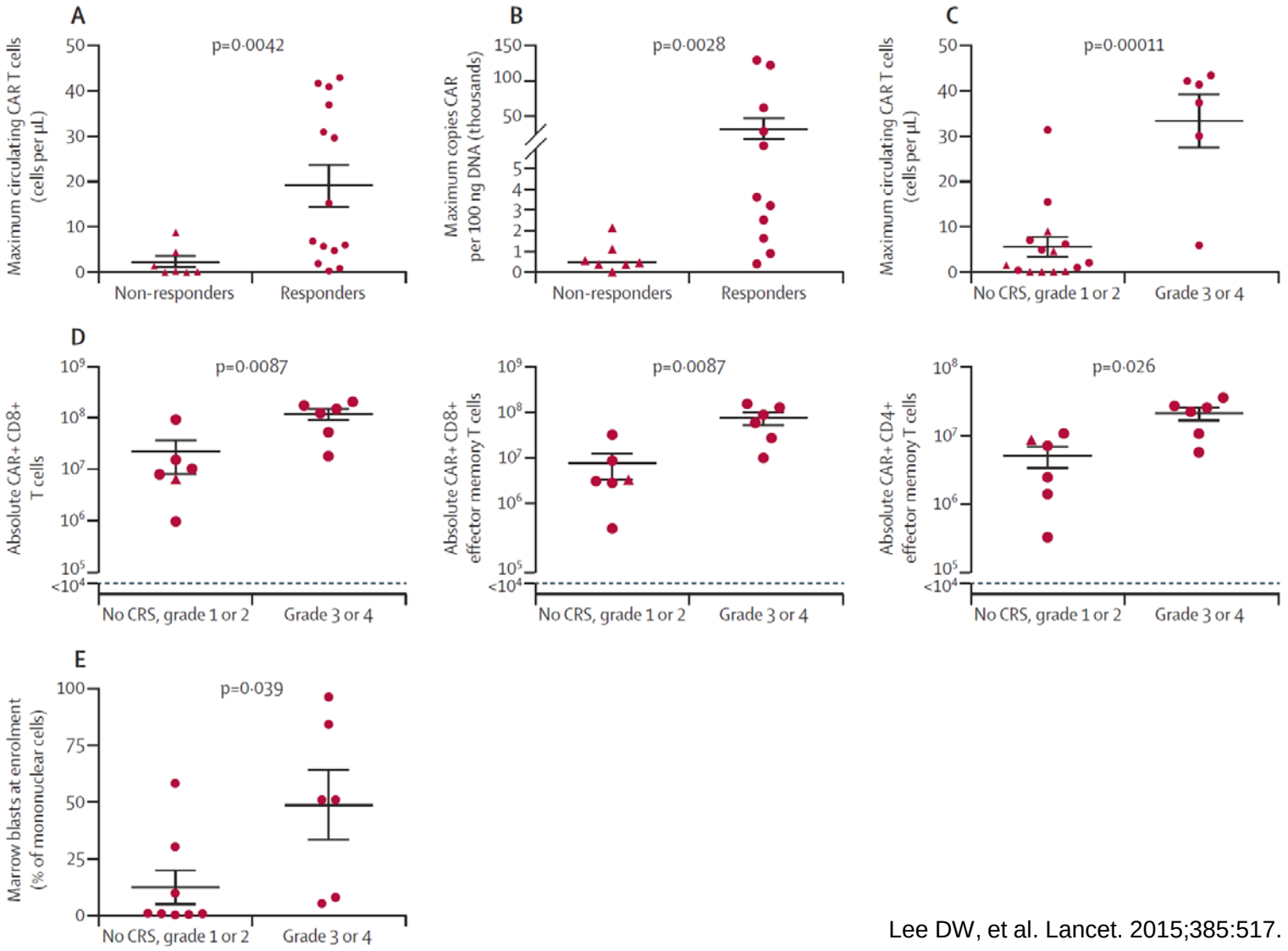






Lee DW, et al. Lancet. 2015;385:517.

CAR T persistence vs CRS biomarkers



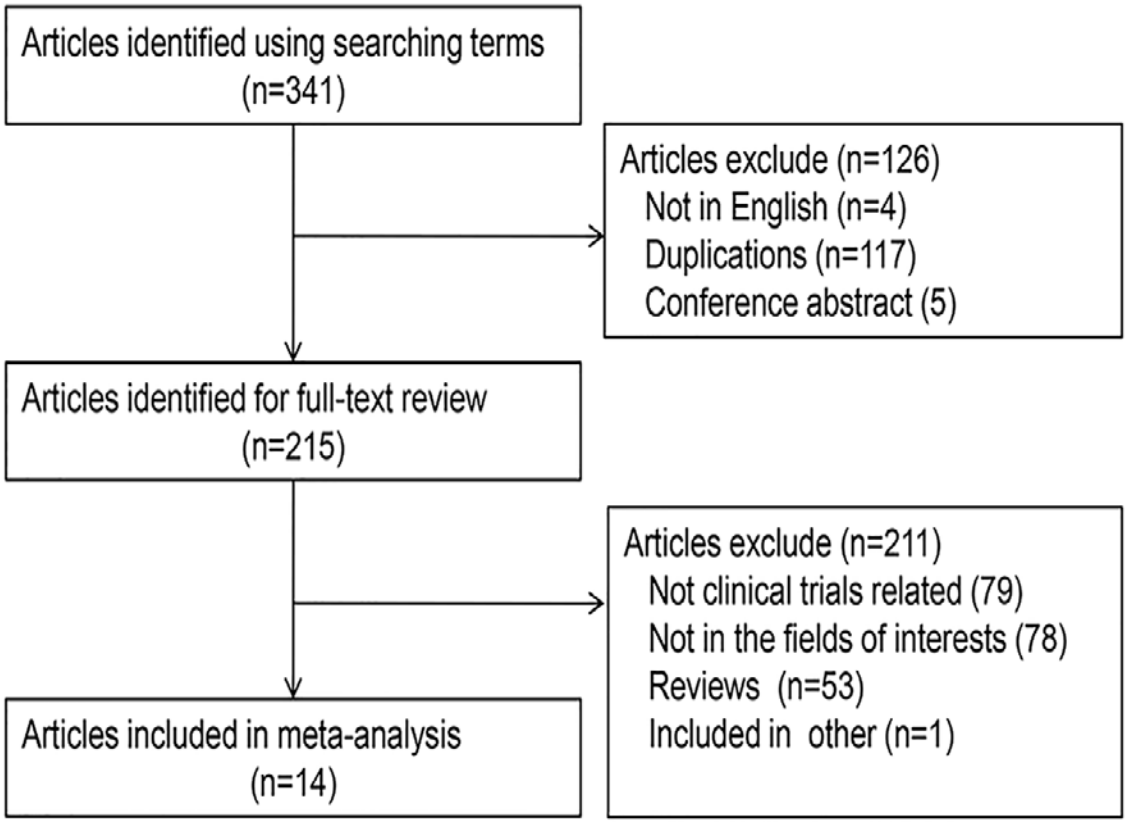
Lee DW, et al. Lancet. 2015;385:517.

Conclusion

- The maximum tolerated dose was 1 x 10^6 CD19-CAR T cells/kg
- All toxicities were fully reversible
  - CRS gr 4 = 14%
  - No prolonged B-cell aplasia

Lee DW, et al. Lancet. 2015;385:517.

Phase I clinical trials: meta-analysis



- 133 pts with B-cell malignancies
- 119 pts with eligible for response rate evaluation

Zhang T, et al. Oncotarget. 2015. 33961.

