Transfusion Medicine ABC's of CAR-T's:

Chimeric Antigen Receptor T Cell Therapy from the Apheresis, Blood Bank and Cellular Therapy Lab Perspective

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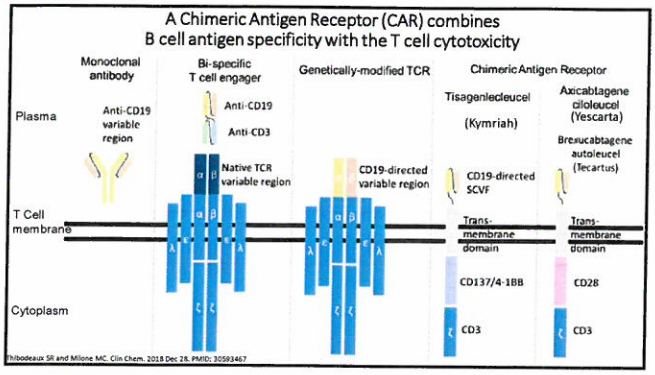
December 09, 2020

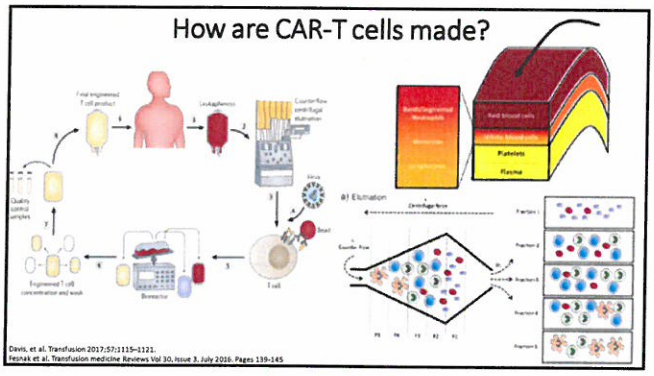
Financial disclosures

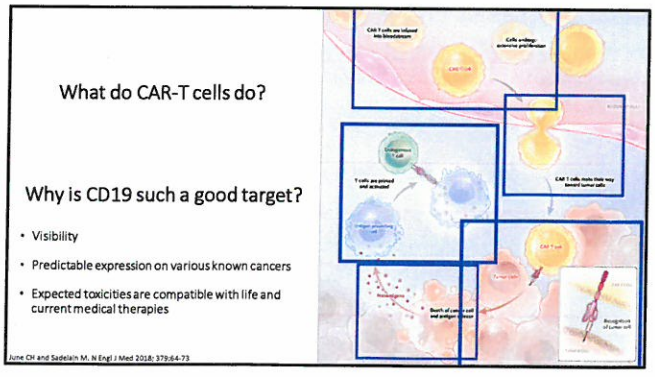
- None
- Mentions of therapies/trade names are for educational purposes only

Learning Objectives

1. **Describe CAR-T cells:**
   - structure
   - function
   - indications
2. **Recognize the role of transfusion medicine services in the care of patients receiving CAR-T cell therapy, including:**
   - Apheresis
   - Blood bank
   - Cellular therapy laboratory


A Chimeric Antigen Receptor (CAR) combines
B cell antigen specificity with the T cell cytotoxicity
Monoclonal antibody
Bi-specific T cell engager
Genetically-modified TCR
Chimeric Antigen Receptor
Tisagenlecleucel (Kymriah)
Axicabtagene ciloleucel (Yescarta)
Brexucabtagene autoleucel (Tecartus)
Plasma
Anti-CD19 variable region
Anti-CD19
Anti-CD3
Native TCR variable region
CD19-directed variable region
CD19-directed SCVF
T Cell membrane
Trans-membrane domain
CD137/4-1BB
CD28
Cytoplasm
CD3
CD3
Thibodeaux SR and Milone MC. Clin Chem. 2018 Dec 28. PMID: 30593467


How are CAR-T cells made?
Platelets
Plasma
Davis, et al. Transfusion 2017;57:1115–1121.
Fesnak et al. Transfusion medicine Reviews Vol 30, Issue 3, July 2016. Pages 139-145


What do CAR-T cells do?
Why is CD19 such a good target?
• Visibility
• Predictable expression on various known cancers
• Expected toxicities are compatible with life and current medical therapies
June CH and Sadelain M. N Engl J Med 2018; 379:64-73

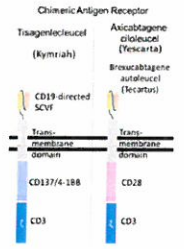
Do CAR-T cells work? Yes (with caveats)

Table 1. Responses to CAR T-Cell Therapy.*

Disease	Response Rate	Comments	Reference
	percent		
Leukemia			
B-cell acute lymphoblastic leukemia (in adults)	83–93	High initial remission rates; unresolved issue is whether CAR T-cell therapy is definitive therapy or should be followed by allogeneic hematopoietic stem-cell therapy	Park et al.,[21] Davila et al.,[36] Turtle et al.[37]
B-cell acute lymphoblastic leukemia (in children)	68–90	Approximately 25% of patients reported to have a relapse with CD19-negative or CD19-low leukemia; CD22 CAR T cells may improve survival among some patients with CD19 relapses	Maude et al.,[38] Maude et al.,[39] Fry et al.,[40] Lee et al.[41]
Chronic lymphocytic leukemia	57–71	Relapse is rare in patients who have a complete response; ibrutinib appears to increase response rates	Porter et al.,[42] Turtle et al.[43]
Lymphoma			
Diffuse large B-cell lymphoma	64–86	Approximately 40–50% of patients reported to have a durable complete response	Turtle et al.,[44] Kochenderfer et al.,[45] Schuster et al.,[46] Neelapu et al.[47]
Follicular lymphoma	71	At a median follow-up of 28.6 mo, the response was maintained in 89% of patients who had a response	Schuster et al.[46]
Transformed follicular lymphoma	70–83	A total of 3 of 3 patients with transformed follicular lymphoma had a complete response	Turtle et al.,[44] Schuster et al.,[46] Neelapu et al.[47]

June CH and Sadelain M. N Engl J Med 2018; 379:64-73

What are CAR-T cells used for?



FDA approval:

August 30, 2017

KYMRIAH (tisagenlecleucel)

Proper Name: tisagenlecleucel
Tradename: KYMRIAH
Manufacturer: Novartis Pharmaceuticals Corporation
Indication:

- Patients up to 25 years of age with B cell precursor acute lymphoblastic leukemia (ALL) that is refractory or in second or later relapse
- Adult patients with relapsed or refractory (r/r) large B cell lymphoma after two or more lines of systemic therapy including diffuse large B cell lymphoma (DLBCL) not otherwise specified, high grade B cell lymphoma and DLBCL arising from follicular lymphoma

October 18, 2017

YESCARTA (axicabtagene ciloleucel)

Proper Name: axicabtagene ciloleucel
Trade Name: YESCARTA
Manufacturer: Kite Pharma, Incorporated
Indications:

- Treatment of adult patients with relapsed or refractory large B cell lymphoma after two or more lines of systemic therapy.

July 24, 2020

TECARTUS (brexucabtagene autoleucel)

Proper Name: brexucabtagene autoleucel
Tradename: TECARTUS
Manufacturer: Kite Pharma, Inc.
Indications:

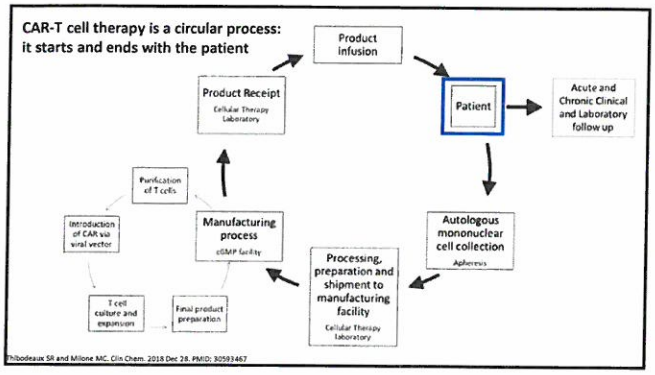
- For the treatment of adult patients with relapsed/ refractory mantle cell lymphoma (r/r MCL)

https://www.hcp.novartis.com/globalassets/migration-root/hcp/eg-plus-assets/kymriah/kymriah_ref_physician_guide_digital.pdf