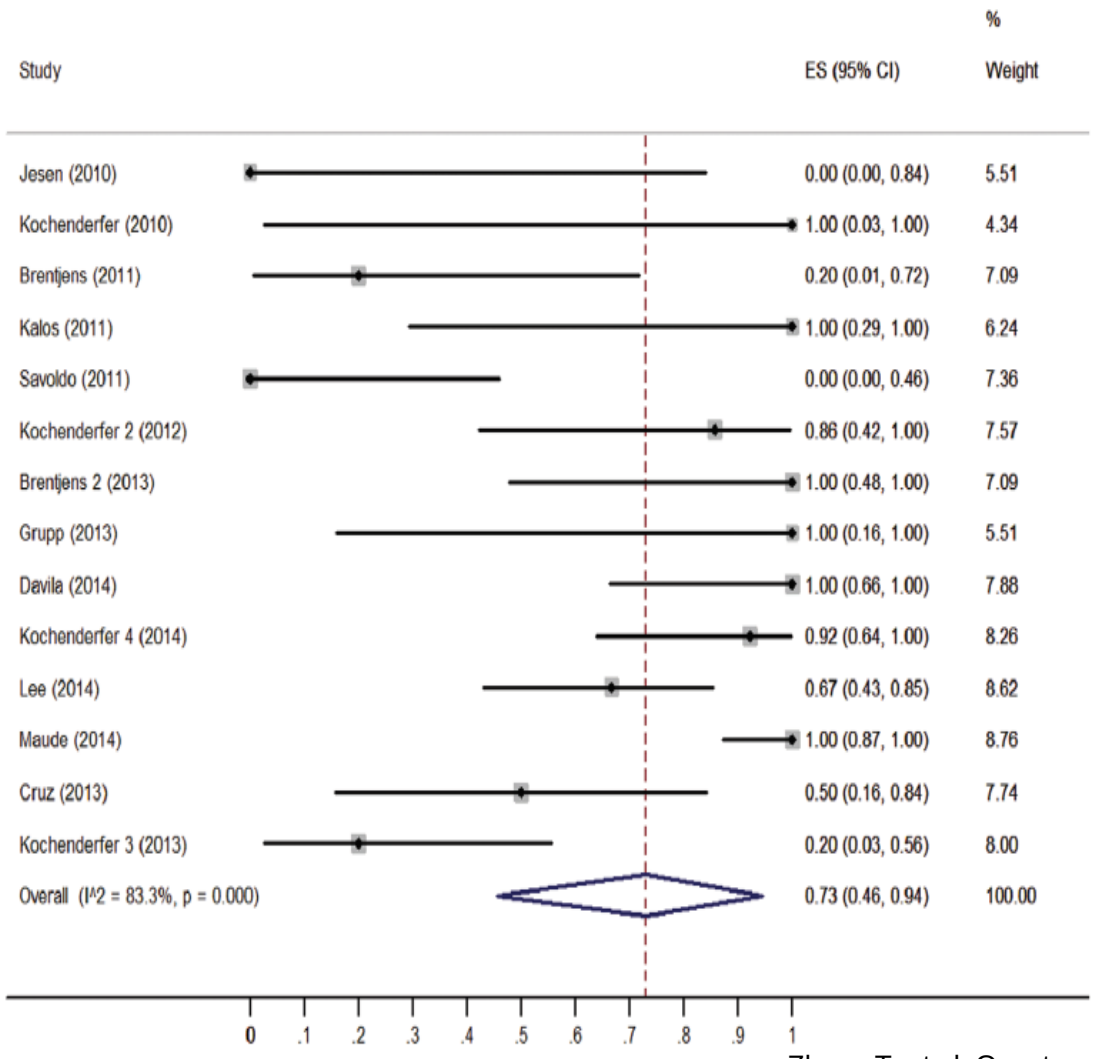
14 Clinical trials

No.[a]	Vector	T cell Original	Cell culture	Transfection method	T Cell treatment	CAR T cell persistence	Diagnosis	Lymphodepletion	Dose[b]	Response	Ref
2	IgG-CD4+ CD3	autologous	3 month	Electroporation	OKT3 + IL-2 +irradiated LCL feeders	1 day	lymphoma	Fludarabine IL-2	10^8-10^9/m^2	2PD	9
1	CD28+ CD28 and CD3	autologous	18-26 day	Lentivirus	OKT3 + IL-2 +irradiated LCL feeders	36 weeks	lymphoma	Cyclophosphamide Fudarabine IL-2	1-3 ×10^8	PR	12
9	CD28 + CD28+ CD3	autologous	6-18 day	Gammaretrovirus	OKT3 + IL-2	5-6 weeks	8CLL 1ALL	4 NA, Cyclophosphamide	0.4-3.0 ×10^7/kg	3NE[c], 1Died, 1CR, 1PD, 3SD	10
3	CD8-CD8 + 4-1BB + CD3	autologous	10 day	Lentivirus	CD3/28 beads	6 month	CLL	Pentostatin cyclophosphamide	1.46 × 10^5/kg 1.0-1.6 × 10^7/kg	2CR, 1PR	6
6	IgG-CD28 + CD3/CD28-CD3	autologous	6-8 day	Gammaretrovirus	OKT3 + IL-2	6 weeks	lymphoma	NA	2-10 ×10^7/m^2	6PR	13
8	CD28 + CD28+ CD3	autologous	24 day	Gammaretrovirus	OKT3 + IL-2	< 20 day or 8 weeks	4lymphoma 4CLL	Cyclophosphamide, Fudarabine, IL-2	0.3-2.8×10^7/kg	1Died, 5PR, 1CR, 1SD	11
5	CD28 + CD28+ CD3	autologous	14 day	Gammaretrovirus	CD3/28 beads	3-8 weeks	ALL	Cyclophosphamide	1.4-3.2 ×10^8/kg	5CR	15
8	IgG-CD28 + CD28+ CD3	allogeneic	5-6 week	Gammaretrovirus	Ad. Pp65+ EBV-LCLs+ IL-2	1-12 weeks	ALL	NA	1.5-12× 10^7/m2	1CR, 1PR, 2CCR, 1SD, 3PD	3
2	CD8-CD8+ 4-1BB+ CD3	autologous	10 day	Lentivirus	CD3/28 beads	6 month	ALL	NA or Cyclophosphamide	0.14-1.2 × 10^7/kg	2CR	4
10	CD28 + CD28+ CD3	allogeneic	8 day	Gammaretrovirus	OKT3 + IL-2	1 month	4 CLL 6 lymphoma	NA	1 × 10^6 /kg	6SD, 2PD, 1CR, 1PR	2
11	lgG+ CD 28	autologous	14 day	Gammaretrovirus	CD3/28 beads	2-3 month	ALL	Cyclophosphamide	3 × 10^6/kg	9CR, 2NE	16
15	CD28+ TCR	autologous	10 day	Gammaretrovirus	OKT3 + IL-2	<75 day	4 CLL, 11 lymphoma	Cyclophosphamide Fudarabine	5 × 10^6/kg	4PR, 8CR, 1SD, 2NE	7
21	lgG+ CD3+CD28	autologous	11 day	retroviruses	CD3/28 beads	68 day	20 ALL, 1 lymphoma	Fudarabine Cyclophosphamide	1-3 × 10^6/kg	3SD, 4PD, 13CR, CiR	8
30	CD8-CD8 + 4-1BB + CD3	autologous	10 day	Lentivirus	CD3/28 beads	up to 11 month	ALL	Fudarabine Cyclophosphamide	0.76-20 x 10^6/kg	27CR, 3NE	14

[a] No.= patients in clinical trial received CD19-CAR T cells; [b] Dose of infused T cells; [c]NE= no response and no evaluation

Zhang T, et al. Oncotarget. 2015. 33961.