Learning Objectives

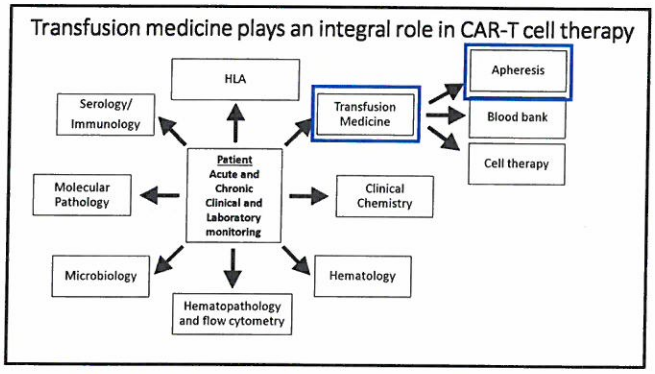
1. **Describe CAR-T cells:**
   - structure — CAR-T cells are genetically modified T cells...
   - function — with B cell antigen specificity and cytotoxic T cell function...
   - indications — in clinical use for CD19+ cancers, and under clinical development for many more potential uses
2. **Recognize the role of transfusion medicine services in the care of patients receiving CAR-T cell therapy, including:**
   - Apheresis
   - Blood bank
   - Cellular therapy laboratory

CAR-T cell therapy is a circular process: it starts and ends with the patient



Thibodeaux SR and Milone MC. Clin Chem. 2018 Dec 28. PMID: 30593467

Transfusion medicine plays an integral role in CAR-T cell therapy



Considerations for CAR-T cell production from an apheresis perspective

Timing	General considerations	Specific considerations
Before apheresis	Patient general health	Patient tolerance of apheresis collection
		Ability to tolerate turnaround time for CAR T cell manufacturing
	Patient diagnosis	Clinical indication for CAR T cells
	Predetermined requirements	FDA approved therapy or clinical study criteria
	Patient access	Peripheral or central venous access
	Laboratory parameters	Precollection CD34+ count may predict if collection will be adequate volume to process
		CBC to determine need for transfusion device priming
		Active peripheral disease may compromise collection
	Prior therapies	Time of last chemotherapy/immunotherapy should be considered for scheduling collection
	Scheduling	Patient geography
		Time slot availability for CAR T cell manufacturers and apheresis collections
During apheresis	Total blood volume	Processing volume determination
	Extracorporeal volume	Determination of need for priming apheresis machine
	Collection goals	Affects processing volume and time spent on apheresis machine
	Anticoagulant used	ACD-A or heparin
	Anticipated hypocalcemia	Calcium supplementation
	Collection efficiency	Specific to institution, apheresis machine, software, operator
After apheresis	Product contents	Determination of need for T-cell selection or lymphocyte enrichment by elutriation
	Cellular composition	Potential to affect manufacturing success
	CAR genetic material	Potential to affect transduction efficiency
	Manufacturing techniques	Media used, system used (ie, automation)
	Manufacturing times	Potential to affect individual turnaround time and overall manufacturing throughput

Chandler et al. Adv Cell Gene Ther. 2019;2:e75. https://doi.org/10.1002/acg2.75

Apheresis collection practices for manufacture of CAR-T cells differ in published literature

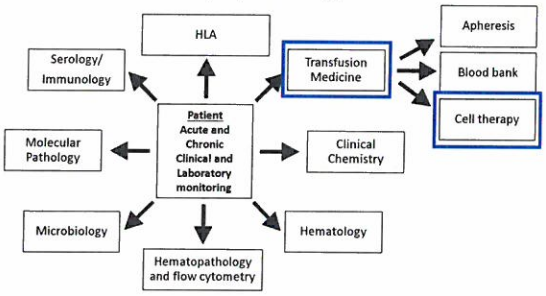
	Ceppi, et al	Allen, et al	Maude, et al	Hutt, et al	Jarisch, et al
ALC threshold	≥100/μL		>500/μL		
CD3+ threshold			>150/μL if ALC <500/μL	>250/μL	
Volume processed	Target 600 mL product	Based on pre-apheresis CD3+ count	Fixed	Target 400×10^6 cells	Based on pre-apheresis CD3+ count

Yescarta BLA: Leukapheresis: 12-15 liter apheresis with target collection of ~5-10 x 10^9 mononuclear cells

Goal of clinical studies → commercialization
could result in different requirements for different therapies in a more permanent way

Maude SL, et al. N Engl J Med 2014;371:1507-17. Ceppi F, et al. Transfusion. 2018 Jun;58(6):1414-1420. Allen ES, et al. Transfusion 2017;57:1153-61. Hutt, et al. 2019 Transfusion and Apheresis Science Volume 58, Issue 4, August 2020, 102769. Jarisch, et al. J Clin Apher. 2020;35:398–405. https://www.fda.gov/media/109189/download

Transfusion medicine plays an integral role in CAR-T cell therapy



Logistics play an important role in successful CAR-T cell manufacturing process

Chen, et al. TRANSFUSION 2019;59:2506–2518

The product label: before and after manufacturing

Collection Label

Chen, et al. TRANSFUSION 2019;59:2506–2518

Logistics play an important role in successful CAR-T cell manufacturing process

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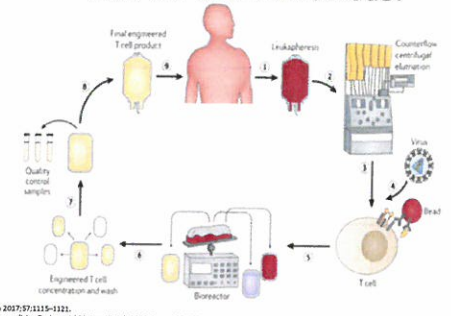
How are CAR-T cells made?



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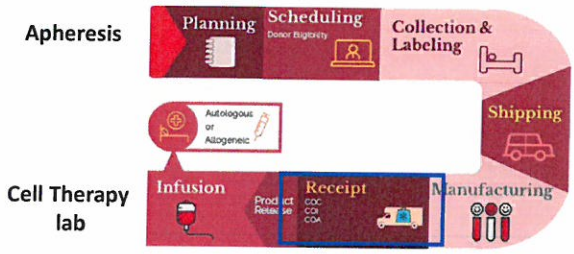


Chen, et al. TRANSFUSION 2019;59:2506–2518

The product label: before and after manufacturing

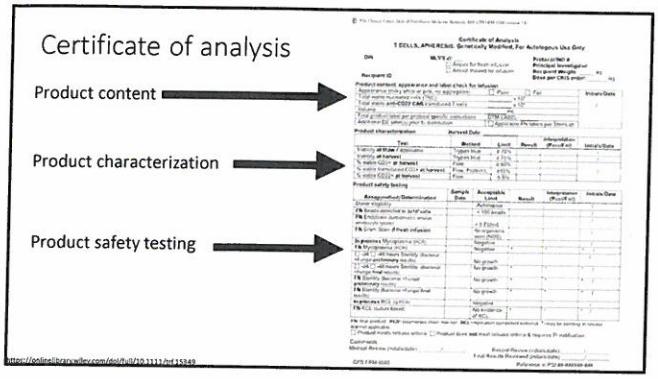
Chen, et al. TRANSFUSION 2019;59:2506–2518