Forest plot for overall response rates



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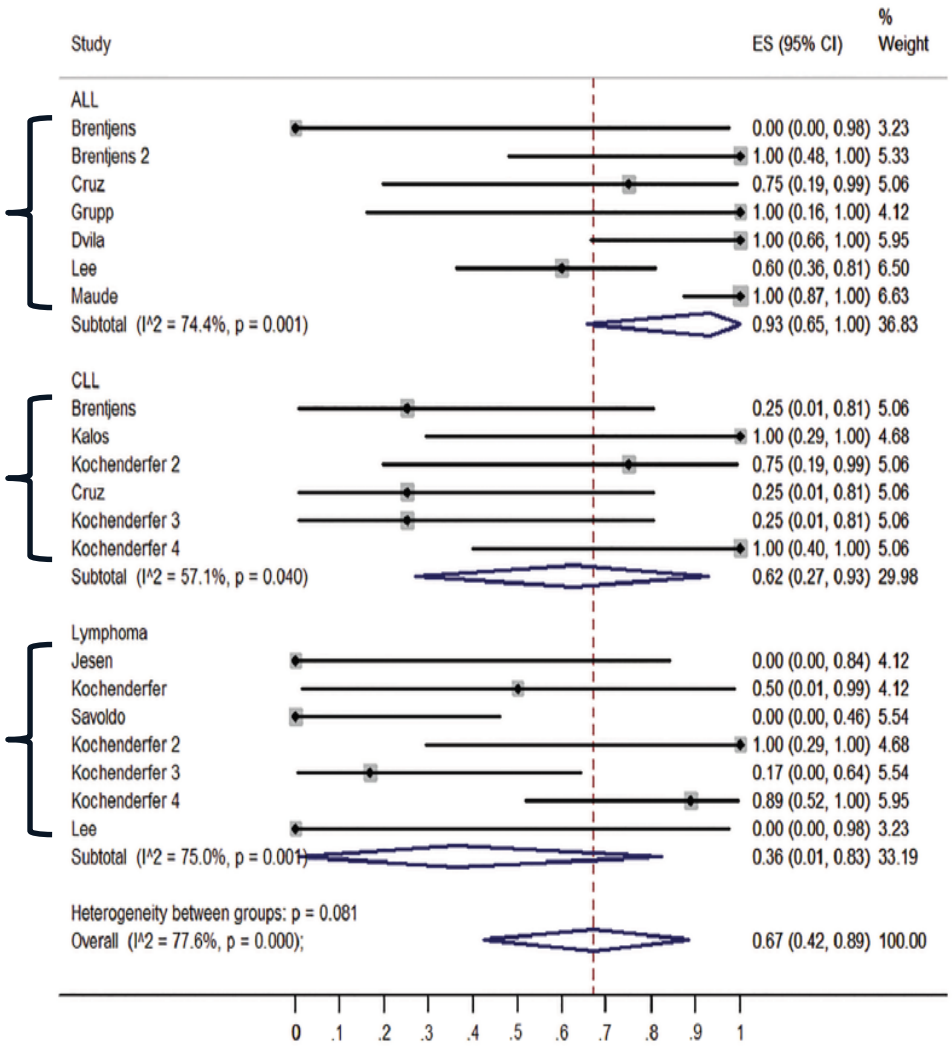
Better response rate in ALL: 93 (65-100)%

ALL

CLL

Lymphoma

Overall



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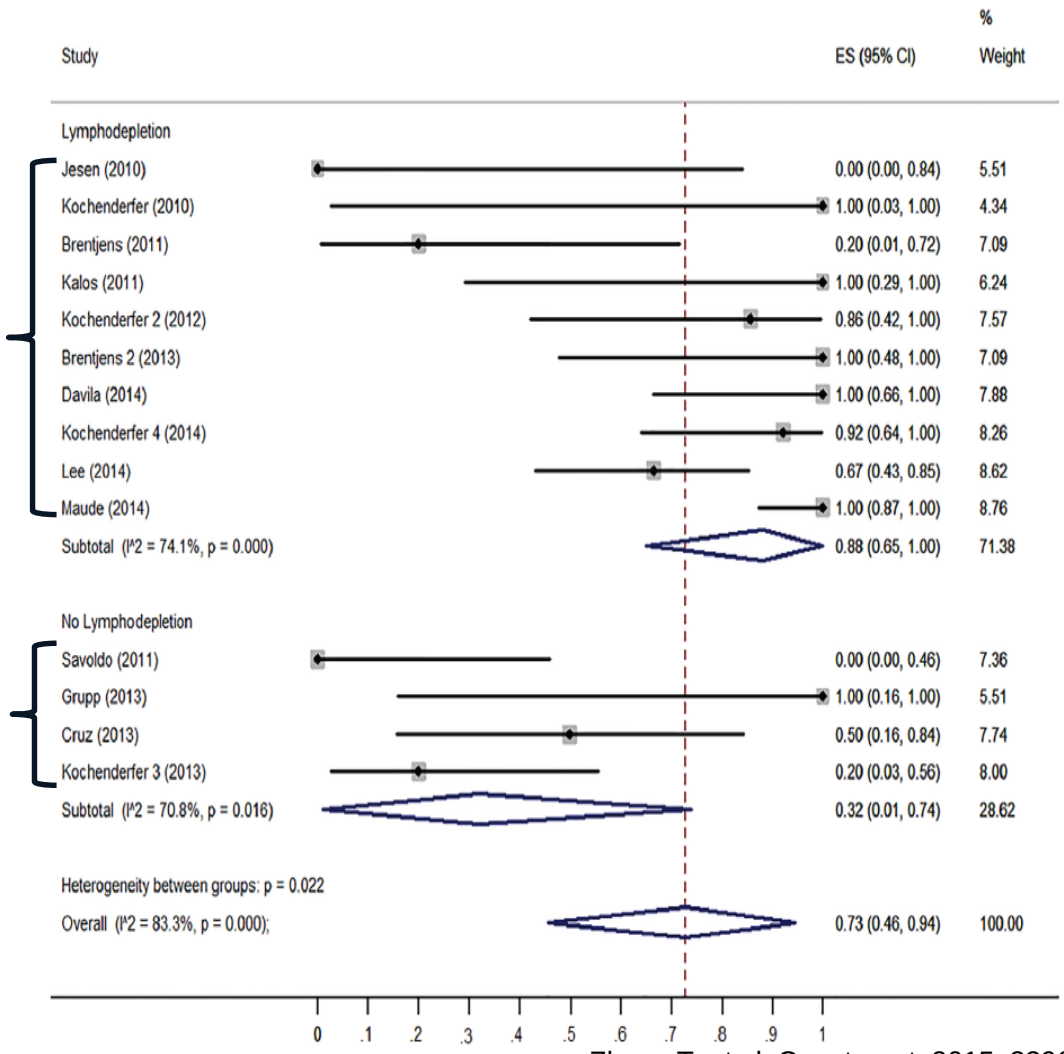
Better response rate with lymphodepletion: 88 (65-100)%

Lymphodepletion

No lymphodepletion



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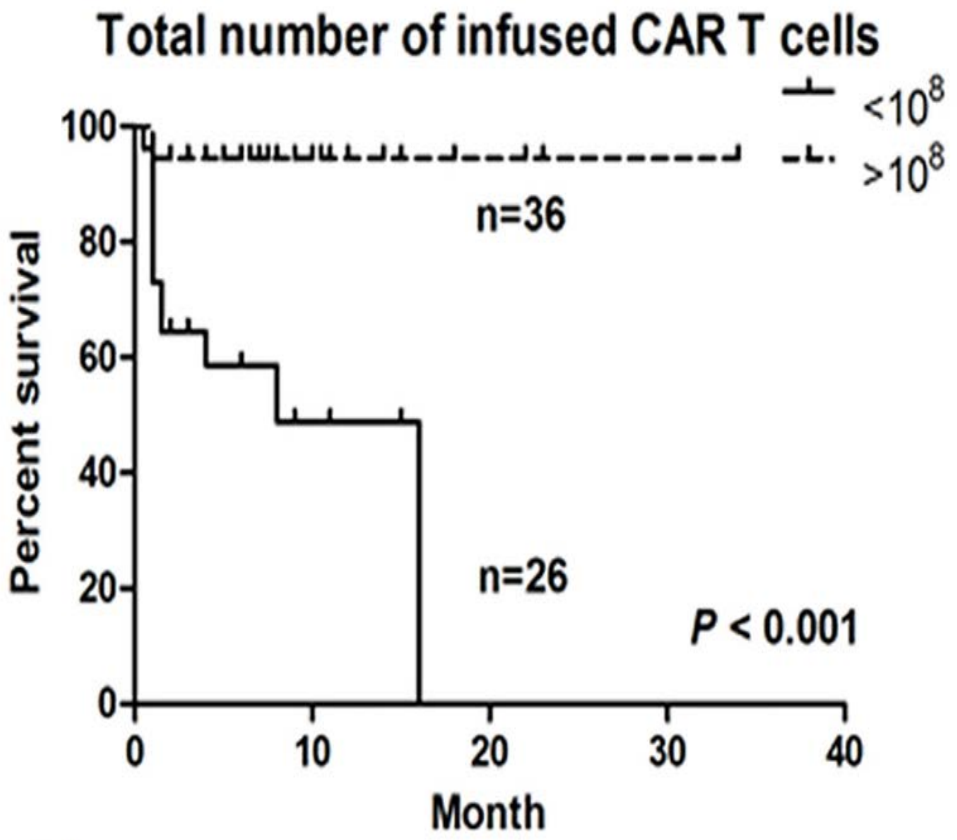
Better progression-free survival with >10^8 infused CAR T cells