Certificate of analysis

Product content

Product characterization

Product safety testing



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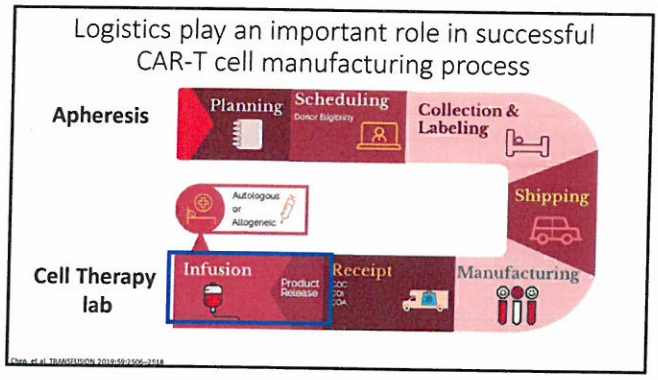
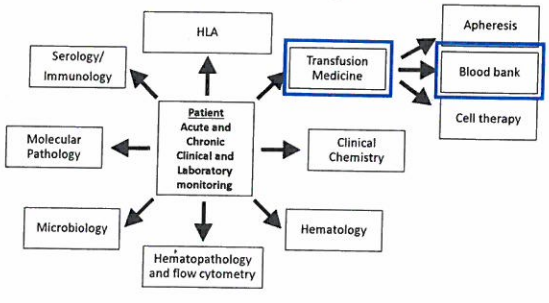


Transfusion medicine plays an integral role in CAR-T cell therapy



Potential blood bank considerations in patients receiving CAR-T cells

Indications for transfusion

- Largely remain the same as other patients who don't receive CAR-T cells
- These patients might have undergone previous therapies (HPC transplants)

Effects on blood bank could be dependent on individual therapies

- CD19 (targets B cells)
- BCMA (aka B cell maturation antigen - targets plasma cells)
- Other?

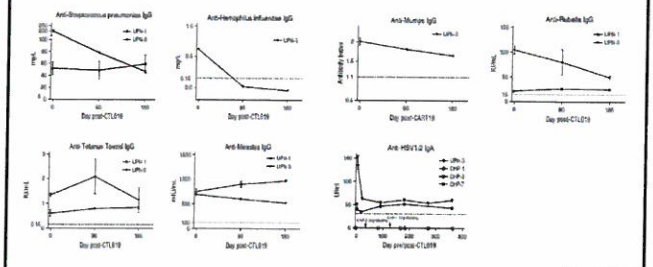
CAR-T cell therapy can be associated with adverse events

Table 2. Reported Toxic Effects of CAR T Cells.

CAR Specificity and Adverse Effect	Reference
CD19 CAR	
B-cell aplasia and hypogammaglobulinemia	Kochenderfer et al.,[32] Kalos et al.[33]
Cytokine release syndrome	Davila et al.,[34] Lee et al.,[34] Teachey et al.[35]
Dermatitis	Rubin et al.[36]
Hematophagocytic lymphohistiocytosis and macrophage activation syndrome	Grupp et al.,[32] Porter et al.,[41] Teachey et al.[35]
Neurologic effects such as ataxia and aphasia	Brudno and Kochenderfer[37]
Cerebral edema	Gust et al.[38]

June CH and Sadelain M. N Engl J Med 2018; 379:64-73

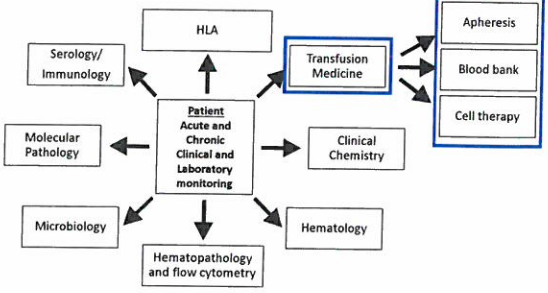
Antibody titers can persist after CD19 CAR-T cell therapy



Bhoj et al. Blood. 2016;128(3):360-370

Serology/ Immunology

Transfusion medicine plays an integral role in CAR-T cell therapy



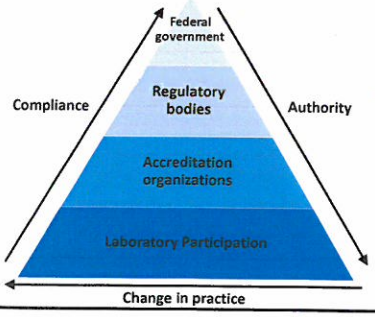
The regulatory landscape for cell therapies (and CAR-T cells) is multi-faceted