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Conclusion

- High clinical response rate of CAR-CD19 T cell-based immunotherapy
- Lymphodepletion & increasing no. of infused CAR-CD19 T cell have positive correlation with the clinical efficacy

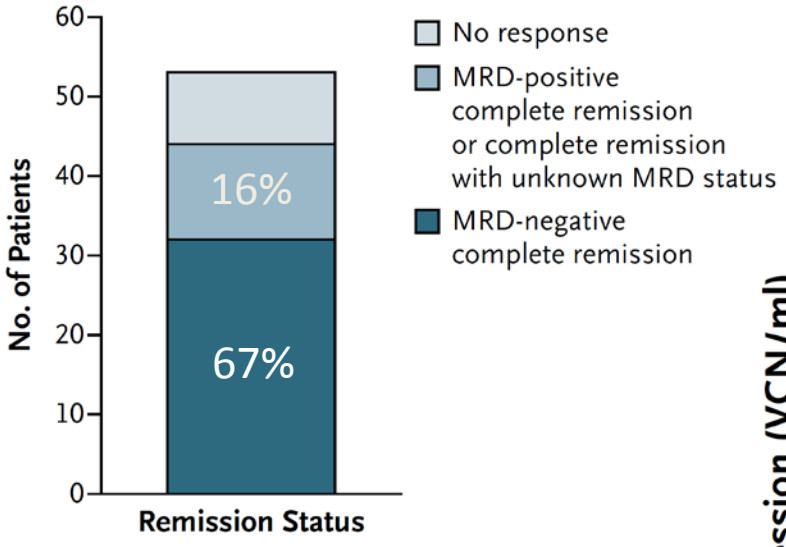
Zhang T, et al. Oncotarget. 2015. 33961.

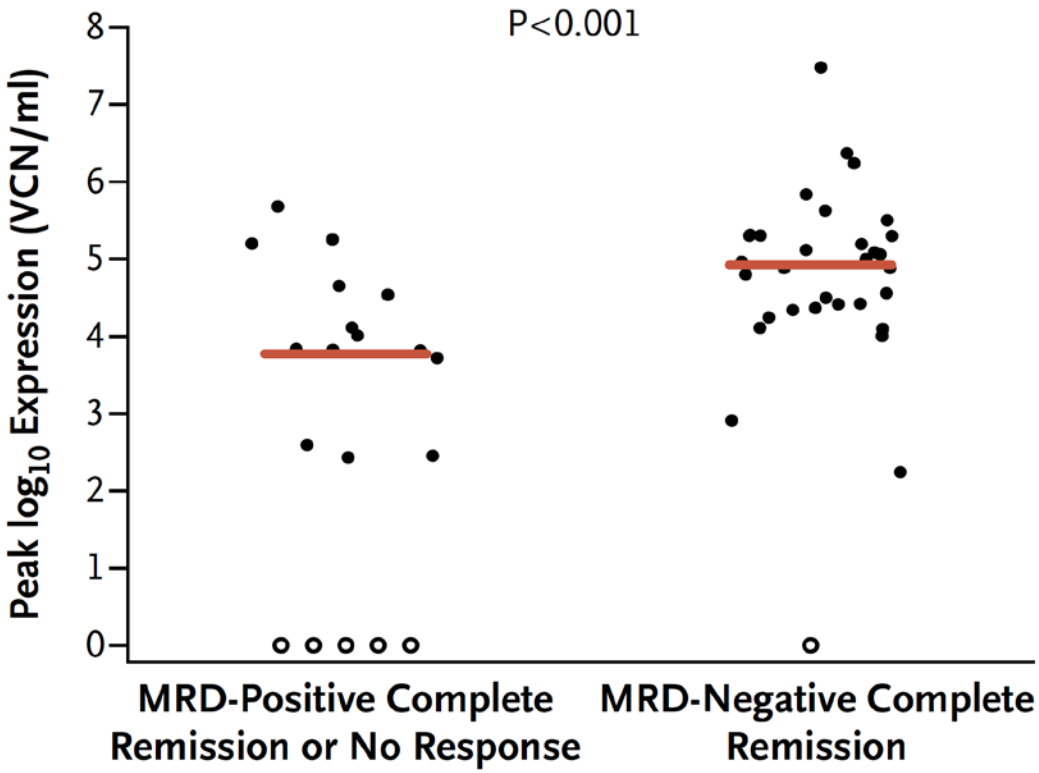
Long-term follow-up of phase 1 trial

- 53 relapsed B-cell ALL, median age 44 yrs
- 23% primary refractory disease; 30% Ph+
- 35% previous allo-HCT; 25% blinatumomab
- Median BM blast – 63%
- Auto 19-28z CAR T cells: 3 x 10^6 cell/kg
- Median F/U time = 29 (1-65) mo

Park JH, et al. NEJM 2018; 378:449.

Response to 19-28z T cells





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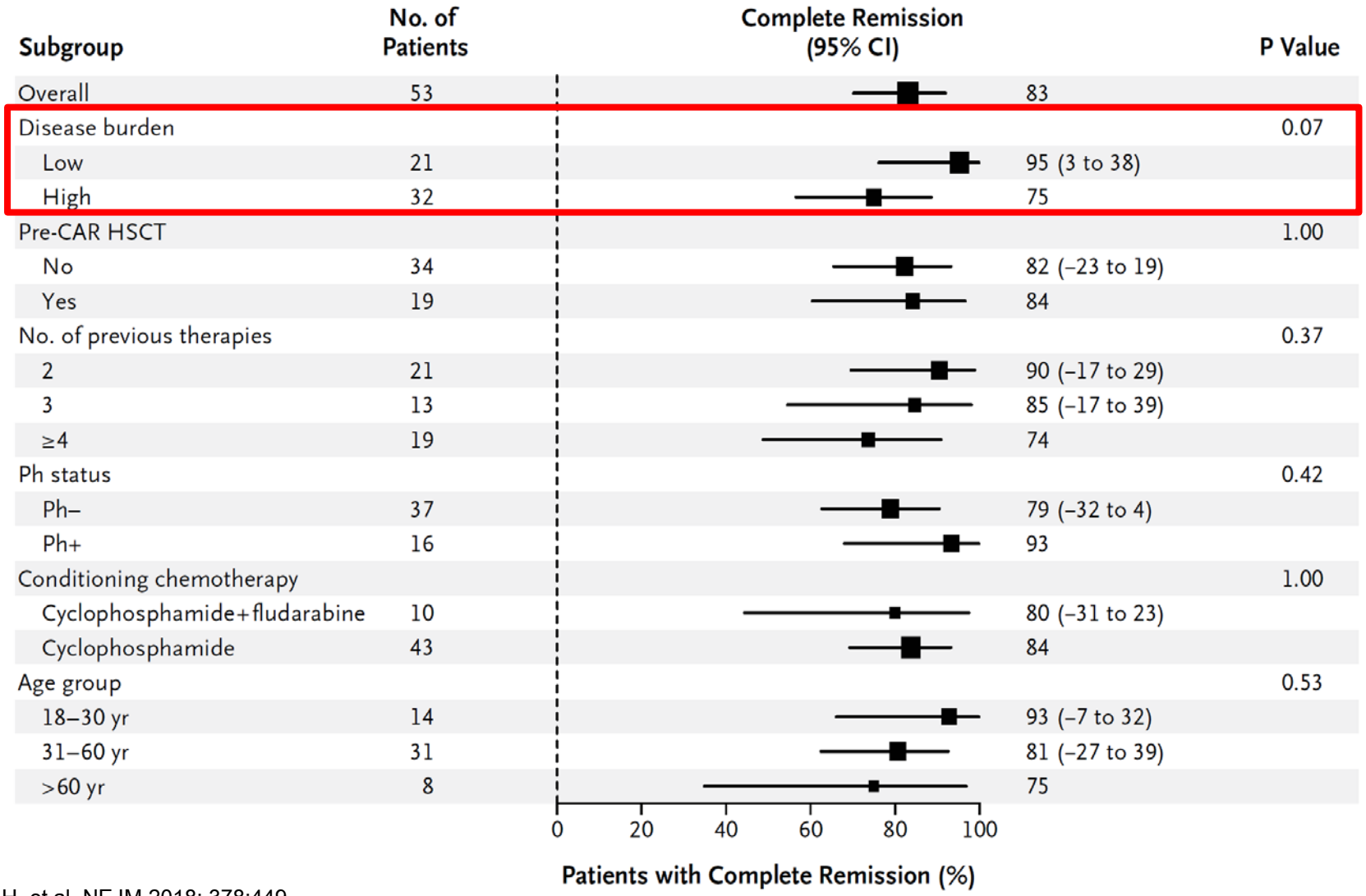
Low disease burden associated with CR



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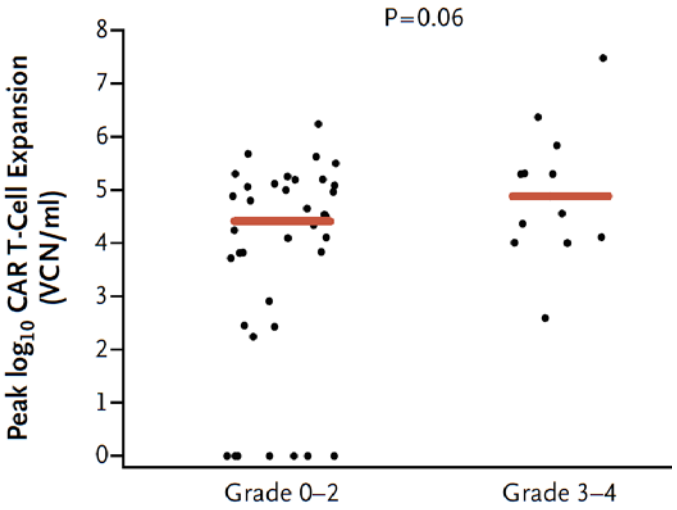
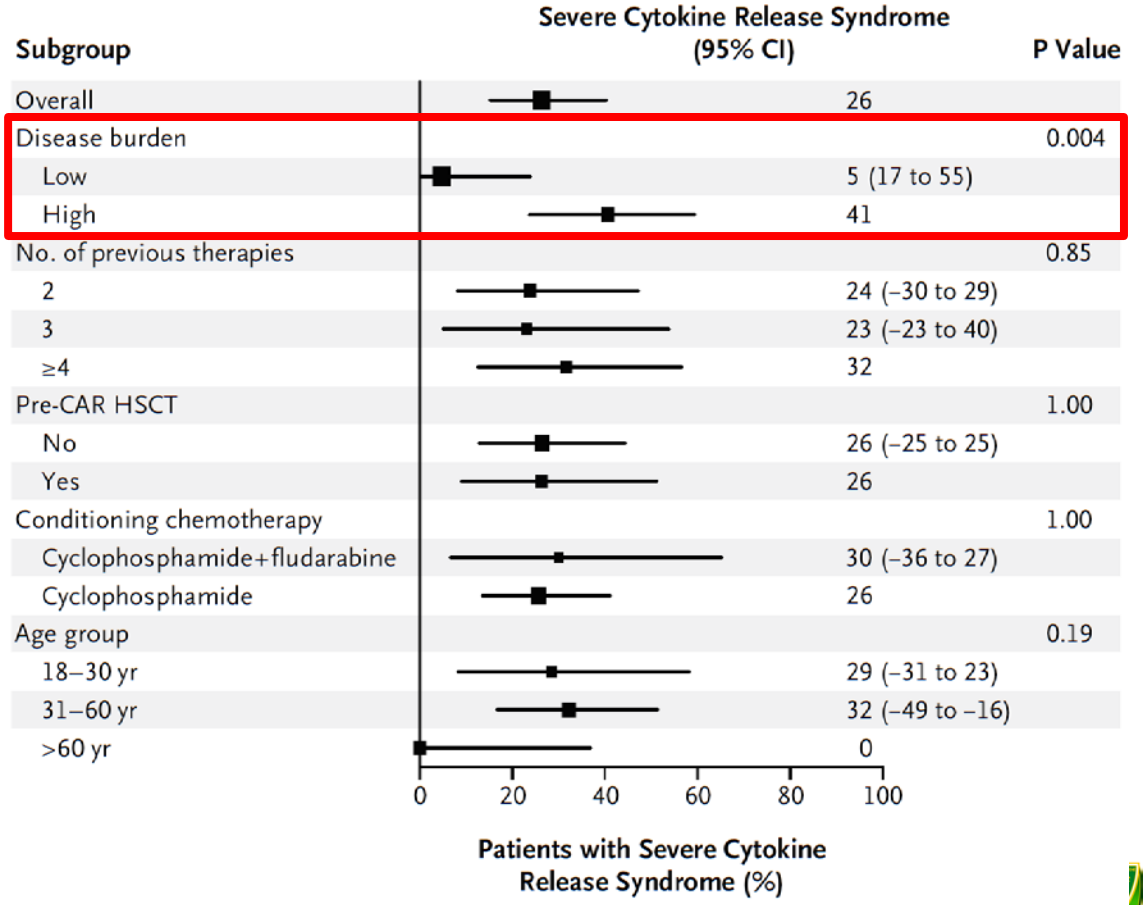
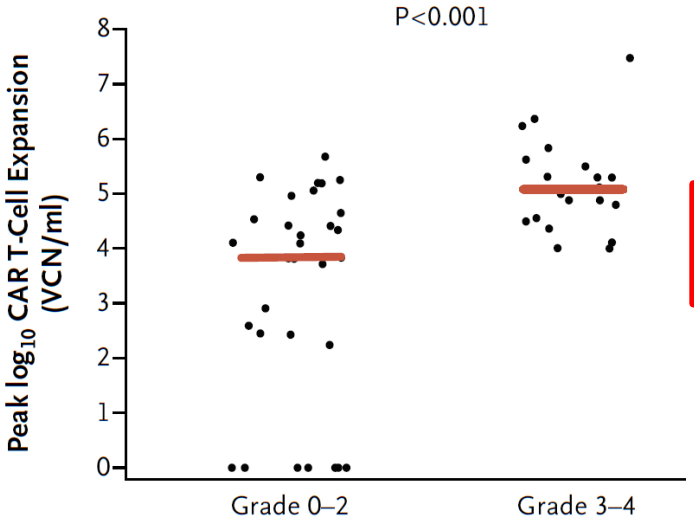
Cytokine release syndrome - 85%