Group	Cell therapy focus	Standards	Accreditation duration
AABB*	Clinical: collection, cord blood banking, processing, storage and distribution	Standards for cellular therapy product donors	2 years
CAP*	Product testing (eg, microbiology, flow cytometry, hematology), collection, processing, storage, and distribution of HPC and tissue	Standards for Laboratory Accreditation; Laboratory Accreditation Manual; All Common, Laboratory General, and Laboratory-Specific Checklists	2 years
FACT*	Clinical program, collection, cord blood banking, processing, storage, and distribution	FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing, and Administration	3 years
		NetCord-FACT International Standards for Cord Blood Collection, Processing, Testing, Banking, Selection, and Release	
JACIE*	Clinical program, collection, cord processing, storage, and distribution	FACT-JACIE International Standards for Cellular Therapy Product Collection, Processing and Administration	4 years
NMDP**	Unrelated products including donor screening, collection, distribution, cord blood banking, processing	National Marrow Donor Program Standards	3 years

*Voluntary, ** required for access to unrelated donors, FACT/AABB required for cord blood, CAP required for microbiology

Reynolds-Smith L, Linenberger M. Collection of Cellular Therapy Products by Apheresis. In: Areman E, Loper K, editors. Cellular Therapy: Principles, Methods and Regulations. Bethesda, MD: AABB; 2016. p306-317.

Ultimately regulation starts and ends with the FDA (in the USA)



- **Federal government**
  regulation of labs and lab testing
- **Regulatory bodies (CMS, FDA)**
  to carry out laws
- **Accreditation organizations**
  for clinical laboratories
- **Laboratory Participation**
  for ongoing compliance

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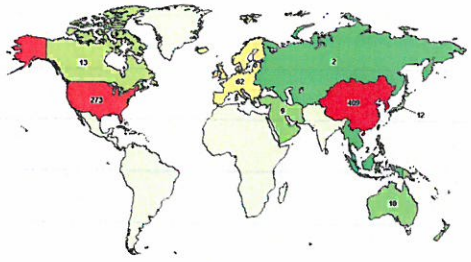
2. **Recognize the role of transfusion medicine services in the care of patients receiving CAR-T cell therapy, including:**
   -
   -
   -

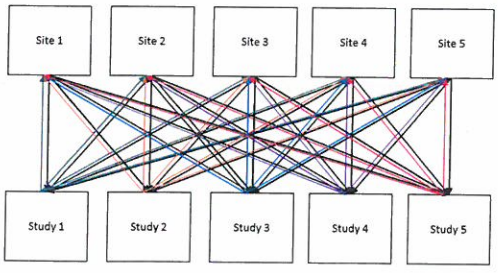
Transfusion medicine is critical for care of patients undergoing therapy with CAR-T cells

Clinical studies for CAR-T cells is global and growing!

762 studies as of 12/8/2020



The complexity of multiple studies with clinical study requirements can present difficulties



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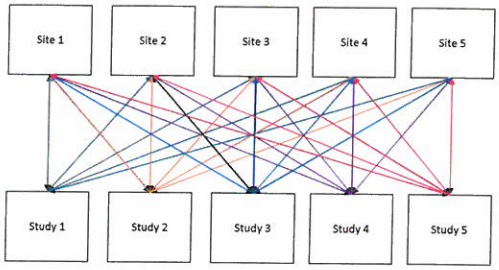

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   evolving

2. **Recognize the role of transfusion medicine services in the care of patients receiving CAR-T cell therapy, including:**
   - Apheresis — is critical for collection of material for CAR-T cells
   - Blood bank — is critical for transfusion support of patients receiving CAR-T cells
   - Cellular therapy laboratory — is critical for processing material for CAR-T cells

Transfusion medicine is critical for care of patients undergoing therapy with CAR-T cells

Thank you!

HAABB

Heart of America Association of Blood Banks