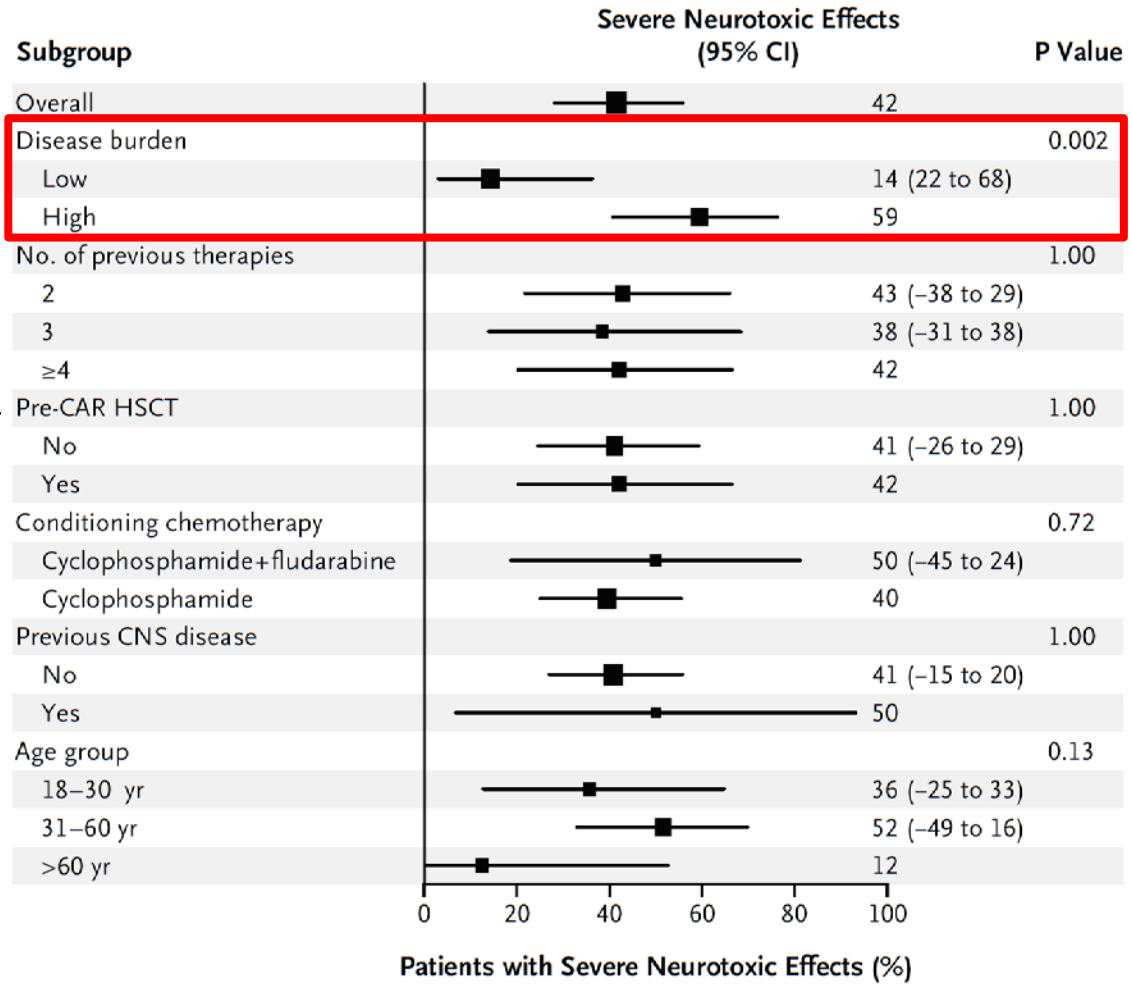




Park JH, et al. NEJM 2018; 378:449.

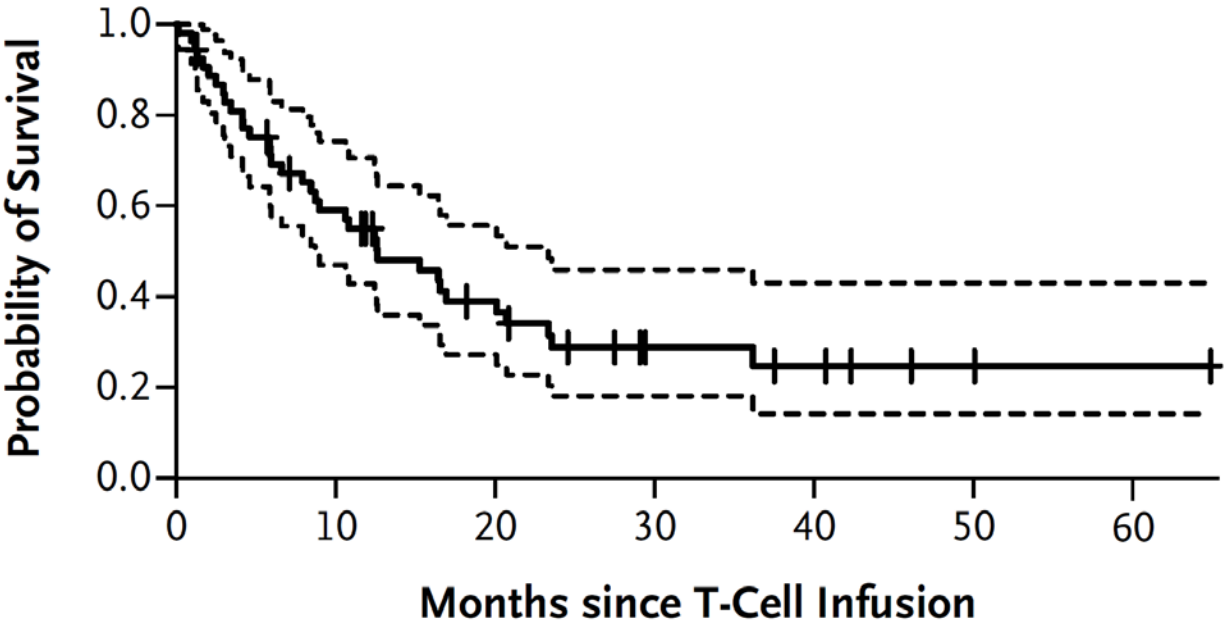
Neurotoxicity – 44%

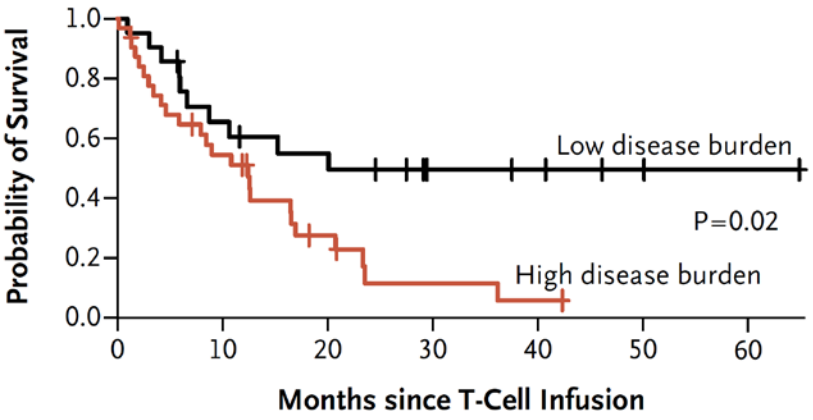


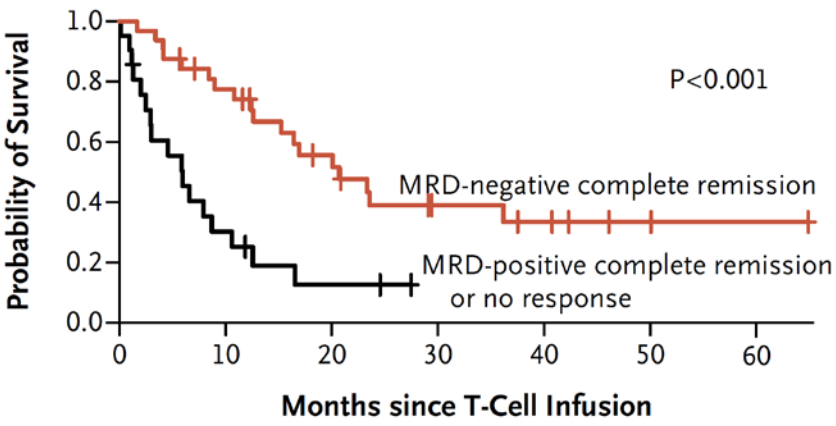


Park JH, et al. NEJM 2018; 378:449.

Median OS = 12.9mo







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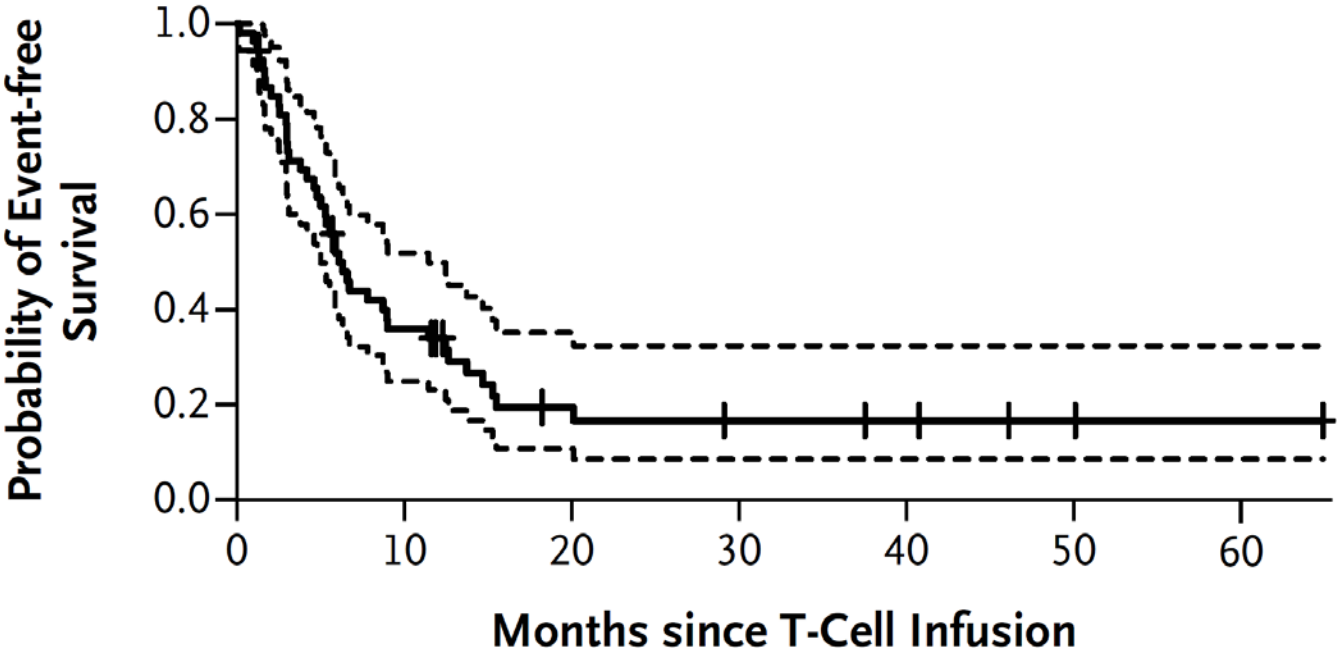
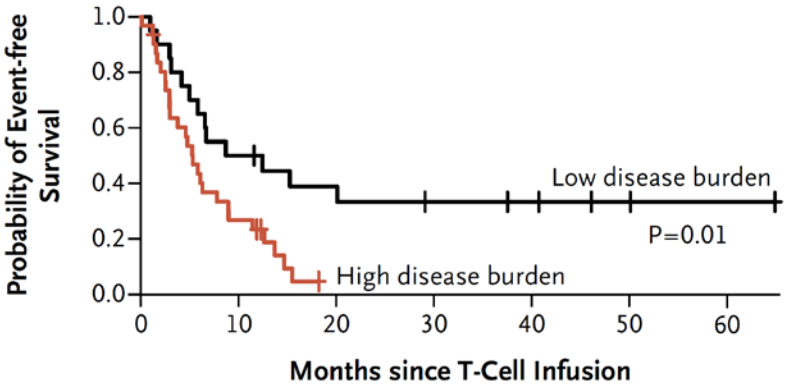
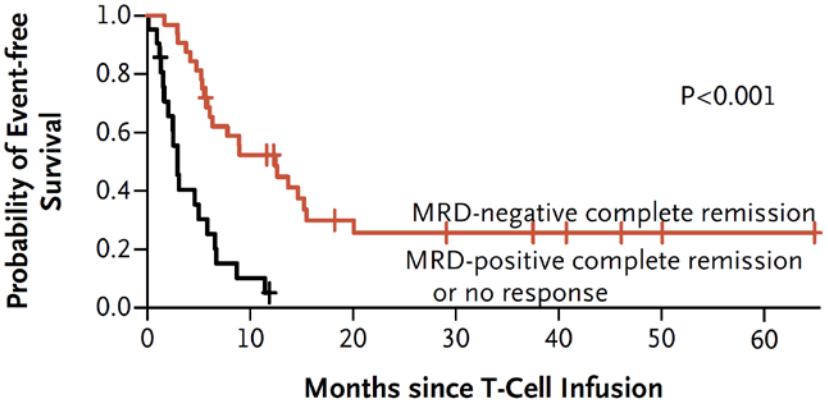
Median EFS = 6.1mo







Park JH, et al. NEJM 2018; 378:449.

Conclusion

- Patients with a low disease burden
  - Significantly longer median overall survival time
  - Markedly lower incidence of CRS & neurotoxic events

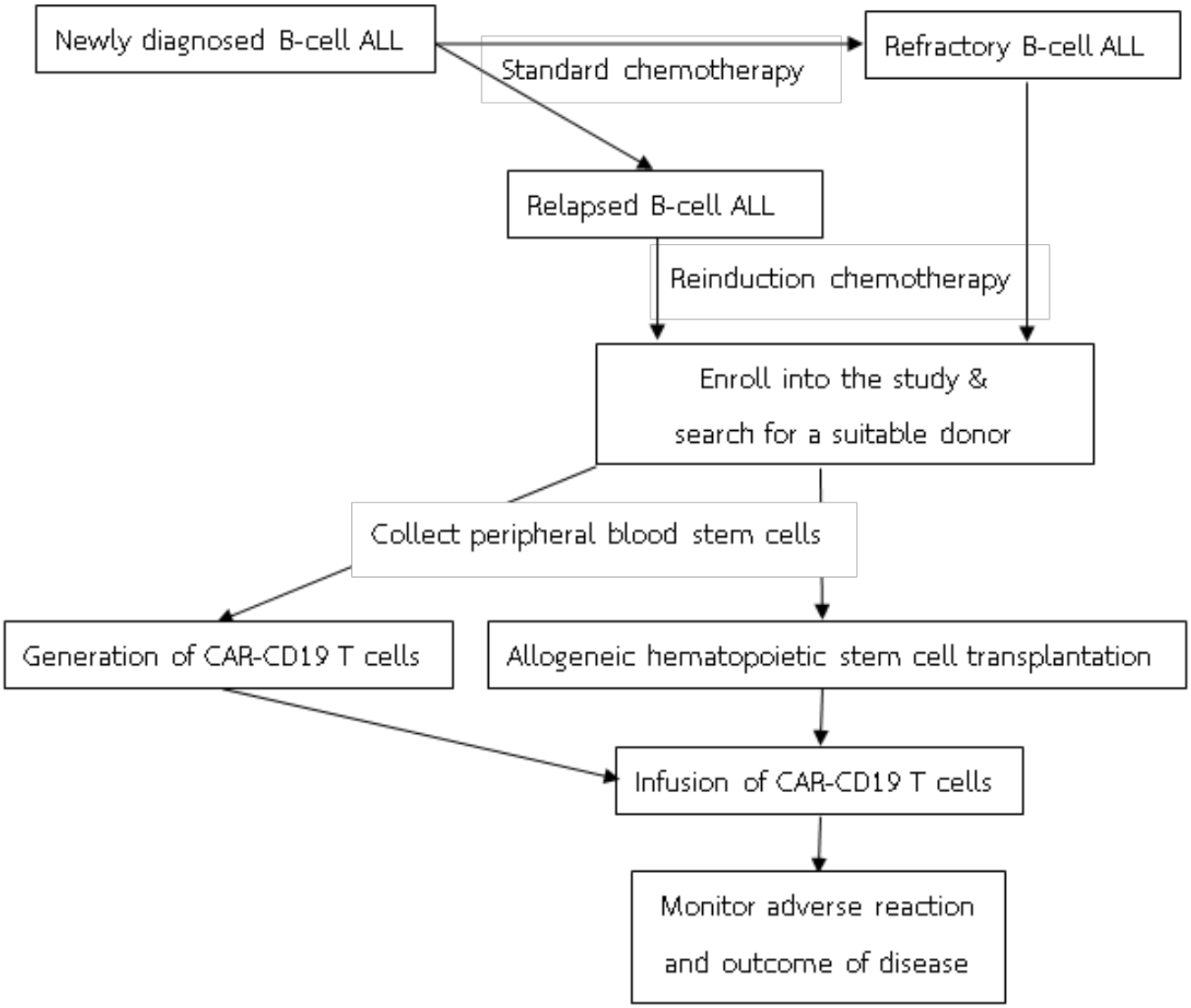
CAR T cells for clinical application

- Be produced and expanded according to GMP guidelines and in larger scale
- GMP-graded materials and reagents will be used during all processes
- All cell preparation will also be conducted in GMP facilities
- Be tested for viability, expression, efficacy and sterility prior to administration

Phase1/2 clinical trial

- Relapsed/refractory B cell leukemia patients
- Age 1-18 y/o
- No co-existing diseases
- Available donors for stem cell transplantation
- Priming with 1.5-3g/m^2 of cyclophosphamide
- Start with $5x10^5$ cell/kg in the first 3 patients

Protocol flow chart



Maximum tolerated dose

	total T cells (cells/kg)
Tier 1	5×10^5
Tier 2	1×10^6
Tier 3	2×10^6
Tier 4	4×10^6

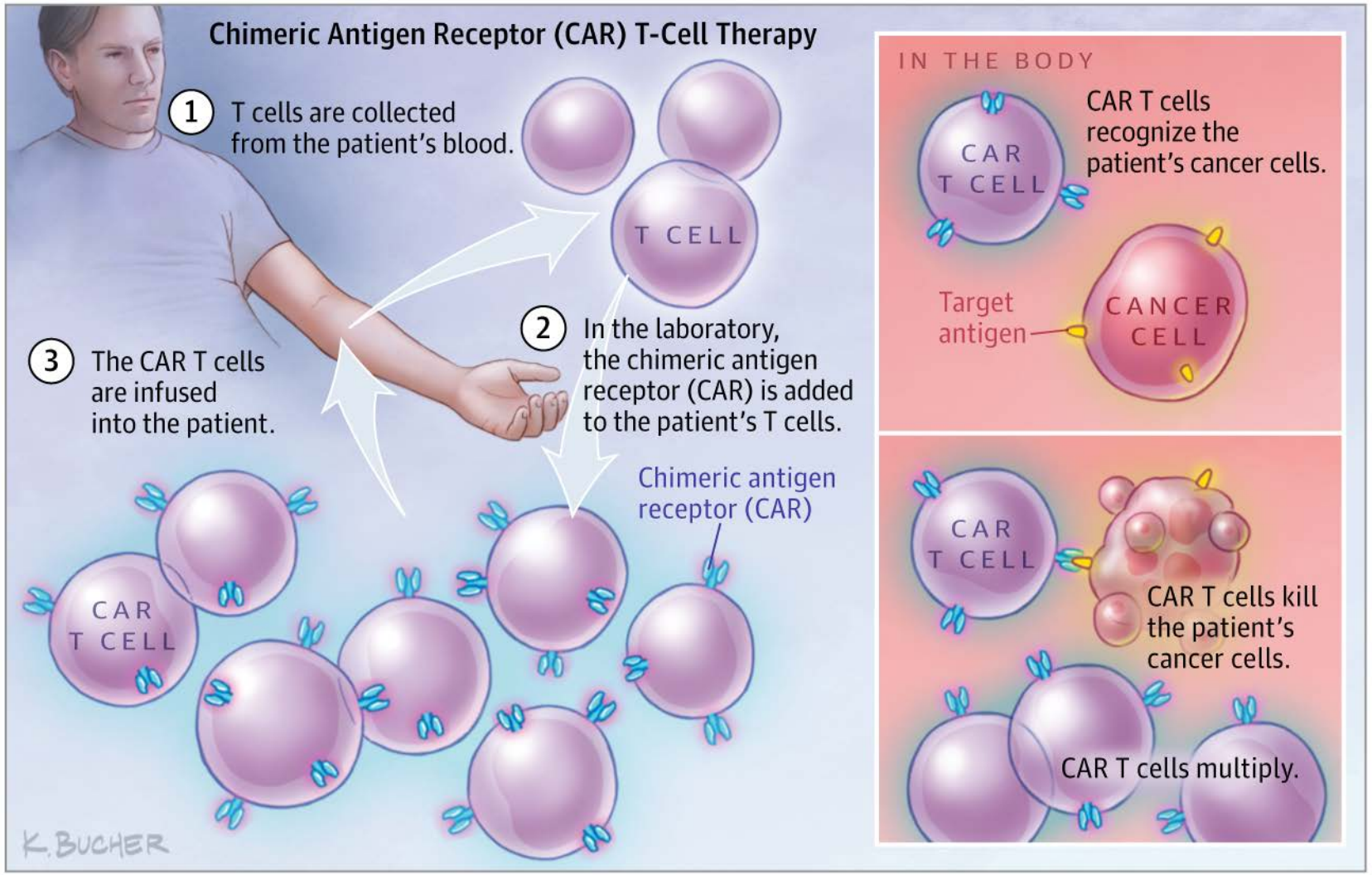
Enrolled subjects	Pts with complications	Plan
Upto 3 pts	0	Increase CAR-CD19 T cell dosage to the next tier
upto 3 pts	1	Enrolled 3 more pts receive CAR-CD19 T cell dosage at the same tier
4 – 6 pts	1	Increase CAR-CD19 T cell dosage to the next tier
4 – 6 pts	2	Assume the CAR-CD19 T cell dose is jmaximum tolerated dose, the next enrolled 3 pts receive the previous CAR-CD19 T cell dose to assure that is no complications

CAR-CD19 T cells release criteria

N=2

Test	Specimen	Specification	RESULT
Viability by trypan blue	Cell product	>70% viable	**97.30%** (Total live cell = 252-295 x 10^6)
Phenotyping	Cell product	>10% CAR-CD19 T cells	**16-21%**
Potency	Cells on day 7 of culture	> 20% lysis at 20:1 of effector to target ratio	**32.92-38.97%**
Mycoplasma test	Final product	Negative	**Negative**
Presence of bacteria by light microscope	Final product	Negative	**Negative**
Sterility Bactec	Final product	Negative	**Negative**

CAR-T against CD19+ leukemic cells



Bridget M, et al. JAMA. 2017;318:2167.


อุตตมัง อุปมา การ
มหาวิทยาลัยมหิดล
๒๕๐๘
คณะแพทยศาสตร์โรงพยาบาลรามาธิบดี
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Question?